INAJOG

EDITORIAL

Vitamin D Supplementation to Reduce the Risk of Preeclampsia: Is It True?

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Hypertensive disorders in pregnancy including preexisting chronic hypertension, pregnancy-induced hypertension, and preeclampsia is one of the most complication in pregnancy and it contributes to the maternal mortality worldwide. Actually, there is preeclampsia in every tenth pregnancy and the incidence of preeclampsia is approximately 2-7%. Several risk factors known in preeclampsia are nulliparity, multifetal gestation, previous preeclampsia, obesity, and preexisting medical conditions.[1]

The pathophysiology of this condition is still unknown, but studies suggest an association to vitamin D status, measure as 25-hydroxyvitamin D (25(OH)D). Linnea B, et al stated that the 25(OH)D concentration at least 30 nmol/l was associated with lower odds ratio for preeclampsia.[1] Another review by David B, et al. pointed out that lower maternal calcitriol levels were associated with preeclamptic women at the diagnosis of disease in the third semester of pregnancy. It hypothesized that low IGF-1 and PTHrP explained the decreased calcitriol levels in observed preeclamptic women. Calcidiol as the marker of vitamin D have been found lower in early-onset severe preeclamptic women.[2] Other studies stated that vitamin D status is influenced by sunlight exposure; nevertheless, preeclamptic women had lower serum calcidiol levels in summer and calcitriol concentrations in winter.[3] Therefore, they concluded that these two vitamin D metabolites have not been found related between healthy and preeclamptic pregnancies.

Regarding the controversies above, we suggest to take the vitamin D supplementation during pregnancy to reach a circulating-calcidiol level of 30 ng/ml and not exceeding 150 ng/ml to avoid the risk of toxicity. Apart from that, the level of calcidiol should be more than 40 ng/ml to optimize the calcitriol production during pregnancy.[2] In conclusion, we should give not only calcitriol, but also calcium to reduce the risk of preeclampsia development.

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Research Article

Characteristics of Maternal Mortality Cases in a Tertiary Hospital

Kajian Karakteristik Kematian Ibu di Rumah Sakit Tersier

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Abstract

Objective: To identify the characteristics of maternal mortality cases in a tertiary hospital in Jakarta, including socio-demographic characteristics, previous medical and obstetric history, and patient's clinical condition on arrival at the hospital.

Method: This was a survey to identify the descriptive data of maternal mortality cases through medical records during study period. Manual review of 51 medical records was conducted for 2 years from January 2013 to December 2014 in Department of Obstetrics and Gynecology, Dr. Cipto Mangunkusumo Hospital (RSCM).

Result: Of 51 cases of maternal deaths, 46 subjects (90.19%) had nine years of minimum education background. There were two subjects less than 21 years old and another was 42 years old. None of these subjects were using intrauterine device (IUD) or implant as the contraceptive methods where 66.6% subjects with underlying disease never used contraception. Ninety-two percent of subjects did antenatal care (ANC) regularly and 80.4% (41 subjects) of them was done in midwives. There were 14 subjects (29.78%) who had ANC in the first trimester of pregnancy. Severe preeclampsia is the most prevalent complication in pregnancy (26 subjects, 65%), which all (100%) patients arrived at RSCM with HELLP Syndrome. Therefore, preeclampsia was the leading cause of death in RSCM.

Conclusion: The characteristics of maternal death in RSCM are prevalent in the group of 25-34 years old with the high school as the educational background. Most of them are multiparity and do not use the long-term contraceptive methods. Preeclampsia is the major cause of maternal death in RSCM.

[Indones J Obstet Gynecol 2016; 4-3: 119-122]

Keywords: maternal mortality, risk factors, tertiary hospital

Abstrak

***Tujuan**: Mengidentifikasi karakteristik kematian ibu di RS tersier di Jakarta yang meliputi karakteristik sosio-demografi, riwayat obstetri dan medis, serta kondisi klinis pasien saat tiba di rumah sakit.*

***Metode**: Penelitian ini merupakan survei untuk mengetahui data deskriptif kematian maternal menggunakan rekam medis selama rentang waktu penelitian. Dilakukan telaah rekam medis 51 kasus kematian ibu yang terjadi selama 2 tahun dari Januari 2013 hingga Desember 2014 di bagian Obstetri dan Ginekologi, RSCM.*

***Hasil**: Dari 51 kasus kematian maternal, 46 subjek (90,19%) berpendidikan minimal sembilan tahun. Terdapat dua subjek yang berusia kurang dari 21 tahun dan satu subjek yang berusia 42 tahun. Tidak ada satupun subjek yang pernah menggunakan metode kontrasepsi IUD atau implan dengan 66,6% subjek yang memiliki penyakit penyerta tidak pernah menggunakan kontrasepsi. Empat puluh tujuh subjek melakukan ANC yang umumnya dilakukan di bidan (41 subjek, 80,4%) dan hanya 14 subjek (29,78%) yang pernah menjalani ANC pada trimester pertama kehamilan. Preeklamsia berat merupakan komplikasi kehamilan yang paling banyak dijumpai (26 subjek, 65%), di mana seluruhnya (100%) tiba di RSCM dengan sindrom HELLP. Komplikasi preeklamsia adalah penyebab kematian utama.*

***Kesimpulan**: Karakteristik kematian maternal di RSCM banyak terjadi pada usia 25-34 tahun dengan latar belakang pendidikan lulus SMA. Kebanyakan dari mereka multiparitas dan tidak menggunakan metode kontrasepsi jangka panjang. Preeklamsia merupakan penyebab kematian maternal di RSCM.*

[Maj Obstet Ginekol Indones 2016; 4-3: 119-122]

***Kata kunci**: faktor risiko, kematian ibu, rumah sakit tersier*

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INTRODUCTION

Maternal deaths according to the Tenth Revision of the International Classification of Diseases (ICD-10) is the death of a woman during pregnancy until 42 days after delivery, irrespective of the length and location of the pregnancy. Apart from that, it can be caused by anything related to pregnancy or aggravated by the pregnancy; it is not due to an accident, yet.[1,2]

Reducing maternal mortality has long been a priority on global health program and it is one of the targets in the United Nations Millennium Development Goals (MDGs) number five. Based on the Household Health Survey, maternal mortality rate (MMR) in Indonesia in 2001 was 396 per 100,000 live births; meanwhile, Indonesia Demographic and Health Survey (IDHS) 2002/2003 stated that the MMR was 307 per 100,000 live births.[3] Actually, this was contrary to the MDGs target in 2015 namely 108 per 100,000 live births.[4]

In 2012, the number of maternal deaths in Jakarta were 97 patients. The highest incidence of MMR was located in East Jakarta (34), followed by

North Jakarta (23). The main causes of maternal death in Jakarta in 2012 were preeclampsia/ eclampsia (39%), bleeding (31%), infection (6%), abortion (2%), prolonged labor (1%) and others. Health status, education, accessibility to the health care, and services by health professionals during pregnancy and delivery influence the MMR.[5]

Dr. Cipto Mangunkusumo Hospital (RSCM) as a national referral center in Indonesia is the center of excellence consisting of education, research, and service for co-assistant, residents, nurses, and other health workers.[6] As the tertiary hospital in Indonesia, it is noteworthy to analyze the characteristics of maternal mortality in order to describe the referral condition to the tertiary hospital in Indonesia. Therefore, this study aims to identify the determinants affecting maternal mortality, including individual and socio-demographic, maternal history, obstetric or reproductive history, and the mother's condition upon hospital admission.

METHODS

This was a descriptive study using cross sectional design which was held in RSCM to identify the characteristics of maternal deaths. The inclusion criteria were all maternal deaths occured in RSCM from January 2013 to December 2014. The exclusion criteria were if we did not find the medical records. This study had been approved by the Research Ethics Committee of the Faculty of Medicine, Universitas Indonesia through a Certificate of Conduct number 441/UN2.F1/ETIK/2015.

RESULTS

During the study period, we found 51 maternal deaths. Of them, 58.8 % (30 subjects) were in the age group of 25-34 years old and the majority of educational background was graduated from high school (36 subjects; 70.6%). A total of 90.2% (46 subjects) finished nine-year education. There were 2 subjects aged less than 21 years old, namely 19 and 18 years old and both of them were primigravida. Meanwhile, there was one 41-year-old patient on her third pregnancy. All of these outliers experienced severe preeclampsia. Almost all subjects (50 subjects; 98.1%) were housewifes. Actually, all subjects in this study used governmental insurance (Jampersal, JKN, BPJS) as financial resource.

Twenty-three subjects from multiparity and grandemultiparity had ever used contraception, whereas 7 subjects (30.4%), 15 subjects (65.3%), 1 subject (4.3%) used pill, injection, condom; respectively. Only one subject with grandemultipara in fifth pregnancy never used any contraceptive methods.

Four subjects (28.57%) delivered assisted by forceps; while, another one else (7.14%) assisted by vacuum extraction. All patients with forceps extraction were caused by uncontrolled blood pressure on severe preeclampsia. The reason of using vacuum extraction was due to dystocia on second stage of labor. All subjects who did not undergo labor (4 subjects) and came with severe preeclampsia were accompanied by multi organ failure at less than 26 weeks of gestational age.

A total of 15 subjects (29.4%) had a history of previous illness. Consisting of two subjects (13.3%) with history of treatment for lung tuberculosis, 3 subjects (20%) with hypertension, 2 subjects (13.3%) with blood disorders (thalassemia and chronic granulocytic leukemia/CGL), 2 subjects (13.3%) with an autoimmune disease (SLE and autoimmune hemolytic anemia/AIHA), 1 subject (6.7%) with hepatitis B, 1 subject (6.7%) with asthma, and 1 subject (6.7%) with obesity. Only 1 subject (6.7%) was known to be infected with HIV. All of the subjects were multigravidas, which 5 of them used contraception, while, 10 subjects never used contraception.

The majority of subjects (41 subjects, 80.4%) had antenatal care done by midwifes. Fourteen subjects (27.5%) had antenatal visits less than 4 times and 4 subjects (7.8%) had never undergo antenatal care. Of 47 subjects who underwent ANC, only 14 subjects (29.8%) who had undergone the ANC in the first trimester.

Table 1. The Characteristics of Subjects

Characteristics	N	%
Contraception		
Yes	23	45.1
No	28	54.9
Parity		
Primi	18	35.3
Multi	32	62.8
Grandemulti	1	19

Previous illnes		
Yes	15	29.4
No	36	70.6
Delivery method		
Vaginal delivery (Spontaneous/ instrumental)	14	27.5
Caesarean section	33	64.7
Not delivered	4	7.8
Antenatal care (ANC)		
Never	4	7.8
Midwife	41	80.4
Doctor	6	11.8
ANC frequency		
Never	4	7.8
< 4 times	14	27.5
≥ 4 times	33	64.7

Fourteen subjects (27.5%) came up with co-morbidities. The co-morbidities of subjects consisted of 10 (19.6%), 12 (23.5%), 8, 2, 1, and 1 subjects having eclampsia, postpartum hemorrhage, uterine atony, placental abruption, placenta previa, and uterine perforation; consecutively. Actually, the postpartum hemorrhage occurred outside RSCM in eight subjects and inside RSCM in 4 subjects were entirely caused by uterine atony. Of the 8 subjects who were referred to RSCM for bleeding, one subject was in third grade of shock, with less than 7 for hemoglobin level. She was applied an intravenous line without urine catether.

One subject in stage II dystocia was primigravidae, referred by midwifes, was in the second stage of labor for 3 hours. Finally, she was delivered by vacuum extraction and after delivering, she got postpartum hemorrhage due to uterine atony and birth canal laceration.

Of all patients who were referred or come with severe preeclampsia or eclampsia, there were 4 subjects without applying the intravenous line because they came without reference letter from secondary health centers. All preeclamptic patients had been given MgSO4 to avoid the convulsion and nifedipine for the blood pressure since at primary health. The majority of subjects (47 subjects, 92.2%) were referral cases from other medical facilities. About 18 subjects (35.3%) were referred in anemic condition.

Table 2. The Characeristics of Patient's Clinical Condition at Arrival

Variables	N	%
Comorbid Disease		
Yes	14	27.5
No	37	72.5
Eclampsia		
Yes	10	19.6
No	41	80.4
Dystocia		
Yes	1	1.9
No	50	98.1
Ante/post partum bleeding		
Yes	12	23.5
No	39	76.5
Hemoglobin		
< 10,5 gr/dl	18	35.3
≥ 10,5 gr/dl	32	62.8
No data	1	1.9
Referral status		
Referred	47	92.2
By own will	4	7.8

There were 14 subjects (27.5%) in co-morbidity pregnancy. Meanwhile, six subjects (15%) had cardiac abnormalities during pregnancy consisting of 2 subjects with atrial septal defect, 3 subjects with valvular heart disease, and 1 subject with aortic aneurysm. The other underlying diseases were such as one subject with HIV, one subject with pulmonary tuberculosis, one subject with pulmonary tuberculosis on going therapy, one subject with systemic lupus eritematosus, and another one with autoimmune hemolytic anemia. While, there were 2 subjects in blood disorder, namely chronic granulocytic leukemia and thalassemia for each patient.

Preeclampsia as a direct result of pregnancy abnormality was found in 26 subjects, whereas 10 of them suffered from eclampsia. All the patients with preeclampsia or eclampsia was on going of HELLP Syndrome at arrival.

Around 36 subjects (70.6%) went through septic shock as the cause of death. Sepsis was the most common in the group of subjects with severe preeclampsia or eclampsia. Meanwhile, postpartum hemorrhage as the cause of death was happened in 8 subjects (15.7%) (shown in Table 3).

Table 3. Cause of Death

Cause of death	N	%
Sepsis	36	70.6
Hemorrhagic shock	8	15.7
CNS disorder	5	9.8
Cardiac abnormality	2	3.9

DISCUSSION

Acknowledging the characteristics of maternal mortality and clinical spectrum in tertiary hospital makes the inspiration for the further research to prevent the maternal mortality. Campbell, et al. through their study stated that optimal intrapartum care was the best strategy to decrease the MMR.[7,8] From these findings, it was possible to change the prevention strategy as most of deaths were due to preeclampsia/eclampsia.[9] Meanwhile, there was a tendency that the majority of deaths were occurred in women doing the regular antenatal care at midwifes without risk factors analysis recorded. Therefore, we should improve the quality of antenatal care at midwifes. This can be achieved through regular training and education about screening and suggesting the midwifes to refer the women with high risk pregnancy.

CONCLUSION

The characteristics of maternal death in RSCM are prevalent in the group of 25-34 years old with the high school as the educational background. Most of them are multiparity and do not use the long-term contraceptive methods. Preeclampsia is the major cause of maternal death in RSCM.

RECOMMENDATION

Further study on early detection and prompt treatment of preeclampsia or eclampsia in primary care facilities should be conducted because pregnancy-induced hypertension is the most common causes of maternal mortality in this study. Improving antenatal care quality in primary center by providing continuing professional education for health workers there. Apart from that, the identification of preeclampsia should remain a priority in the clinical setting considering the contribution of these disease in MMR. The antenatal care utilization depends not only on number of visits, but also the health professional should be able to detect any problem rising during pregnancy and making the right referral. Finally, the ability to space and limit the pregnancy, especially in high risk group becomes the key point to reduce the MMR.

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Research Article

Maternal and Fetal Outcome on Pregnancy in Advanced Maternal Age

Luaran Maternal dan Fetal pada Kehamilan Ibu Usia Lanjut

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Abstract

Objective: Pregnancy in advanced maternal age (AMA) was classified as high risk. The study aims to provide a better description of pregnancy outcome in AMA.

Method: The cross sectional study was to review the demography, medical and obstetrics problems, mode of delivery, maternal and fetal outcome in Tangerang General Hospital as a referral center in Banten. The data were taken from medical records of 35-year-old and above women age who delivered on the period of January to December 2014.

Result: The total number of delivery and live birth was 6,107 and 5,926 respectively, including 1,548 (25.36%) pregnancies in AMA. The prevalence of grandmultiparities was 11.4%. The average education level was mostly elementary. The prevalence of hypertension in pregnancy was 34.6%. There was increasing trend of miscarriages from 10.8% in 35-40-year-old group to 25% in above 45-year-old group. Pregnancy complications were higher consisting of 3.2% multifetal, 16.6% malpresentation, and 7.1% placenta previa. Cesarean section rate was 33.6%. Maternal near-missed cases were 56 of 1000 and the maternal mortality rate (MMR) was 932 of 100,000 live births. The prevalence of stillbirth and perinatal mortality were 5.3% and 12.9%; respectively.

Conclusion: Adverse maternal and fetal outcome were higher in AMA as it takes special attention and multidiscipline-approached care started from preconception, antenatal, preparation of delivery, and also postpartum care.

[Indones J Obstet Gynecol 2016; 4-3: 123-127]

Keywords: advanced maternal age (AMA), fetal outcome, maternal outcome, pregnancy

Abstrak

***Tujuan**: Kehamilan ibu usia lanjut memiliki risiko tinggi. Rumah Sakit Umum (RSU) Tangerang adalah pusat rujukan di provinsi Banten, sehingga diharapkan penelitian ini dapat menggambarkan luaran kehamilan pada ibu usia lanjut.*

***Metode**: Penelitian ini menggunakan studi potong lintang yang melakukan kajian pada karakteristik demografi, masalah medis, obstetri, metode persalinan, luaran fetal dan maternal di RSUD Tangerang sebagai pusat rujukan Provinsi Banten. Data diambil dari rekam medis kasus kehamilan ibu usia lanjut di RSUD Tangerang dari Januari 2014 sampai Desember 2014.*

***Hasil**: Terdapat 6.107 persalinan, 5.926 kelahiran hidup di mana 1.548 (25,36%) ibu usia lanjut. Sebanyak 11,4% grandemultipara, pendidikan rata-rata SD. Prevalensi hipertensi 34,6%. Abortus meningkat sebanding usia ibu, dari 10,8% pada usia 35-40 tahun sampai 25% pada usia ≥ 45 tahun. Komplikasi kehamilan lebih tinggi dari penelitian lain, yaitu kehamilan kembar 3,2%, malpresentasi 16,6% dan plasenta previa 7,1%. Seksio sesarea sebanyak 33,6% persalinan. Kejadian maternal near missed dan AKI yaitu 56 per 1000, dan 932 per 100.000 kelahiran hidup. Prevalensi kematian janin intrauterin dan perinatal adalah 5,3% dan 12,9%.*

***Kesimpulan**: Luaran maternal dan fetal yang tidak diinginkan sampai dengan kematian ibu dan janin lebih tinggi pada ibu usia lanjut, sehingga diperlukan perhatian dan penanganan khusus dimulai dari perencanaan kehamilan, asuhan antenatal, persiapan persalinan, dan asuhan nifas yang baik.*

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***Kata kunci**: kehamilan ibu usia lanjut, luaran fetal, luaran maternal*

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INTRODUCTION

Pregnancy in advanced maternal age (AMA) is generally defined as pregnancy in women aged 35 years old or older.[1] The trend of delayed childbearing is increasing, especially in developed countries, since last decade.[2] This situation also happened in Indonesia as it was reported by Indonesia Demographic Health Survey (IDHS) in 2013.[3] This accelerating demographic shift becomes major clinical and public concern.[4] Pregnancy in AMA is prone to have higher complications, whether it is caused by general degenerative process or decrease of reproductive function.[5]

Tangerang General Hospital is a tertiary and referral center in Banten as one of the most crowded provinces in Indonesia.[3] Annual Report of Tangerang General Hospital in 2013 recorded 7,534 deliveries, including 1,683 (22.3%) pregnancies in AMA.[6] Therefore, this study aims to provide a better description of pregnancy outcome in AMA.

METHODS

The cross-sectional study reviewed the demography, medical and obstetrics problems, mode of delivery, maternal and fetal outcome in Tangerang General Hospital, Banten. The data were taken

from medical records of patients with the inclusion criteria of 35 year-old and above women delivering in Tangerang General Hospital from January 1, 2014 to December 31, 2014. Data entry and analysis used Microsoft Excel 2010.

RESULTS

The Characteristics of Patients

There were 6,107 deliveries and 5,926 live births in Tangerang General Hospital from January 2014 to December 2014. Of those, 25.3% or 1,548 cases were pregnancy in AMA. There were nineteen missing cases or incomplete medical records secondary to Death on Arrival (DOA) cases or transferred to other departments.

Table 1 described the advanced maternal age during pregnancy whereas the majority of cases were due to multiparities. While majority of patients were only completed elementary school and the average time was spent for schooling was around 8.27 years.

Maternal Outcomes

There were 38% pregnancies with medical disorders. Hypertension was the most common medical disorders as 529 cases (34.6%) followed by 17 cases (1.1%) of diabetes, 1 case (0.1%) of HIV and 33 cases (2.2%) of other diseases.

Table 1. Maternal Age to Parity and Education Level

	Parity			Education level				
Age	**≤1**	**2 - 5**	**6 ++**	**Less**	**Elm**	**JHs**	**SHs**	**HD1**
35 - <40	76	1060	103	22	546	404	238	29
Percent	6.1	85.6	8.3	1.8	44.1	32.6	19.2	2.3
40 - <45	10	195	61	5	124	51	61	25
Percent	3.8	73.3	22.9	1.9	46.6	19.2	22.9	9.4
≥ 45	2	12	10	5	11	4	3	1
Percent	8.3	50.0	41.7	20.8	45.8	16.7	12.5	4.2
Total	88	1267	174	32	681	459	302	55
	5.8	82.9	11.4	2.1	44.5	30.0	19.7	3.6

Notes:
Less : not completed Elementary school
Elm : completed Elementary school
JHs : completed Junior High school
SHs : completed Senior High school
HD1 : Diploma and higher

Table 2. Maternal Age to Obstetrics Problems and Mode of Delivery

	First Timester			Second & Third Trimester					Mode of Delivery			
Age	**Ab**	**Ect**	**Mol**	**MuP**	**PS**	**MalP**	**PlP**	**PlA**	**Sp**	**CS**	**Vc**	**Fc**
35 - <40	134	14	5	32	199	198	86	23	650	371	57	8
Percent	10.8	1.1	0.4	2.6	16.1	15.9	6.9	1.8	59.8	34.3	5.2	0.7
40 - <45	45	3	8	15	25	49	22	6	136	64	8	2
Percent	16.9	1.1	3.0	5.6	9.4	18.4	8.2	2.2	64.8	30.5	3.8	0.9
≥ 45	6	0	0	2	2	7	0	0	10	7	0	1
Percent	25.0	0	0	8.3	8.3	29.1	0	0	55.5	38.9	0	5.6
Total	185	17	11	49	226	254	109	29	796	442	65	11
Percent	12.1	1.1	0.7	3.2	14.8	16.6	7.1	1.9	60.7	33.6	4.9	0.8

Ab : Abortion
Ect : Ectopic Pregnancy
Mol : Molar Pregnancy
MuP : Multiple Pregnancies
PS : Previous Scar
MalP : Malpresentation
PlP : Placenta Previa
PlA : Placental Abruption
Sp : Spontaneous vaginal delivery
CS : Cesarean Section
Vc : Vacuum Extraction
Fc : Forceps Extraction

**The number and percentage of cases were based on the diagnosis in the labor ward and operating theater reports. It meant one patient might have two diagnosis of the above complication.*

Table 2 showed obstetric problems in the first trimester lead to nonviable conception and problems which revealed in later gestational age. The rate of CS was high (33.6%) because AMA and multiparities had higher risk problems such as malpresentation, previous CS, placenta previa, placental abruption, and multiple pregnancy.

There were total of 66 maternal near-missed cases (56 in 1,000 live births) required special care in ICU or HCU on admission and there were 11 maternal mortality cases (the maternal mortality rate/MMR 932 per 100,000 live births).

Fetal Outcomes

Study of fetal outcomes on pregnancy in AMA showed regardless of spontaneous or medically indicated, premature delivery comprised 26.4% of all deliveries; while, term deliveries were occurred in 849 cases (58%), serotines deliveries were in 13 cases (0.9%), and the rest were less than 20 weeks which could be abortion, molar pregnancy, and ectopic pregnancy. As recorded, another severe outcome as fetal congenital anomaly was presented in 35 cases (2.96%).

Table 3. Birth Weight

Birth weight (grams)	Cases	%
500 - 999	28	2.2
1000 - 1499	42	3.4
1500 - 2499	188	15.0
2500 - 3999	971	77.5
4000 - 4499	21	1.7
4500+	2	0.2
Total	1251	100.0
Abortus, ectopic, molar, non-institutional delivery	278	
Total	1529	

The incidence of Extremely Low Birth Weight (ELBW), Very Low Birth Weight (VLBW), and Low Birth Weight (LBW) were 2.2 %, 3.4%, and 15%; respectively. However, the incidence of macrosomia (>4000 grams) was 1.9%. After excluding early pregnancy loss and non-institutional delivery, there were 5.3% intrauterine fetal death and 12.9% perinatal death.

DISCUSSION

The Age Specific Fertility Rate (ASFR) is the number of live births per 1,000 women in a specific age group for particular geographic area in the point of time annually.[7] Age specific fertility rate (ASFR) for 35-year-old and above Indonesian women was 16.76%; however, this study reported the prevalence was 25.3%.[8] Higher proportion of pregnancy in AMA in this institution showed that this group had major clinical and public health problem because AMA is consistently associated with adverse pregnancy outcomes.

In this study, 5.8% of pregnancy in AMA experienced conceiving for the first time. The rate was lower than in Europe (11%).[2] It showed that pregnancy in AMA in this institution was caused by multiparity compared to delayed childbirth.[9] Indonesia Demographic Health Survey (IDHS) 2013 reported the average number of children for 40-49 year-old women were 3.2 in Indonesia, 3.8 in Banten, and 4.8 in this institution.[3]

In 2013, the average duration of schooling for more than 15 years was 8,62 in Banten Province.[10] Most patients had the educational level only elementary graduated with the average duration of schooling was 8.27 years. The patients' educational level was under the average of Banten province whereas the older population had lower education level.

Education is very important and it contributed to society behavioral changes toward personal or family health. Apart from the formal education, preventive health knowledge needs to be improved through health counseling and seminar for pregnant lady or her family members. For example, Chile was able to reduce 93.8% MMR in 50 years drastically from 293.7 to 18.2 per 100,000. Learning all contributing factors, women educational attainment was the main factors. Educational status will affect birth rate, delivery process attended by skill professional health, sanitation, and personal hygiene.[11]

Medical disorders in this study were 38% consisting of 34.6% cases of hypertension. However, the prevalence of hypertension in pregnancy was reported to be 10-15% in England.[2] It was two to three times more frequent. Therefore, the further prospective study should be conducted to determine the causes of this event. The prevalence of diabetes on pregnancy in AMA was 1.1%, as this

number was lower than in United Kingdom (2-5%) in spite of same screening guideline between Indonesia and United Kingdom.[2] This finding was not correlated with the fact that being older and Asian had more prone to diabetes. Diabetes screening may not performed well in daily practice.

This study found 0.1% of pregnancy in AMA suffering from HIV. In Indonesia, the HIV prevalence in pregnancy was 0.38% which was reported by Ministry of Health (MOH) in 2011. The HIV screening should be offered to all pregnant women as HIV in society like iceberg phenomena whereas the actual incidence is higher.[12]

There was also an increasing trend of miscarriages from 10.8% in 35-40 years old, 16.9% in 40-45 years old to 25% after 45 years old. These numbers were lower than other countries such as Denmark which had 24.6%, 51%, and 93.4%; respectively, because of poor documentation and low educational level in society.[2]

In this study, the prevalence of ectopic pregnancy was 1.1%. This was also documented lower than other countries such as Denmark which showed 1.4% at the age of 21 and 6.9% at the age of more than 44.[2]

Molar pregnancy in this study was reported 4 per 1,000 at the age of 35-40 and 30 per 1,000 at the age of 40-45. This number was much higher than 1 per 1,000 in United Kingdom.[13]

Multiple pregnancy was reported 32 per 1,000 live births which was three times higher on pregnancy in AMA than younger women. However, it was lower than other developed countries as in developed countries had higher Artificial Reproductive Therapy (ART) for pregnancy in AMA, such as in England which was resulting 110 per 1,000 live births.[7]

The prevalence for placenta previa was 7.1% which showed almost twice than reported in Europe, such as 3.7% in Turkish because pregnancy in AMA in Indonesia had the characteristics of higher parity, CS, and curettage. They were the risk factors for placenta previa.[14]

In 1995-2005, Yaniv, et al. reported higher risk of fetal malpresentation in nulliparous in AMA group (6.9% in younger than 35 year-old women and 12.4% in AMA).[15] In this study, the result described 16.6% of fetal malpresentation cases. Higher fetal malpresentation may be caused by multiparity as the tonus and elasticity of abdominal wall were malformed by age, trauma, and stretch due to repeatedly being pregnant.

Caesarean section rate in this study pointed out 33.6%. Majority of the causes were due to cephalopelvic disproportion, previous CS, malpresentation, and fetal distress.[16] Previous CS was documented to be 14.8% in AMA and 4.9% in younger women.[6]

The maternal mortality rate (MMR) in Tangerang general hospital was calculated to be 607.5 per 100,000 live births equal to 1.7 times than MMR in Indonesia globally which counted for 359 per 100,000 live births.[10] This was occurred due to the Tangerang General hospital is a tertiary hospital in Banten Province. As a comparison to other tertiary hospitals in a developing country such as Western Maharashtra Hospital India in 2001-2010, the MMR was 302.23/100,000 live births equal to 1.42 times higher than MMR globally (212 per 100,000 live births).[17]

There were 66 maternal near-missed cases (55.9 in 1,000 live births) that required special care in ICU or HCU during admission, contributing to 11 maternal mortality cases (MMR 932.2 per 100,000 live births in AMA population). It showed that AMA carries high risk for complications.

There was no difference for prevalence of congenital anomaly in older women for this study compared to general population since fetal anomaly and chromosomal screening was not offered to the patients. Premature delivery was 55 per 1,000 deliveries lower than reported by Laopaiboon, et al. which indicated 67 per 1,000 delivery.[1] This was due to the limitation of NICU availability in the observed hospital.

There was no difference for prevalence of congenital anomaly in older women, while the prevalence of stillbirth in AMA was twice than younger women.[18] The perinatal mortality in AMA population was 12.9%. This data was associated with study by Reddy and Ko, et al. in 2001 that they showed the increment of IUFD was 1.28 times in 35-39 year-old women and 1.79 times in 40 year-old women compared to younger women.[19]

The limitations of the study were not included all AMA cases in population; as retrospective study highly depended on consistency and accuracy of documentation. Incorrect entries may lead to wrong conclusion.

CONCLUSIONS

Adverse maternal and fetal outcomes such as maternal near-missed, maternal death, stillbirth, and perinatal mortality were higher in AMA, as it needs special attention and multidiscipline-approached care started from preconception, antenatal, preparation of delivery, and postpartum care.

A meticulous documentation is vital for formulating epidemiological and management strategies that may have considerable implications for the provision of resources in the obstetric and pediatric specialties.

It is also useful for hypotheses-generating studies, or studies directed at one specific association between risk factors and maternal or fetal outcomes in order to confirm or refute possible association. This is in view of the future planning and implementation of the policies and guidelines in the management of advanced maternal age as high risk group.

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Research Article

Heme Oxygenase-1 Level in Normotensive Pregnancy and Preeclampsia with Severe Features

Kadar Heme Oxygenase-1 pada Kehamilan Normotensi dan Preeklamsia dengan Karakteristik Perburukan

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Abstract

Objective: To understand the relationship of heme oxygenase-1 (HO-1) level between normotensive pregnancy and preeclampsia with severe features.

Method: The cross sectional study was conducted in the Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sam Ratulangi/Prof. Dr. R. D. Kandou General Hospital Manado. The subjects consisted of 26 pregnant women with normal blood pressure and 26 women with severe features of preeclampsia. We took the patients' history, general physical examination, and laboratory assessment. The blood samples were taken from normotensive women more than 20 weeks of pregnancy and preeclamptic women with severe features more than 20 weeks of pregnancy. The data obtained was processed using SPSS 20.0 software. We did the non-parametric Mann-Whitney test to analyze the relationship between heme oxygenase-1 (HO-1) level in normotensive pregnancy and preeclampsia with severe features.

Result: The level of heme oxygenase-1 (HO-1) in normotensive pregnant women was at 3.24 (SD 0.58) ng/ml (95% CI 3.00-3.47), and the level of heme oxygenase-1 (HO-1) of preeclamptic women with severe features was 3.92 (SD 0.73) ng/ml (95% CI 3.62-4.21). The result of Mann-Whitney test showed p value of 0.001 which meant that there was significant difference in the level of heme oxygenase-1 (HO-1) between normotensive women and pre-eclamptic women with severe features.

Conclusion: There was the relationship between the level of heme oxygenase-1 (HO-1) and the incidence of preeclampsia with severe features.

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Keywords: heme oxygenase-1 (HO-1), normotensive pregnancy, preeclampsia with severe features

Abstrak

***Tujuan**: Untuk memahami hubungan kadar heme oxygenase-1 (HO-1) pada kehamilan normotensi dan preeklamsia dengan karakteristik perburukan.*

***Metode**: Penelitian ini merupakan studi potong lintang. Penelitian ini dilaksanakan di Bagian Obstetri dan Ginekologi FK Universitas Sam Ratulangi/BLU RSUP Prof. Dr. R. D. Kandou Manado. Subjek penelitian terdiri dari 26 pasien dengan kehamilan normotensi dan 26 pasien dengan kehamilan preeklamsia dengan karakteristik perburukan. Dilakukan anamnesis, pemeriksaan fisik umum dan kemudian dicatat dalam status penelitian, kemudian perempuan dengan kehamilan normotensi >20 minggu dan perempuan hamil >20 minggu dengan preeklamsia dengan karakteristik perburukan diambil sampel darah dan dibawa ke laboratorium. Data yang diperoleh diolah dengan komputer menggunakan perangkat lunak program SPSS 20,0. Uji non parametric Mann-Whitney digunakan untuk menganalisa hubungan antara kadar HO-1 pada kehamilan normotensi dan preeklamsia berat.*

***Hasil**: Pasien hamil normotensi memiliki kadar HO-1 sebesar 3,24 (SD 0,58) ng/ml (95% IK 3,00-3,47), pasien hamil preeklamsia dengan karakteristik perburukan memiliki kadar HO-1 sebesar 3,92 (SD 0,73) ng/ml (95% IK 3,62-4,21). Uji Mann-Whitney menunjukkan bahwa p=0,001 yang berarti bahwa terdapat perbedaan bermakna kadar heme oxygenase-1 (HO-1) pada pasien normotensi dan preeklamsia dengan karakteristik perburukan.*

***Kesimpulan**: Terdapat hubungan bermakna antara kadar HO-1 dengan kejadian preeklamsia dengan karakteristik perburukan.*

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***Kata kunci**: heme oxygenase-1 (HO-1), kehamilan normotensi, preeklamsia dengan karakteristik perburukan*

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INTRODUCTION

Preeclampsia as one of the most devastating obstetric problems is still not solved completely. Preeclampsia was occurred in approximately 1.8 to 16.7% of pregnancies, whereas the incidence of preeclampsia was varied among countries.[1] Based on the systematic review conducted by the WHO, about 16% of maternal deaths in developed countries, including the United States, were caused by hypertension in pregnancy and its complications. This complication passed through other major causes, such as bleeding (13%), abortion (8%), and sepsis (2%).[2] In Indonesia itself, the average incidence of preeclampsia was 3-10% and the maternal mortality rate (MMR) was approximately

4.91%, ranging from 8,739 to 170,725 deaths.[3] Several factors have been associated with the incidence of preeclampsia. In general, these risk factors can be classified as maternal factors (extreme age, parity, history of preeclampsia), medical risk factors (chronic hypertension, diabetes mellitus or renal disease) and factors of the placenta (hyperplacentosis on twin pregnancy and gestational trophoblastic diseases). Nevertheless, these predisposing factors cannot be explained with clear pathogenesis.[4]

In addition to the dominance of several risk factors, a lot of theories have been developed to explain the pathogenesis of preeclampsia, including genetic, immunological, oxidative stress, inflammation, hypoxia, angiogenic, and hormonal factors. Many theories tend to agree that the initial problem is the disruption of trophoblastic invasion, unfortunately, there are still no theories with satisfactory answers which correlating among each factors to the complication of preeclampsia.

In recent years, various evidences show that heme oxygenase-1 (HO-1) is very important in pregnancy. Therefore, we need to understand the mechanisms underlying the protective effect of HO-1. These mechanisms can be varied depending on the reproductive phase of the HO-1 involvement. The microenvironment in which HO-1 performs its action also greatly affects the protective mechanisms. Study on the mechanisms underlying the emergence of HO-1 effects to the process of pregnancy is through the release of CO by HO-1. This process not only occurs in the reproductive process,[5,6] but also in several inflammatory processes.[7,8] This study aims to understand the relationship of HO-1 level between normotensive pregnancy and preeclampsia with severe features.

METHODS

This was a cross sectional study which conducted at the Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sam Ratulangi/ Prof. Dr. R. D. Kandou General Hospital Manado, from March 1st to May 31st 2015. The inclusion criteria include pregnant women with more than 20 weeks of gestational age, having normal blood pressure or preeclampsia with severe features, they would like to fill the informed consent and participate in the study. Meanwhile, the exclusion criteria consist of pregnant women with diabetes mellitus, kidney disorders, heart diseases, chronic hypertension, premature rupture of membranes (PROM), clinical signs of infection, multiple pregnancy, in utero fetal death (IUFD), and also not willing to participate in this study. The data was processed by computer using SPSS 20.0 software. Non parametric Mann-Whitney test was used to analyze the relationship between the level of HO-1 in normotensive pregnancy and preeclampsia with severe features.

Table 1. Characteristics of Subjects

Characteristics	Normal Blood Pressure		Preeclampsia with Severe Features	
	N	%	N	%
Parity				
Primiparous	9	34.61	8	30.76
Multiparous	17	65.38	18	69.23
Gestational Age				
20-28 week	6	23.07	6	23.07
29-36 week	10	38.46	10	38.46
≥ 37 week	10	38.46	10	38.46
Education				
Elementary	1	3.84	4	15.38
Junior high school	2	7.69	3	11.53
Senior high school	23	88.46	15	57.69
University	0	0	4	15.38
Occupation				
Housewife	18	69.23	17	65.38
Student	3	11.53	1	3.84
Private company employee	5	19.23	4	15.38
Government employee	0	0	4	15.38

Table 2. Descriptive Analysis of the Heme Oxygenase-1 (HO-1) Level Differences between Normotensive Pregnancy and Preeclampsia with Severe Features.

Study Variables	Normal Blood Pressure	Preeclampsia with Severe Features	p
Heme oxygenase-1 (HO-1)			
Mean	3.2388	3.9154	0.001
95% Confidence Interval	3.00 - 3.48	3.62 - 4.21	
Minimum	2.41	2.88	
Maximum	4.39	5.67	
Standard Deviation	0.58	0.73	

RESULTS

We got 26 pregnant women with normal blood pressure and the other 26 women with preeclampsia with severe features. Based on the data presented in Table 1, most of the subjects are multiparous (65.38% in normotensive pregnancy and 69.23% in preeclampsia with severe features). The subjects with 29-36 weeks and ≥37 weeks of gestational age had the same percentage between women with normal blood pressure and preeclampsia with severe features (38.46% for each). Most of them were graduated from senior high school (88.46% in normotensive pregnancy and 57.69% in preeclampsia with severe features) and they worked as housewives (69.23% in normotensive pregnancy and 65.38% in preeclampsia with severe features).

Table 2 depicted the differences of HO-1 level in normotensive pregnancy and preeclampsia with severe features women. Of 26 samples in normotensive pregnancy, the average level of HO-1 was at 3.24 ng/ml (95% CI 3.00-3.48), with a standard deviation of 0.58 ng/ml. Meanwhile, of 26 samples in preeclampsia with severe features, the average value of HO-1 level was 3.92 ng/ml (95% CI 3.62-4.21), with a standard deviation of 0.73 ng/ml. The statistical analysis described there was significant relationship between the levels of HO-1 and the incidence of preeclampsia with severe features.

DISCUSSION

This study showed that the level of HO-1 in maternal serum during pregnancy were significantly higher in preeclampsia women with severe features compared with normal blood pressure women. Further more, this study showed a positive correlation between the levels of serum HO-1 and the severity of preeclampsia.

Actually, heme oxygenase is an important enzyme system in the human body. There are three isoforms of heme oxygenase, consisting of inducible HO-1, constitutive HO-2, and HO-3 which function are still unknown.[9] The importance of this enzyme and its catalytic product in the maintenance of normal pregnancy to term has recently been disclosed. Heme oxygenase catalyzes the oxidation of heme into carbon monoxide (CO), biliverdin, and iron, as well as it has a key role in tissue protection against oxidative stress.[10] Today, it is clearly understood that the HO-CO-biliverdin system is involved in the formation of normal placenta and in the hemodynamic control of placenta and fetal tissues. Heme oxygenase system regulation in the placenta is a complex process and it partly depends on the local concentration of glucose and oxygen.[11,12]

Some studies found a decrease in the expression and/or activity of HO-1 in human placenta in pregnancy or in individuals with hypertensive disorders;[9] while, other studies reported a decrease occurred only in the level of HO-2 and not in the level of HO-1.[13,14] Several other studies had shown that there was no difference of HO-1 protein level between pregnancies with mild preeclampsia and uncomplicated pregnancy.[10] However, all these studies were conducted on the levels of HO in the placenta, and only few data were available regarding the changes in maternal serum of HO-1 level in normal pregnancies and pregnancies complicated by preeclampsia. Our results showed that HO-1 level was increased in plasma of preeclampsia women with severe features compared with women with normal blood pressure. There were only two studies conducted by Eide, et al. and Vitoratos, et al. which resulted similar to our study. They found the increased level of HO-1 in serum and decidual tissue, accompanied by changes in the expression

of the decidua. This result supported the role of these substances in oxidative stress and excessive inflammatory responses in the pathogenesis of maternal preeclampsia.[15,16]

Reactive oxygen species (ROS) will be sequestered by antioxidants, which may have non-structural proteins, such as vitamin E, C and A, as well as by metabolites, such as glutathione, ubiquinone and uric acid. Antioxidants based protein include catalase, heme oxygenase, glutathione peroxidase and thioredoxin peroxidase.[17] Normal pregnancies are characterized by a transient increase ROS production which neutralized by the induction of antioxidant defense mechanisms.[18] Preeclampsia is associated with the increased of oxidative stress not only in the placenta, but also scattered in the maternal circulation. It is thought to be part of a systemic inflammatory response.[19,20] Increased oxidative stress occurs as a result of excessive ROS production or due to interference in antioxidant capacity;[17] however, oxidative stress is closely related to the severity of preeclampsia clinically.[21] Heme oxygenase-1 enzyme experiences rapid upregulation through oxidative stress and the induction of HO-1 may protect the cells through binding metaloporphyrin catalyze pro-oxidants, such as heme, bile pigments (biliverdin, bilirubin) that has function as free radicals.[22] We found an increased serum HO-1 in preeclampsia with severe features compared with normal blood pressure group. Previous report showed that the level of oxygen radical absorbance based on the direct neutralization of free radicals was unchanged in women with mild preeclampsia.[23]

In contrast, free irons and particularly carbon monoxide are produced from HO-1 which is mediated by heme catabolism.[24] Previous study explained an increase of serum iron and carboxyhemoglobin concentration in preeclampsia, which reflected the raising of heme and erythrocyte turnover; also it demonstrated that this endogenous production could change the maternal and fetal oxygenation.[25] Thus, there might be a potential link between HO-1 and the severity of preeclampsia. Indeed, based on our results, the serum level of HO-1 seemed to be related to the severity of the disease in preeclamptic women with severe features. Although in those study the serum levels of HO-1 were positively correlated with both the overall study population and in women with preeclampsia, we could not describe the correlation in control group. Vitoratos, et al. through their study showed that the increased activity of HO-1 serum in severe preeclampsia would persist long after childbirth. It explained the key role of persistent oxidative stress, the increased vascular resistance, and the chronic excessive maternal inflammatory response in the pathophysiology of preeclampsia.[16] Therefore, further clinical trial with larger samples and repeated measurement during pregnancy is required to properly evaluate the exact role of HO-1 both in normal pregnancy and preeclampsia.

There are several limitations to our study. The limited number of patients could reduce the strength of this study and in creased the chances of error. As a result, a significant correlation might be missed. The measurement of HO-1 level was only performed once during pregnancy. It would be more accurate if the repeated measurement of maternal serum HO-1 level was performed every week to allow a better assessment of the temporal changes and explain the further interaction.

CONCLUSION

This study assesses the level of maternal serum HO-1 in preeclampsia with severe features as well as in normal pregnancy. Increased level of HO-1 is found in preeclampsia with severe features compared with normal pregnancy, reflecting the significant correlation between HO-1 level and the severity of preeclampsia.

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Research Article

Classic Antiphospholipid and Antiphosphatidylserine Antibody Profile in Suspected Antiphospholipid Antibody Syndrome Patient

Gambaran Hasil Pemeriksaan Antibodi Antifosfolipid Klasik dan Anti Fosfatidilserin pada Pasien yang Dicurigai Sindrom Antifosfolipid

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Abstract

Objective: To compare the classic examination results of antiphospholipid (aPS) and antiphospatidylserine (aPL) antibody profile to establish the diagnosis from suspected antiphospholipid antibody syndrome (APS) patient in order to state the subsequent treatment strategies.

Method: This descriptive cross-sectional study design was conducted at outpatient clinics of Dr. Cipto Mangunkusumo Hospital (RSCM) from January to December 2015. The laboratory test was held in Clinical Pathology Laboratory RSCM/Faculty of Medicine Universitas Indonesia (FKUI) and in corporation with Prodia laboratory.

Result: All of normal patients did not have positive result in any laboratory examination (Lupus Anti-coagulant (LA), anticardiolipin (aCL), anti-β2 glycoprotein I (anti-β2GPI), and aPS). In patient suspected APS, 11 (37.1%) patients had positive aCL, 7 (25.9%) patients had positive anti-β2GPI, and 11 (37.1%) patients had positive aPS. The most positive cross laboratory examination was between aCL and aPS (25.9%). In this study, we found the most positive test result was aCL and aPS (62.9%). From this study, suspected APS patient who had negative result in classic laboratory examination, but showing the positive result in aPS was in 5 (18.5%) patients.

Conclusion: All normal pregnant patients do not have any positive classic examination and aPS. Meanwhile, in patients with suspected APS, less than 20% patients show positive result of aPS with negative result in classic laboratory examination.

[Indones J Obstet Gynecol 2016; 4-3: 138-141]

Keywords: antiphospatidylserine, antiphospolipid syndrome

Abstrak

***Tujuan**: Untuk memberikan perbandingan hasil dari pemeriksaan klasik antibodi antifosfotidilserin (aPL) dan antifosfolipid (aPS) untuk menegakkan diagnosis tersangka sindrom antibodi antifosfolipid sehingga dapat menentukan rencana tata laksana berikutnya.*

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***Hasil**: Semua pasien hamil normal tidak memiliki satu pun pemeriksaan klasik (antikoagulan lupus (LA), antikardiolipin (aCL), anti-β2 glikoprotein I (anti-β2GPI) serta aPS) yang positif. Pada pasien yang dicurigai APS, 11 (37,1%) pasien memiliki pemeriksaan aCL yang positif, 7 (25,9%) pasien memiliki pemeriksaan anti-β2GPI positif, dan 11 (37,1%) pasien memiliki pemeriksaan aPS yang positif. Hasil silang pemeriksaan aPL positif terbanyak adalah antara aCL dan aPS yaitu sebesar 25,9%. Pada penelitian ini didapatkan dua pemeriksaan laboratorium yang positif terbanyak (62,9%), yaitu aCL dan aPS. Dari penelitian ini, didapatkan pada pasien yang dicurigai APS tetapi memiliki hasil negatif terhadap aCL, anti-β2GPI, dan LA, ternyata sebanyak 5 (18,5%) pasien memiliki hasil positif pada pemeriksaan aPS.*

***Kesimpulan**: Seluruh pasien hamil yang normal tidak menunjukkan hasil pemeriksaan klasik dan aPS positif. Sementara itu, pasien yang dicurigai APS, hanya 20% yang menunjukkan hasil aPS positif dengan hasil pemeriksaan klasik negatif.*

[Maj Obstet Ginekol Indones 2016; 4-3: 138-141]

***Kata kunci**: antiphospatidylserine, antiphospolipid syndrome*

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INTRODUCTION

Intra Uterine Fetal Death (IUFD) was defined as a fetal death which occurs in more than 20 weeks of gestational age. Several factors underlie this condition such as fetus (chromosomal abnormality, infection), placenta (placental abruption, placental insufficiency), or maternal (metabolic disorder hypercoagulability state, etc.).[1]

Hypercoagulable state is one of IUFD common causes from maternal factors which are due to the antiphospholipid syndrome (APS). This syndrome is an autoimmune disease characterized by the presence of antiphospholipid antibodies (aPL) and at least one of clinical manifestations, such as arterial or venous thrombosis or fetal death.[2] The incidence of APS in women experiencing IUFD was quite high around 20-40%.[3,4] The perinatal mortality rate in Indonesia was generally about 77 per 1,000 live births whereas 30 stillborn fetuses were caused by APS.[5]

The high incidence of IUFD due to APS makes us to improve the knowledge in diagnosing and performing subsequent management. To diagnose APS, it takes at least 1 clinical criteria namely the presence of one or more unknown causes from fetal deaths at the over 10 weeks of gestational age and supported by laboratory criteria which is the presence of aPL.[2,6,7] There is variety of already known aPL, such as lupus anticoagulant (LA), anticardiolipin (aCL), antiphosphatidylserine (aPL), antiphosphatidylethanolamine (aPE), many more.[8]

The LA and aCL antibodies are first discovered to diagnose APS. One of the screening for LA was through the examination of activated partial thromboplastin time (aPTT).[2] Meanwhile, the examination of aCL has progressed from time to time. Many studies have shown that aCL was not directly bind to the negatively charged phospholipids, but it stuck to the beta-2 Glycoprotein I (β2GPI), a plasma protein or "cofactor" which was attached directly to negatively charged phospholipids.[3]

However, patients sometimes show negative classic examination results (aPL aCL, anti- β2GPI, and LA) in clinically suspected APS. Thus, some experts put those patients in the category of seronegative APS.[9] Meanwhile, other experts are opposite to the above categories which they suggests that it is caused by aPL contained on the patient instead of an antibody to cardiolipin, and β2GPI; however, the antibodies to phosphatidylserine (aPL) is part of the inner cell membrane.[10,11] Study by Matzner, et al. stated that aPL was most often identified as the aPS (20.5%) and aPE (19.1%).[12] Therefore, this study aims to compare the classic examination results of antiphospholipid (aPS) and antiphospatidylserine (aPL) antibody profile to establish the diagnosis from suspected antiphospholipid antibody syndrome (APS) patient in order to state the subsequent treatment strategies.

METHODS

This study was conducted with a descriptive cross-sectional design at outpatient clinics of dr. Cipto Mangunkusumo Hospital (RSCM) from January to December 2015. The laboratory test was taken in Clinical Pathology laboratory RSCM/Faculty of Medicine Universitas Indonesia (FKUI) and Prodia laboratory. The inclusion criteria for the group of normal pregnancy were the one who never experienced any of the Sydney criteria; while, group of APS were the patients suspected of APS (fetus death at more than 10 weeks of gestational age) in accordance with one of the clinical criteria for APS (revised in 2006 in Sydney). The exclusion criteria for normal pregnancies were women having experienced one or more clinical episodes of arterial thrombosis, venous or small blood vessels in the tissue or any organ; and patients with infection (fever, or other symptoms of infection). The exclusion criteria for APS were patients with genetic abnormalities (abnormal morphology of the parents); fetus with abnormalities (confirmed by direct examination and the results of ultrasonography); patients with the quality of the gametes were not good (to be seen on the patients aged > 35 years old); patients with metabolic disorders (elevated blood sugars more than 200). We did the consecutive sampling for all patients who come in and meet the inclusion criteria in the study until the required number of subjects met.

RESULTS

In this study, we got 54 patients who met the inclusion criteria as study subjects. Of the 54 patients, 27 patients had normal pregnancy and 27 patients with suspected APS. As seen in Table 1, all normal pregnant patients did not have any positive of classic examination (LA, aCL and anti-β2GPI) and aPS.

Table 1. Classic aPL Examination Profile (aCL, anti- β2GPI, and LA), and aPS in Normal and Suspected APS Patients

Antibody Examination	Normal P;regnancy (n=27) n (%)	Suspected APS (n=27) n (%)
LA		
Normal	27 (100)	25 (92.5)
Prolonged	0 (0)	2 (7.5)
aCL		
Negative	27 (100)	17 (62.9)
Positive	0 (0)	11 (37.1)
anti-β2GPI		
Negative	27 (100)	20 (74.1)
Positive	0 (0)	7 (25.9)
aPS		
Negative	27 (100)	17 (62.9)
Positive	0 (0)	11 (37.1)

In patients with suspected APS, 11 (37.1%) patients had positive aCL examination, 7 (25.9%) patients had positive anti-β2GPI examination, and 11 (37.1%) patients had positive aPS examination. We tried to look at the classic cross-examination of aPL (aCL, anti- β2GPI, and LA), and aPS in patients suspected of having APS (results not normal or positive). From this study, it was found in 2 (7.5%) patients having positive cross between LA and aCL, LA and anti-β2GPI, as well as LA and aPS. The most positive aPL of cross-examination result was between aCL and aPS (25.9%) (Table 2).

DISCUSSION

In patients with suspected APS, 11 (37.1%) patients had positive aCL examination, 7 (25.9%) patients had positive anti-β2GPI examination, and 11 (37.1%) patients had positive aPS examination. We intend to compare the aPL level of subjects suspected APS with normal pregnant subjects which aims to prove that the levels of aPL in normal pregnancy will not increase.

From this study, it was found in 2 (7.5%) patients had a positive cross between LA and aCL, LA and anti-β2GPI, as well as LA and aPS. This was because only two patients were positive for LA examination. However, sometimes, patients with clinically suspected APS has negative result of classic examination aPL (aCL, anti-β2GPI, and LA). Some researchers put it in the category of seronegative aPS.[9] The diagnosis of seronegative antiphospholipid syndrome (SN-APS) is used in patients with clinical manifestations leading to the APS, but classic examination results of aPL (aCL, anti-β2GPI, and LA) are persistently negative. Until now, the best management of these patients is still unclear. Examination of aPL in addition to classical examination may improve the ability to diagnose APS. However, the availability of aPL in routine laboratory examination is still limited. Patients with typical clinical manifestations of APS, it can have negative results on the examination of LA including IgG and IgM aCL and anti-β2GPI. However, there are several considerations which some patients with negative results on classic examination may have antibodies against other phospholipid membranes, such as phosphatidylserine (PL), phosphatidic acid (PA), phosphatidylinositol (PI), phosphatidylethanolamine (PE) which are not included in the examination of blood routine.[9,10,13] This study investigated the antibodies to phosphatidylserine (PS) as a part of the inner cell mem-

Table 2. Cross-examination Profile of Classic aPL Examination (aCL, anti-β2GPI, and LA) and aPS in Suspected APS Patients (abnormal or positive result)

Examination	LA n (%)	aCL n (%)	anti-β2GPI n (%)	aPS n (%)
LA n (%)		2 (7.5)	2 (7.5)	2 (7.5)
aCL n (%)			3 (11.1)	7 (25.9)
anti-β2GPI n (%)				3 (11.1)
aPS n (%)				

Table 3. Classic aPL Examination Profile (aCL, anti-β2GPI, and LA), and aPS in Suspected APS Patients (only one positive examination)

Antibody Examination	Suspected APS n (%)
LA	2 (92.5)
aCL	11 (62.9)
anti-β2GPI	7 (74.1)
aPS	11 (62.9)

In this study, two most positive laboratory result (62.9%) were aCL and aPS (Table 3). From this study, it was found that in patients with suspected APS, it depicted the negative result against aCL, anti-β2GPI, and LA; nevertheless, it showed the positive result on aPS examination as many as five (18.5%) patients (Table 4).

Table 4. The Proportion of Patients who were Clinically Suspected of Having APS which Had Negative aPL Classic Examination Results (aCL, anti-β2GPI, and LA), but Positive aPS Result

Antibody Examination	Suspected APS n (%)
aPS	5 (18.5)

brane.[10,11] From this study, it was found that in patients with suspected APS, but having negative result against aCL, anti-β2GPI, and LA, it turned out as much 5 (18.5%) patients having positive results on aPS examination (Table 4). This was consistent to study conducted by Matzner, et al. which showed aPL as the most frequently identified antibody and followed by aPS.[12]

The diagnosis of SN-APS is an important issue for the clinician to decide additional examination in diagnosing APS. If the classic examination has a negative result and a thorough evaluation of genetic and acquired conditions leading to thrombosis can be ruled out, we need the additional examination. In the future, it is possible that aPL as the additional examination will be included into the criteria.[9,11]

The strength of this study was that we tried to see the profile of clinically APS suspected patient who pointed out the negative of standard laboratory result, but positive result of APS. Unfortunately, the limitation of this study was we had not done the repeated serology examination after 12 weeks applied as gold standard. The study did not examine patients who had a trip of APS positive test results and did not review the outcome of pregnancy.

CONCLUSION

All normal pregnant patients do not have any positive classic examination and aPS. Meanwhile, in patients with suspected APS, less than 20% patients show positive result of aPS with negative result in classic laboratory examination.

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Research Article

Classic Antiphospholipid and Antiphosphatidylserine Antibody Profile in Suspected Antiphospholipid Antibody Syndrome Patient

Gambaran Hasil Pemeriksaan Antibodi Antifosfolipid Klasik dan Anti Fosfatidilserin pada Pasien yang Dicurigai Sindrom Antifosfolipid

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Faculty of Medicine Universitas Indonesia/
Dr. Cipto Mangunkusumo Hospital
Jakarta

Abstract

Objective: To compare the classic examination results of antiphospholipid (aPS) and antiphospatidylserine (aPL) antibody profile to establish the diagnosis from suspected antiphospholipid antibody syndrome (APS) patient in order to state the subsequent treatment strategies.

Method: This descriptive cross-sectional study design was conducted at outpatient clinics of Dr. Cipto Mangunkusumo Hospital (RSCM) from January to December 2015. The laboratory test was held in Clinical Pathology Laboratory RSCM/Faculty of Medicine Universitas Indonesia (FKUI) and in corporation with Prodia laboratory.

Result: All of normal patients did not have positive result in any laboratory examination (Lupus Anti-coagulant (LA), anticardiolipin (aCL), anti-β2 glycoprotein I (anti-β2GPI), and aPS). In patient suspected APS, 11 (37.1%) patients had positive aCL, 7 (25.9%) patients had positive anti-β2GPI, and 11 (37.1%) patients had positive aPS. The most positive cross laboratory examination was between aCL and aPS (25.9%). In this study, we found the most positive test result was aCL and aPS (62.9%). From this study, suspected APS patient who had negative result in classic laboratory examination, but showing the positive result in aPS was in 5 (18.5%) patients.

Conclusion: All normal pregnant patients do not have any positive classic examination and aPS. Meanwhile, in patients with suspected APS, less than 20% patients show positive result of aPS with negative result in classic laboratory examination.

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Keywords: antiphospatidylserine, antiphospolipid syndrome

Abstrak

***Tujuan**: Untuk memberikan perbandingan hasil dari pemeriksaan klasik antibodi antifosfotidilserin (aPL) dan antifosfolipid (aPS) untuk menegakkan diagnosis tersangka sindrom antibodi antifosfolipid sehingga dapat menentukan rencana tata laksana berikutnya.*

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***Kesimpulan**: Seluruh pasien hamil yang normal tidak menunjukkan hasil pemeriksaan klasik dan aPS positif. Sementara itu, pasien yang dicurigai APS, hanya 20% yang menunjukkan hasil aPS positif dengan hasil pemeriksaan klasik negatif.*

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RESULTS

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CONCLUSION

All normal pregnant patients do not have any positive classic examination and aPS. Meanwhile, in patients with suspected APS, less than 20% patients show positive result of aPS with negative result in classic laboratory examination.

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Research Article

The Level of Ca-125 in Pre- and Post-operative of Endometriosis

Kadar Ca-125 pada sebelum dan setelah Operasi Endometriosis

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Abstract

Objective: To determine the Ca-125 level in pre- and post-operative of endometriosis and its correlation to endometriosis stage and severity of dysmenorrhea.

Method: This was a cross-sectional study design conducted at Dr. Wahidin Sudirohusodo dan some affiliated hospitals. We took the patients undergoing laparoscopy or laparotomy consecutively.

Result: The mean value of preoperative Ca-125 level in stage I-II was 21.53 (SD 12.64) IU/ml vs 72.52 (SD 8.52) IU/ml in stage III-IV. The mean value of postoperative Ca-125 level was 14.82 (SD 10.00) IU/ml (stage I-II) vs 61.03 (SD 8.43) IU/ml (stage III-IV); they were significantly different (p<0.001). There was a significant correlation between Ca-125 level and the stage of endometriosis (r=0.43 and 0.52; p=0.005). We observed mild to severe dysmenorrhea for stage I-II; while, patients had moderate to severe dysmenorrhea in stage III-IV (p<0.001) and both of them showed a significant correlation (r=0.318 and 0.537; p<0.05). The Ca-125 level in stage I-II endometriosis was lower than stage III-IV.

Conclusion: Level of Ca-125 will decrease after endometriosis surgery. There is a strong correlation between preoperative level of Ca-125 and endometriosis stage. The severity of dysmenorrhea has the correlation to the stage of endometriosis.

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Keywords: Ca-125, dysmenorrhea, endometriosis

Abstrak

***Tujuan**: Mengetahui kadar Ca-125 sebelum dan setelah operasi endometriosis dan hubungannya dengan stadium endometriosis dan derajat dismenorea.*

***Metode**: Penelitian dilakukan di BLU RS Dr. Wahidin Sudirohusodo dan rumah sakit afiliasinya dengan menggunakan desain potong lintang pada pasien laparoskopi/laparotomi.*

***Hasil**: Rerata kadar Ca-125 preoperatif pada endometriosis stadium I-II adalah 21,53 (SB 12,64) IU/ml vs 72,52 (SB 8,52) IU/ml pada stadium III-IV. Rerata kadar Ca-125 pascaoperasi adalah 14,82 (SB 10,00) IU/ml (stadium I-II) vs 61,03 (SB 8,43) IU/ml (stadium III-IV), terdapat perbedaan bermakna kadar Ca-125 baik pre- maupun pascaoperasi (p<0,001) dan menunjukkan korelasi bermakna antara kadar Ca-125 preoperatif dan stadium endometriosis (r=0,431 dan 0,524; p=0,005). Dismenorea derajat ringan sampai berat terdapat pada stadium I-II sedangkan derajat sedang sampai berat pada stadium III-IV (p<0,001) dan keduanya menunjukkan korelasi bermakna (r=0,318 dan 0,537; p<0,05). Kadar Ca-125 endometriosis stadium I-II lebih rendah dibandingkan stadium III-IV.*

***Kesimpulan**: Kadar Ca-125 menurun pascaoperatif. Terdapat hubungan antara kadar Ca-125 preoperatif dengan stadium endometriosis. Derajat dismenore berkorelasi dengan stadium endometriosis.*

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***Kata kunci**: Ca-125, dismenorea, endometriosis*

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INTRODUCTION

Endometriosis occurs around 10-15% of reproductive age women. Endometriosis causes a lot of problem, which one of them is infertility. Many studies have proved this relationship; however, the causal relationship between both of them are still controversial.[1]

Actually, the prevalence of endometriosis is unknown. The definitive diagnosis of endometriosis can be performed through laparoscopy or surgery; meanwhile, it will not do to the women without specific symptoms of physical examination suggesting to the diseases. Therefore, there are various indications for surgical treatment for endometriosis.[2,3]

Apart from the infertility problem faced by endometriosis, the pain of endometriosis impacts to quality of life which can be assessed through Health-Related Quality of Life (HRQOL). The pelvic pain is the major causes of productivity loss in reproductive age women. Therefore, we as the health professional should put more awareness to the disease in primary health center by early diagnostic and prompt treatment to improve the productivity of women.[4]

A non-invasive diagnostic test of endometriosis is required to prevent the delay between onset of symptoms and diagnosis of disease. The diagnosis of endometriosis is difficult to determine due to the variation of clinical symptoms. We should consider

endometriosis if patients are complaint about the dysmenorrhea. Dysmenorrhea as a common gynecologic disorder in women with endometriosis at childbearing age is the sensation of pain during menstruation. It usually makes cramping and is located on the lower abdomen.[3,5] Dysmenorrhea affects health and quality of life. Many studies conducted on different populations reported the prevalence was ranged between 20% and 94%.[5-7]

Currently, laparoscopy is the gold standard to confirm diagnosis and treatment of endometriosis. Unfortunately, this procedure has not been performed in most hospitals in Indonesia. Therefore, a simple modality prior to surgery is required to diagnose the endometriosis.

The tumor marker of Ca-125 is the most extensively investigated and used as the peripheral biomarker of endometriosis. The Ca-125 is produced by endometrial and mesothelial cells and released into circulation via the endothelial lining of capillaries in response to inflammation. However, Ca-125 level in the peripheral lacks diagnostic power as a single biomarker of endometriosis.[8] The level of Ca-125 usually elevates in endometriosis, especially in moderate and severe stage of endometriosis. Mol, et al. through their study showed the routine examination of serum Ca-125 in infertile patients could identify a subgroup of patients who are more likely to get benefit from early laparoscopy though, Ca-125 had limited diagnostic performance.[9] Although its limited diagnostic performance, Ca-125 was still examined prior to surgery and it was used as the monitoring tool of the endometriosis development after treatment in patients with epithelial ovarian cancer, endometriosis, endometrial cancer, and also cervical adenocarcinoma.[10]

Therefore, this study aims to determine Ca-125 level pre- and post surgery. Apart from that, we would like to know the correlation to the stage of endometriosis and severity of dysmenorrhea.

METHODS

This cross-sectional study was conducted between May and September 2015 in Dr. Wahidin Sudirohusodo and some affiliated hospitals. The samples were taken from women at reproductive age who had been diagnosed with endometriosis and dysmenorrhea. They would perform the laparoscopy or laparotomy. Then, we collected the blood samples from patients pre- and postoperative to measure the Ca-125 level at Prodia Diagnostic Laboratory Makassar. The endometriosis stage was determined by the American Society of Reproductive Medicine (ASRM) criteria at the time of surgery.[11] The severity of dysmenorrhea associated with endometriosis was measured with Numerical Rating Scale (NRS). Data were analyzed using Mann-Whitney test, Fisher exact test, Wilcoxon and Spearman correlation test.

RESULTS

There were sixty subjects confirmed endometriosis during surgical whereas nineteen subjects had stage I-II and forty-one subjects had stage III-IV. There were no statistically different in age, marital status, infertility, duration of marriage, parity and contraception between stages of the disease (Table 1).

Table 1. Subject Characteristics

Characteristics	Endometriosis Stages n (%)		p
	I-II (n = 19)	III-IV (n = 41)	
Age (year)	31.2	32.7	0.271
Marital Status			
Not married	5(26.3)	7(17.1)	0.493
Married	14(73.7)	34(82.9)	
Infertility			
Primary	12(85.7)	23(67.6)	0.292
Secondary	2(14.3)	11(32.4)	
Duration of Marriage			
<5 years	12(85.7)	22(64.7)	0.181
≥5 years	2(14.3)	12(35.3)	
Parity			
0	7(77.8)	10(47.6)	
<3	2(22.2)	9(42.9)	0.273
≥3	0	2(9.5)	
Contraception			
Non user	12(85.7)	27(79.4)	
OC (hormonal)	2(14.3)	5(14.7)	0.646
IUD (non-hormonal)		2(5.9)	

The mean level of Ca-125 preoperative for stage I-II endometriosis was 21.52 (SD 12.64) IU/ml and 14.82 (SD 10.0) IU/ml for postoperative. In stage

III-IV, preoperative level of Ca-125 was 72.52 (SD 8.52) IU/ml and 61.03 (SD 8.43) IU/ml after the surgical. Actually, the Ca-125 level preoperative for stage I-II endometriosis was higher than stage III-IV. There were significant differences for all of endometriosis stages on Ca-125 level in preoperative and postoperative ($p<0.001$). The stage I-II endometriosis was strongly correlated ($r=0.524$) with preoperative level of Ca-125; while, there was moderate correlation in stage III-IV ($r=0.431$). There was significant correlation between Ca-125 level and all of the endometriosis stages ($p<0.05$) (Table 2).

Patients in stage I-II experiencing mild, moderate, and severe pain of dysmenorrhea was 31.6%, 57.9%, and 2%; respectively. Meanwhile, patients with stage III-IV experienced moderate (51.2%) and severe (48.8%) pain of dysmenorrhea. There were significantly different ($p<0.001$) between severity of dysmenorrhea and endometriosis. The severity of dysmenorrhea had strong correlation with stage I-II ($r=0.537$; $p=0.018$) compared with stage III-IV (Table 3).

DISCUSSION

The results showed a significant difference between Ca-125 level and all of the stages in endometriosis. This study also showed a correlation between the degree of dysmenorrhea and endometriosis stage.

The mean age at diagnosis of endometriosis was between 25 and 35 years old.[3] This study found similar average age of diagnosis for endometriosis where the mean age for stage I-II was 31.2 years old and 32.7 years old for stage III-IV. Actually, our subjects characteristic were alike between groups because they showed the normal distribution ($p>0.05$). Increased age in women affects fecundity and lower fertility. Subfertility due to endometriosis can occur through two mechanisms namely the distortion of adnexal anatomy that inhibits or prevents ovum capture after ovulation and excess of prostaglandins, metalloproteinases, cytokines and chemokines which is resulted into chronic inflammation. This chronic inflammation will impair the ovarian, tubal or endometrial function leading to the disorders of folliculo genesis, fertilization, or implantation. The first mechanism provides a logical explanation for infertility in women with advanced stage of endometriosis. The second mechanism is valid for women with mild stage of endometriosis; nevertheless, the cause of infertility from mild to moderate endometriosis is still controversial.[3,12]

The use of contraception both hormonal and non-hormonal was not significantly associated with all of the stages in endometriosis due to small sample size. Chapron, et al. study showed that there was no relationship between the new user of contraceptive pills and endometriosis. This study also indicated that the history of oral contraceptive user for treating severe primary dysmenorrhea was associated with endometriosis, especially in the type of deep infiltrating endometriosis. However, this did not mean that the use of oral contraception would increase the risk of developing endometriosis. The history of contraceptive pill user

Table 2. Pre- and Post-operative of Ca-125 Level

Endometriosis Stages	Ca-125 Level (mean (SD) IU/ml)		*p*	Ca-125 Preoperative	
	Preoperative	Postoperative		*r*	*p*
I-II	21.53 (12.64)	14.82 (10.00)	<0.001	0.524	0.021
III-IV	72.52 (8.52)	61.03 (8.43)	<0.001	0.431	0.005

Table 3. Dysmenorrhea and Endometriosis Stages

Severity of Dysmenorrhea	Endometriosis Stages (%)		*p*	Stage I-II		Stage III-IV	
	I-II (n=19)	III-IV (n=41)		*r*	*p*	*r*	*p*
Mild	6 (31.6)	0					
Moderate	11 (57.9)	21 (51.2)	0.000	0.537	0.018	0.318	0.043
Severe	2 (10.5)	20 (48.8)					

could use as the marker for women with endometriosis and deep infiltrating endometriosis.[13] A study by Vercellini, et al also reported that the incidence of endometriosis was lower in new user of oral contraceptive pills than old user.[14]

Preoperative level of Ca-125 in stage I-II was lower than stage III-IV. The Ca-125 level in endometriosis was varied on the period of study, although the Ca-125 level increased at advanced stage of endometriosis compared with stage I-II. Barbieri, et al. in their study measured serum Ca-125 level preoperative in 147 patients performed diagnostic laparoscopy or laparotomy. The result pointed out that the serum Ca-125 level increased in stage III or IV endometriosis than in control group. Around 54% of patients with stage III or IV endometriosis and 0% of control group had the level of Ca-125 more than 35 IU/ml.[15] The result was consistent to the previous studies that the level of Ca-125 was higher in advanced stage of endometriosis which the value was more than 35 IU/ml.

Preoperative abdominal preparation showed that women with high level of Ca-125 more than 65 IU/ml (the normal range was under 35 IU/ml) was more likely to have dense omental adhesion, ruptured endometrium as or cul-de-sac obliteration.[3] Another study on 44 women who underwent laparoscopy to confirm the diagnosis of endometriosis showed that the level of Ca-125 was 33.98 IU/ml vs 9.3 IU/ml in control.[16]

Correlation test result showed there was a strong correlation between elevated level of Ca-125 and the stage of endometriosis. The Ca-125 level increases in women with advanced stage, especially during early pregnancy and normal menstruation in women as well as acute PID or leiomyomata.[3] Therefore, the time of blood sampling for measuring Ca-125 level during menstrual cycle affects the result because the high level of Ca-125 occurs during menstruation and immediately after menstruation.[17] Similar finding result concerning higher Ca-125 level during the menstrual phase was presented by Abrao, et al. They stated that the level of Ca-125 marker reached the highest value during the menstrual phase between the first and third day compared with the lutheal phase. Increased marker of Ca-125 in women with endometriosis occurs because the level of Ca-125 is higher in ectopic endometrium compared with entopic endometrium.[18] The increase in Ca-125 can also happen because of the inflammatory reaction altering the endothelial permeability, thereby it allows the Ca-125 enter into circulation.[15]

Another factor that also affects the level of serum Ca-125 is the age. However, several studies reported conflicting results; some studies reported that the level of Ca-125 decreased as increasing the age; while, another study showed the contrary result.[19] The effect of age on level of Ca-125 for all stages in this study was not analyzed.

Laparoscopy for endometriosis is used to remove lesions, adhesion, and cysts through excision, electrocautery, laser vaporization, and repair of pelvic anatomy. Apart from that, it is the preferred method for the treatment of infertile women with mild-moderate endometriosis. Uterosacral nerve ablation and pre-sacral neurectomy may also be considered to relieve the chronic pelvic pain.[20]

The assessment of the dysmenorrhea severity showed that mild to severe dysmenorrhea was occurred in stage I-II of endometriosis; while, moderate to severe dysmenorrhea was happened in stage III-IV. This suggested that the severity of dysmenorrhea increased as the increasing stage of the disease. In a prospective study by Chapron, et al showed that the assessment of the dysmenorrhea severity was similar in women with superficial endometriosis, cystic ovarian endometriosis, and deep infiltrating endometriosis where the assessment was measured using a visual analog scale. Besides, the severity of pain was higher in women without endometriotic lesion.[21] The relationship between the stages of endometriosis based on the Revised American Society Fertility was inconsistent to the dysmenorrhea. Several studies reported a correlation between the stage of endometriosis and dysmenorrhea.[22,23] Other studies indicated there was no correlation.[24,25] Study by Chapron, et al. showed a linear relationship between the stage of endometriosis and the severity of endometriosis through severe adnexal adhesion and obliteration of the Douglas pouch. It was strongly correlated to the degree of visceral adhesion infiltration or deep infiltrating endometriosis implant.[21] This explanation supported the result in this study that moderate to severe dysmenorrhea was occurred in stage III-IV; while, mild to severe dysmenorrhea in stage I-II was due to endometriotic implant superficial infiltration.

The pain due to mild endometriosis was more likely to be associated with the focal inflammation

from cyclic bleeding surrounding the peritoneal implant or the action of inflammatory cytokines by a large number of macrophages and other immune cells in the peritoneal fluid of women with endometriosis. Cytokines are produced by most cell types including endometriotic tissue which has the role in the pathogenesis of endometriosis and infertility.[26] Severe dysmenorrhea and dyspareunia are common symptoms of endometriosis. The intensity of pain because of deeply infiltrating endometriosis is correlated with the depth of penetration and proximity or direct invasion of the nervous system. However, the severity of endometriosis does not have correlation with the number and severity of dysmenorrhea. Meanwhile, women with advanced stage of endometriosis may experience mild or without dysmenorrhea as well as the dysmenorrhea in women with mild or moderate endometriosis.[3]

CONCLUSION

The Ca-125 level is decreased after surgical. There is a strong correlation between preoperative level of Ca-125 and endometriosis stage. The severity of dysmenorrhea has the correlation to the stage of endometriosis. The examination of preoperative level of Ca-125 and severity of dysmenorrhea is able to identify the possibility of endometriosis although it is not recommended to determine the stage of endometriosis.

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Research Article

Incidence of Post-Operative Urinary Retention after Pelvic Organ Prolapse Reconstruction

Insidensi Retensio Urin Pascarekonstruksi Prolaps Organ Panggul

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Abstract

Objective: To determine the incidence of post-operative urinary retention after pelvic organ prolapse reconstruction and associated factors.

Method: This was a prospective cohort study conducted in Dr. Cipto Mangunkusumo and another associate hospital. We recruited women planned for pelvic organ prolapse reconstruction from April 2013 to April 2015. Inclusion and exclusion criteria were women with pelvic organ prolapse (2nd, 3rd and 4th degree) without prior urinary retention, drugs affecting bladder function, and history of bladder injury. After surgery, urinary catheter was applied for 24 hours. Six hours apart from urinary catheter released, residual urine was measured. Urinary retention was defined as residual urine more than 100 ml.

Result: Of 200 subjects, 59 of them (29.5%) classified as having urinary retention. No association found between age, body mass index (BMI), degree of prolapse, degree of cystocele and urinary tract infection toward urinary retention. Total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation and reconstruction duration more than 130 minutes were associated with urinary retention (relative risk (RR) 3.66; 95% CI 2.91-4.60; p<0.001 and 1.66; 95% CI 1.07-2.59; p=0.02; respectively).

Conclusion: The incidence of post-operative urinary retention after pelvic organ prolapse reconstruction is quite high. Reconstruction duration and type of reconstruction are associated with urinary retention.

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Keywords: pelvic organ prolapse reconstruction, postoperative urinary retention, risk factor

Abstrak

***Tujuan**: Untuk mengetahui insidensi retensio urin pascarekonstruksi prolaps organ panggul dan faktor-faktor yang berhubungan.*

***Metode**: Penelitian kohort prospektif ini dilaksanakan di RS Dr. Cipto Mangunkusumo dan RS jejaring dengan mengikutsertakan perempuan yang hendak mengalami rekonstruksi prolaps organ panggul dalam rentang waktu April 2013 hingga April 2015. Kriteria inklusi dan eksklusi subjek meliputi perempuan dengan prolaps organ panggul derajat 2, 3 dan 4 tanpa riwayat retensio urin sebelumnya, konsumsi obat-obatan yang dapat menyebabkan retensio urin dan tanpa cedera kandung kemih. Pascarekonstruksi, subjek dilakukan pemasangan kateter urin selama 24 jam. Kemudian, enam jam pascapelepasan kateter, dilakukan pengukuran residu urin pada kandung kemih. Retensio urin didefinisikan dengan didapatkannya residu urin lebih dari 100 ml.*

***Hasil**: Dari 200 subjek, ditemukan 59 subjek (29,5%) mengalami retensio urin. Tidak ada hubungan antara faktor risiko umur, Indeks Massa Tubuh (IMT), derajat prolaps, derajat sistokel, dan kejadian infeksi saluran kemih terhadap retensio urin. Jenis prosedur total vagina hysterectomy + kolporafi anterior + kolpoperineorafi + sacrospinous fixation, dan durasi operasi > 130 menit berhubungan dengan retensio urin dengan RR 3,66; 95% IK 2,91-4,60; p<0,001 dan RR 1,66; 95%IK 1,07-2,59; p=0,02.*

***Kesimpulan**: Insidensi retensio urin pascarekonstruksi prolaps organ panggul cukup tinggi. Durasi operasi dan jenis rekonstruksi berhubungan dengan retensio urin.*

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***Kata kunci**: faktor risiko, prolaps organ panggul, retensio urin pascaoperasi*

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INTRODUCTION

Pelvic organ prolapse (POP) causes morbidity for women as it can decrease the quality of life, both physically and sexually. As a main management procedure, POP reconstruction surgery has been performed 200.000 times annually in USA.[1,2] However, this number was only representative for those who underwent POP reconstruction, not a total population of women suffered from POP as it was estimated 24% of women aged above 20 years old.[3] Until now, the management of POP was based on stadium and symptoms of the patients through both conservative and surgical procedures as available options.[4] One of complications after reconstruction surgery that could prolong the duration of hospitalization and patients quality of life was the post operative urinary retention.[5]

The incidence of post-operative urinary retention was varied among institutions. In a study reporting patients underwent vaginal mesh procedure, post urinary retention after POP reconstruc-

tion was accounted among 34% subjects.[6] Meanwhile, 29% patients underwent colporaphy experienced urinary retention.[7]

The main cause of post-operative urinary retention was the anesthetic and anatomical injury related to POP reconstruction.[8,9] Theoritically, all types of reconstruction have risk to cause post operative urinary retention.[10] Other possible risk factors for post operative urinary retention consisted of age, body mass index (BMI), and urinary tract infection.[11] Management of post-operative urinary retention was through chatheterization. However, the use of catheter itself could lead to urinary tract infection. Current data estimated that urinary tract infection after colporaphy was around 10-46%.[12,13] In the other hand, the use of catheter in a short duration could lead to abnormal spontaneous bladder emptying, which increased the risk of urinary retention.[14] Identification of risk factors associated with POP reconstruction is important for clinicians to be aware of susceptible patients and get prepared for further patients management. Currently, there is still a few prospective studies evaluating potential risk factors for post operative urinary retention among patients undergoing POP reconstruction in Indonesia. Therefore, this study aims to determine the incidence of post-operative urinary retention after pelvic organ prolapse reconstruction and factors associated with it.

METHODS

This was a prospective cohort study which recruited women underwent POP reconstruction in two referral public hospitals, Dr. Cipto Mangunkusumo and another associate hospital. This study was held from April 2013 to April 2015.

The inclusion criteria of the study were women underwent POP reconstruction for 2nd, 3rd, and 4th degree of POP. Subjects with prior history of urinary retention, taking drugs that potentially affect bladder function, and prior bladder injury were excluded. As this study tried to evaluate the potential risk factors in cohort study, we used rule of thumb formula to estimate the sample size. Prior study revealed that the incidence of post urinary retention could be around 29%. Thus, we calculated the sample size as ten times risk factors divided into the incidence rate.

We identified age, body mass index (BMI), degree of prolapse, degree of cystocele, duration of surgery, type of reconstruction, and urinary tract infection as potential risk factors. As a result, 241 women were calculated as the minimum sample size. We recruited the samples consecutively.

All subjects who would like to participate in this study had to sign the informed consent. All subjects got prophylactic antibiotic (1 gram of co-amoxiclav) one hour prior POP reconstruction. Patients were allowed to drink after surgery (around 2,000 cc in 24 hours). Twenty-four hours after POP reconstruction, the catheter was released by trained nurse or doctor in charge in recovery room. Six hours apart, patients were asked to urinate by themselves. Catheter was then applied to measure residual urine in bladder. Urine dipstick test was used to screen for urinary tract infection.

Urinary retention was defined as loss of spontaneous urinary process six hours after catheter being released with residual urine in bladder more than 100 ml. Body mass index was classified according to Asia-Pacific criteria by World Health Organization.[15] Degree of prolapse was classified based on Pelvic Organ Prolapse Quantification System (POP-Q).[16] Urinary tract infection was assessed using one positive result from either nitrit or leucocyte esterase test by dipstick.[17,18]

Data analysis was performed using SPSS statistics for Windows version 22. The numerical data were analyzed for its normality using Kolmogorov-Smirnov test. Dependent variable was urinary retention; while, independent variables were the risk factors. Duration of operation cut-off for categorization was assessed statistically using receiver operating curve (ROC). Chi-square or Fisher exact as its alternative were used for categorical data analysis. Independent t-test was also used for numerical bivariate analysis. We took the p value less than 0.05 as statistically significant. All bivariate analysis with initial $p<0.2$ were entered for multivariate analysis using logistic regression test.

RESULTS

Of 200 subjects recruited, 59 of them (29.5%) were experienced post-operative urinary retention. The median of residual urine among subjects at six hours after urinary catheter being released was 112.2 (7-780 for the minimum-maximum) ml. Most of the subjects aged above 50 years old (170 subjects; 85.0%). The characteristics of patients were presented in Table 1. Meanwhile, the type and du-

ration of POP reconstruction were shown in Table 2. The median duration of POP reconstruction was 130.0 minutes. We determined more than 130 minutes as a cut-off point to classify prolong duration of POP reconstruction. Fifty-one percent subjects experienced more than 130 minutes duration of POP reconstruction. Total vagina hysterectomy + anterior colporaphy + colpoperineoraphy was the most frequent type of reconstruction being used among subjects (128 subjects; 64.0%). Sixty-six subjects (33.0%) were positive for either nitrit or leukocyte esterase.

Table 1. Patients' Characteristics (n=200)

Characteristics	**n**	**%**
Age (median, min-max)	60.5 (34.0-87.0) years old	
≤ 50 years old	30	15.0
> 50 years old	170	85.0
BMI (median, min-max)	24.0 (15.6-35.2) kg/m^2	
Underweight	8	4.0
Normoweight	68	34.0
Overweight	58	29.0
Obesity I	56	28.0
Obesity II	10	5.0
Degree of prolapse		
Second degree	59	29.5
Third and fourth degree	141	70.5
Degree of cystocele		
Second degree	39	19.5
Third and fourth degree	161	80.5
Hospital		
Dr. Cipto Mangunkusumo	140	70.0
Another associate hospital	60	30.0

Table 2. Duration and Type of POP Reconstruction

Reconstruction Procedure	**n**	**%**
Duration in minutes (median, min-max)	130.0 (45.0-360.0)	
≤ 130 minutes	103	51.5
> 130 minutes	97	48.5
Type of reconstruction		
Colpocleisis	28	14.0
Colpoperineoraphy	4	2.0
Total vagina hysterectomy +anterior colporaphy + colpoperineoraphy	128	64.0
Anterior et posterior colporaphy	26	13.0
Total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation	6	3.0
Anterior colporaphy + colpoperineoraphy + sacrospinous fixation	8	4.0

Bivariate analysis between various risk factors for post operative urinary retention was shown in Table 3. Duration of POP reconstruction more than 130 minutes and TVH + anterior colporaphy + colpoperineoraphy + sacrospinous fixation were two identified factors which had association with post operative urinary retention with relative risk (RR) 1.66 (1.07-2.59 95% CI; p=0.02) and 3.66 (2.91-4.60 95% CI; p<0.001); respectively. Age, BMI, degree of prolapse, and degree of cystocele did not have association with urinary retention. The correlation analysis using Spearman test between duration of reconstruction and residual urine had a coefficient of 0.18 (p=0.012). However, TVH + anterior colporaphy + colpoperineoraphy + sacrospinous fixation could not include into multivariate analysis as this factor had a cell with 0 (zero) value; thus, the odds ratio (OR) could not be calculated statistically. From multivariate analysis, only duration of POP reconstruction more than 130 minutes showed a significant association with urinary retention (RR = 1.8) (shown in Table 4).

Table 3. Bivariate Analysis between Postoperative Urinary Retention and its Risk Factors

Risk factors	Post-operative urinary retention		Relative Risk (95% IK)	p
	Yes (n=59)	No (n=141)		
Age (years old)	60.0	61.0		0.59[a]
(median, min-max)	(37.0-74.0)	(34.0-87.0)		
≤ 50 years (%)	7 (23.3)	23 (76.7)	1	0.42[b]
> 50 years (%)	52 (30.6)	118 (69.4)	1.31 (0.66-2.60)	
BMI	24.2	23.8		0.60[a]
(median, min-max)	(16.2-35.2)	(15.6-34.1)		
Not obese (%)	38 (29.5)	91 (70.5)	1	0,98[b]
Obese (%)	21 (29.6)	50 (70.4)	1.00 (0.64-1.57)	
Degree of prolapse (%)				
mild	17 (28.8)	42 (71.2)	1	0.89[b]
Advanced (3&4)	42 (29.8)	99 (70.2)	1.03 (0.64-1.66)	
Degree of cystocele (%)				
mild	9 (23.1)	30 (76.9)	1	0.33[b]
advanced (3&4)	50 (31.1)	111 (68.9)	1.07 (0.94-1.24)	
Duration of reconstruction in minutes	150	120		0.05[a]
(median, min-max)	(60-360)	(45-285)		
≤ 130 minutes (%)	23 (22.3)	80 (77.7)	1	0.02[b]
>130 minutes (%)	36 (37.1)	61 (62.9)	1.66 (1.07-2.59)	
Type of POP reconstruction (%)				
Colpocleisis	6 (21.4)	22 (78.6)	0.69 (0.33-1.46)	0.31[b]
Colpoperineoraphy	0	4 (100.0)	0.33 (0.02-4.63)	0.19[b]
Total vagina hysterectomy + anterior colporaphy + colpoperineoraphy	41 (32.0)	87 (68.0)	1.28 (0.80-2.05)	0.29[b]
Anterior et posterior colporaphy	3 (11.5)	23 (88.5)	0.36 (0.12-1.06)	0.03[b]
Total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation	6 (100)	0	3.66 (2.91-4.60)	<0.001[c]
Anterior colporaphy + colpoperineoraphy + sacrospinous fixation	3 (37.5)	5 (62.5)	1.28 (0.51-3.23)	0.61[c]
Urinary tract infection				
Nitrit or leukocyte esterase positive	21 (31.8)	45 (68.2)	1.12 (0.72-1.75)	0.61[b]
Negative	38 (28.4)	96 (71.6)	1	

[a]*Mann-Whitney test,* [b]*Chi-Square test,* [c]*Fisher-exact test*

Table 4. Regression Analysis of Duration of POP Reconstruction

Duration	Urinary retention (%)	No urinary retention (%)	OR (95% CI)	Probability	RR
> 130 minutes	36 (61.0)	61 (43.3)	2.05 (1.10-3.82)	0.409	1.80
≤ 130 minutes	23 (39.0)	80 (56.7)		0.227	refference

**Probability =1/(1+exp (-y)) with y as the regression equation; y = constant+β\ixi*

DISCUSSION

In this study, we found that the incidence of post-urinary retention was 59 subjects (29.5%). Previously, there was similar study using case control method which found that the post-urinary retention after POP reconstruction in Dr. Cipto Mangunkusumo Hospital. Priyatini, et al. found the incidence of post-urinary retention after POP reconstruction was 29%.[19] Our study improved the quality of previous study by using prospective cohort method compared to retrospective study conducted before. The criteria of urinary retention in this study was also similar with Priyatini, et al. study which meant residual urine more than 100 ml at six hours after urinary catheteter being released. Another study described that the incidence of post urinary retention after surgery was around 5-70%.[20] Especially for patients undergoing urogynecology surgery, the incidence of urinary retention after surgery was reported to range from 2.5 to 43%.[21]

This study found there was no association between obesity based on BMI also age and urinary retention after POP reconstruction. Previously, it has been known that these two factors were preoperative risk factors for post-operative urinary retention.[21] Pelvic organ prolapse reconstruction is the highest risk for urinary retention in the field of gynecology. Prolapse repair surgery can cause the alteration in vesico-urethral junction which interfering the urination. In addition, the procedure involves the retropubic space and vagina. This can cause misdisruption of nerve branches which contributes to neuropathy and finally, the neuropathy impacts to the urinate sensation. In addition to age and BMI, advanced degree of prolapse (3rd and 4th degree) and urinary tract infection (UTI) were also known to increase the risk of urinary retention.[21] Urinary retention and UTIs are two related processes. The presence of residual urine more than 30 mL after reconstruction was related to the risk factor for urinary retention.[22] In this study, advanced degree of prolapse was not associated with the urinary retention. A study by Hakvoort, et al. showed that urinary retention was associated with the degree of cystocele. Advanced degree of cystocele (3rd and 4th degree) increased the risk of post-operative urinary retention after POP reconstruction.[10] Unfortunately, we did not assess this risk due to advanced degree of cystocele in the study.

In this study, it was found that the total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation was associated with urinary retention after POP reconstruction with the relative risk 3.66; 95% CI 2.91 to 4.60 (p <0.001). Moreover, the duration of surgery more than 130 minutes was associated with the urinary retention. It was noteworthy that 100% of subjects with total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation had duration more than 130 minutes. A study by Priyatini, et al. held in Dr. Cipto Mangunkusumo hospital patients concluded that there were no risk factors for the type of procedure which was related to urinary retention after POP reconstruction.[19] Geller, et al. declared intraoperative factors such as blood loss more than 100 ml, administration of fluids more than 750 ml and more doses of anesthesia due to longer duration of surgery were associated with urinary retention.[21] This result was similar to study by Lamonerie, et al. which explained that the duration of surgery more than 120 minutes increased the risk of urinary retention with OR 3.03, 95% CI 1.39-6.61.[23]

The use of bupivacaine spinal anesthesia in all subjects of this study was not the reason for high number of patients experienced urinary retention. From the literature, it was known that spinal anesthesia was a risk factor for urinary retention as it interrupted the afferent and afferent nerve fibers disrupting the micturition reflex. Blockade of afferent nerve fibers would cause bladder analgesia while blockade of the afferent nerve fibers interfered with detrusor muscle motor activity.[24] However, as the measurement of post void residual (PVR) at 24 hours post-procedure, the anesthetic

effect had disappeared. It was known that the time to return to normal voiding function in patients with bupivacaine was up to 362 minutes (7-8 hours).[21]

The limitation of this study included insufficient sample size targets. In this study, 241 subjects were expected to be fulfilled. However, due to the limited time of the study, only 200 subjects collected for analysis.The results of this study showed statistically significant ($p<0.05$) for certain risk factors; therefore, it was not mandatory to calculate the power of this study. Besides, UTI identification based solely on urine dipstick test. Although we did not use gold standard of urine culture, positive results of nitrites or leukocyte esterase test had been reported to have a sensitivity and specificity more than 80% and 77%; respectively.[18]

CONCLUSION

There is no relationship between age, BMI, degree of prolapse, degree of cystocele, or post-reconstruction ISK and the post operative urinary retention. Prolong duration of reconstruction and total vagina hysterectomy + anterior colporaphy + colpoperineoraphy + sacrospinous fixation are associated with post-operative urinary retention after POP reconstruction.

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Research Article

The Prevalence and Risk Factors of Stress Urinary Incontinence in Postpartum

Prevalensi dan Faktor Risiko Inkontinensia Urin Tipe Tekanan Pascasalin

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Abstract

Objective: To know the incidence of stress urinary incontinence in postpartum and determine the relationship among age, parity, infant birth weight, mode of delivery, episiotomy and perineum rupture.

Method: This study used cross sectional analytic design. Qualified subjects from inclusion criteria were interviewed by researchers using pre-defined MESA questioner. Subjects with stress urinary incontinence were found from the questionnaire result. The acquired data was measured and analyzed using SPSS v. 22.0 software and discussed using available literature.

Result: From 162 subjects, 36 cases (22.22%) had stress urinary incontinence, 47.22% aging ≥ 35 years old, 72.22% had multiple pregnancies, 88.89% had per vaginal delivery. Using multivariate logistic regression test, we found there was a relationship between stress urinary incontinence with age and parity ($p<0.05$).

Conclusion: There is a relationship between stress urinary incontinence with women aging > 35 years old and multiple parities.

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Keywords: multiple parities, post-partum, stress urinary incontinence

Abstrak

***Tujuan**: Mengetahui prevalensi stres inkontinensia urin pascapersalinan, mengetahui apakah terdapat hubungan antara faktor usia, paritas, berat lahir bayi, cara melahirkan, episiotomi dan ruptur perineum dengan kejadian stres inkontinensia urin pascapersalinan.*

***Metode**: Penelitian ini menggunakan desain penelitian potong lintang analitik. Sampel yang memenuhi kriteria inklusi dilakukan wawancara oleh peneliti dengan menggunakan kuesioner MESA yang telah ditetapkan. Dari hasil MESA Questionnaire Incontinence didapatkan sampel menderita stres inkontinensia urin. Data yang diperoleh diolah dengan menggunakan perangkat lunak SPSS versi 22.0 dan dilakukan pembahasan menggunakan teori kepustakaan yang ada.*

***Hasil**: Seratus enam puluh dua responden yang memenuhi kriteria sebanyak 36 kasus (22,22%) mengalami SIU. usia ≥35 tahun 47,22%, multigravida (72,22%), partus pervaginam (88,89%). Hasil uji regresi logistik multivariate ($p<0,05$) terdapat hubungan antara stres inkontinensia urin dengan usia dan paritas.*

***Kesimpulan**: Terdapat hubungan antara stres inkontinensia urin dengan usia 35 tahun dan multiparitas.*

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***Kata kunci**: multiparitas, pascasalin,stres inkontinensia urin*

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INTRODUCTION

Stress Urinary Incontinence (SUI) is an involuntary outflow of the urine which causes social and hygiene problem.[1] This problem is more common in postpartum women and it will affect their physical and quality of life because the symptoms may change their body image and confidence due to the unpleasant odor. Apart from that, it causes the limitation to their daily activities, such as shopping, dancing, playing with their children, laughing, and sneezing.[1,2]

In 2006, World Health Organization (WHO) stated that 200 million people globally had urinary incontinence. In North America, the number of it reached 13 million whereas it attacked to 85% women and 18-21% of them had SUI at 6 week-postpartum period. It was related to the prolonged active labor period, macrosomia, and assisted vaginal delivery.[1,2]

Urinary incontinence is a medical condition problem for women which the prevalence is twice higher than men. Several studies reported the prevalence of it was around 20-30% and actually, this number was too small compared to the reality of unreported cases.[3] According to Yunizaf, et al. in 2002, these cases were alike iceberg phenomenon in Indonesia because it was difficult to accept the stigma of urinary incontinence in women, particularly after laboring. Besides, the women are usually ashamed to get this condition.[4]

Stress urinary incontinence is a common problem in postpartum period. In USA, 18-21% of stress urinary incontinence was found at 6 week-postpartum women. In cases of vaginal delivery, SUI happens due to the excess stretch to the anterior levator ani muscles group causing the weakness and the damage to muscles which finally, it cannot maintain their function properly in closing the urethra from bladder pressure.

Muscle and nerve tissue damage in vaginal delivery is the main contributing factor to the urinary incontinence. In addition, pregnancy itself is a source of mechanical and hormonal change contributing to this symptom.[5] In vaginal delivery, the stretch stress occurs not only to the anterior levator muscle group, but also to the neck of bladder, urethra sphincter muscle, and its ligament.[4] Several risk factors were found to increase the incidence of SUI in postpartum, such as age, parity, delivery, birth weight, episiotomy, spontaneous perineum rupture, assisted vaginal delivery by vacuum or forceps. Some studies concluded that SUI in postpartum was contributed to 40-60% from the total incidence of incontinence.[4,5]

Pregnancy and delivery are the predisposition to urinary incontinence and is the risk will be higher in multiple parities.[1] The weakness of muscle group from the first pregnancy is untreated and the pressure from the sequential pregnancies causes the stress to endure.[6,7] Stainton, Strahle and Fethney in 2005 confirmed that women with urinary incontinence in their first pregnancy had 4.14 times higher risk to have urinary incontinence in their sequential pregnancies.[8]

The first delivery, macrosomia baby, and parities are some causes of becoming complication in delivery.[2,3] Daneshgari, et al. found that the length of delivery time could cause damage to nerve and levator ani muscle group.[3]

Several studies concluded that urinary incontinence was a serious health problem in postpartum period and it could easily disrupt the daily activity.[9-11] Therefore, this study aims to know the incidence of SUI in postpartum and determine the relationship among age, parity, infant birth weight, mode of delivery, episiotomy and perineum rupture.

METHODS

This study used analytical cross-sectional design for 162 subjects who were postpartum with vaginal and abdominal delivery for more than 42 days in Prof. Dr. R.D. Kandou General Hospital and other affiliated hospitals from December 2015 to May 2016. Data was collected using interview and Medical, Epidemiological, and Social aspects of Aging (MESA) questionnaire.

We included all 15-45-year-old postpartum women using vaginal or abdominal delivery who came to Prof. Dr. R.D. Kandou General Hospital and affiliated hospitals and they would like to participate in this study. Subjects qualifying through inclusion criteria were interviewed and asked to fill out the questionnaire. From the MESA questionnaire, we sorted the samples showing SUI. We excluded the patients having complications on their pregnancy (chronic hypertension, pregnancy-induced hypertension, diabetes, IUFD), a history of pelvic abnormalities and urinary tract diseases, undergoing major surgery (hysterectomy, myomectomy, cystectomy, or salpingectomy) after childbirth and 3-month postpartum, also they had history of urinary incontinence complaint.

The MESA Incontinence Questionnaire consists of 15 questions including 9 questions for the SUI type and 6 others for the urge urinary incontinence (UUI) type. We give the score for each question. The maximum score was 27 of 9 questions for the SUI and 18 of 6 questions for UUI. To determine the predominant type of SUI, the percentage score obtained divides into the maximum possible total score. The SUI is considered predominant when the score as a percentage of the stress is greater than the urge (SUI $\geq$ 25% UUI).

Each patient enrolled in this study was adjusted to the research ethic principles. Patients who were willing to participate in the study after giving the counseling had to sign on the informed consent. As the compensation of the willingness to join our study, we gave Kegel tutorial video for 8 minutes in CD to practice at home. This education and practice were considered as the first-line treatment of SUI.

RESULTS

This study was held from December 2015 to May 2016 by involving 162 subjects and 36 of them

were diagnosed with SUI (22.22%) in postpartum. Other obtained risk factors were age, parity, infant birth weight, delivery method, episiotomy, and perineum rupture.

From the data based on subjects' characteristics, we found that SUI happened in less than 35 years old age group was 52.78% and 47.22% for more than 35 years old. Most of subjects (72.22%) were multiparity. Based on educational level, 4 (11.11%), 12 (33.33%), 3 (8.33%), 12 (33.33%), and 5 (13.89%) subjects were uneducated, elementary school, junior high school senior high school, and undergraduate graduate; respectively. Based on occupation, most of subjects (75%) were housewives.

Stress urinary incontinence was happened in <35-year-old postpartum women (19 subjects, 52.78%) and 17 subjects (47.22%) for ≥ 35-year-old postpartum women. According to the number of parities, 26 subjects (72.22%) had multiple parities. The educational level of subjects was mainly elementary and high school graduate (33.33% for each). Most subjects were also housewives (75%).

Table 2. The Distribution of SUI

	Category	Stress Urinary Incontinence	
		N	%
Age (years)			
	<35	19	52.8
	≥35	17	47.2
Parities			
	Single	10	27.8
	Multiple	26	72.2
Infant Birth Weight (grams)			
	< 4000	33	91.67
	≥ 4000	3	8.33
Mode of Delivery			
	Vaginal birth	32	88.89
	SC	4	11.11
Episiotomy			
	Yes	8	22.22
	No	28	77.78
Perineum Rupture			
	Yes	12	33.33
	No	24	66.67

Table 1. Subjects' Characteristics

	Characteristics	Stress Urinary Incontinence		Normal	
		N	%	N	%
Age	<35 years old	19	52.78	101	80.16
	≥ 35 years old	17	47.22	25	19.84
Parity	Single parity	10	27.78	57	45.23
	Multiple parities	26	72.22	69	54.77
Education	Uneducated	4	11.11	4	3.17
	Elementary School	12	33.33	16	12.69
	Junior high School	3	8.33	27	21.43
	Senior High School	12	33.33	56	44.44
	Undergraduate	5	13.89	23	18.25
Occupation	Housewives	27	75.00	88	69.84
	Entrepreneur	6	16.67	23	18.25
	Housemaid	3	8.33	15	11.90

Table 3. The Bivariate Analysis for the Relationship among Age, Parity, Mode of Delivery, Episiotomy to SUI

Independent Factors	OR	95% C.I.		p-value
		Lower	Upper	
Age	3.509	1.389	8.868	0.008
Parity	3.014	1.036	8.775	0.043
Mode of delivery	2.664	.886	8.014	0.081
Episiotomy	0.354	.113	1.108	0.075

Table 3 pointed out the relationship between several associated factors and SUI. Of the several factors, only age and parity had the influence to the occurrence of SUI. More than 35-year-old women had 3.509 (95% CI 1.389-8.868) times to be SUI. Apart from that, multiparity had 3.014 time to have SUI.

DISCUSSION

We measured the relationship between SUI and patients' age. Stress urinary incontinence was found in 17 cases in ≥ 35-year-old age group (47.22%). There was 3.5 times higher risk of more than 35-year-old women to have SUI. In this study, we concluded that SUI incidence would raise as the increase of age.

Thomason, et al. reported that many women aging from 35-50 years old had SUI symptoms.[1] The notion was supported by Glazener in 2006 reporting that older women had higher chance to acquire SUI compared with younger women (OR 2.02, 95% CI, 1.35-3.02).[12] Meanwhile, Kondon, et al. found that SUI was more prevalent in > 40-year-old women.[3,5] In this study, we also found that more incidence of SUI were occurred in those with advance age. Lower urinary tract may decrease the endurance as the increase of age. The muscle strength to close the urethra decrease as people grow older.[1,2]

Based on their babies' birth weight, SUI was found higher in babies born with <4000 grams (91.67%) compared to ≥ 4000 grams (8.33%). Multivariate logistic regression test showed that birth weight (p>0.25) had no significant relationship with the SUI. In this study, we indicated more SUI incidence in women with history of delivery <4000 grams. Susanto, et al. study in Palembang and Santy in Jakarta also pointed out the same result. Statistical analysis could not find a significant relationship between birth weight and SUI.[13-15] From the literature, pressure and stretching of the hip muscles by the fetus during vaginal delivery is one of the causes for the damage muscle; thus, bigger babies create higher pressure to the structure. This study had more samples of <4000 gram of birth weight; hence, it was unable to find a significant relationship between SUI and birth weight.

Glazener in 2006 concluded that birth weight had no significant relationship with incontinence either in labor or after delivery.[12] The same study was conducted by Ali HS, et al. in Pakistan which found that the SUI mostly happened in 3-month postpartum (53.3%) and there was no significant result (p=0.946) between incontinence and birth weight.[13]

Based on parities, multiple parities group had more incidence of SUI compared with single parity. The multivariate logistic regression test showed a significant relationship between multiparity and SUI. Pregnancy and delivery are the predisposition factors for urinary incontinence and it is found higher in women with multiple parities compared with single parity.[1] The reason is the weakness of muscle group in the first pregnancy went untreated and the recurrent pressure for the sequential pregnancy causes more injury to the muscles.[6,7]

In this study, we found that those with multiple parities had 3 times higher risk for SUI compared with first parity. Nygaard in 2006 concluded that after their first delivery, women had twice risk for incontinence in their next.[16] Chaandini in Michigan also found that SUI was found mainly in women with multiple parities (23 subjects from 131 total).[17] Delivery causes the stretching of auxiliary muscle tissue during labor and disrupts the function of urethra sphincter in contraction.[3]

Vaginal delivery group had 32 cases (88.89%) of SUI. Multivariate logistic regression test did not indicate the relationship between mode of delivery and SUI. This result was supported by previous finding by Mason in Liverpool which found that SUI in postpartum was 31%.[6] The more parity rate, the higher the SUI incidence. There was no difference in SUI prevalence in vaginal delivery and assisted vaginal delivery. The incidence of SUI in caesarean section was lower compared with vaginal spontaneous delivery.[3,18]

In study held in Pakistan, they examined a total of 141 women in their 3-month postpartum and

they did not found the significant result between SUI and mode of delivery. Similar study by Glazener in 2006 identified other factors related to incontinence in pregnancy and postpartum. The study was conducted in 3,405 women with single parity and 29% of them had urinary incontinence. Vaginal delivery method might cause higher incontinence incidence compared with those with caesarean section (OR 0.28, 95% CI 0.19-0.41) and there was no significant relationship between vaginal delivery and caesarean section.[12]

Eight subjects (22.22%) from those who underwent episiotomy had SUI, whereas 28 subjects (77.78%) who did not undergo episiotomy had SUI. Using multivariate logistic regression test, we found no significant relationship between SUI and episiotomy. According to DeLancey, the hip can be divided into 4 levels namely level 1 consists of endopelvic fascia, level 2 consists of hip diaphragm muscles, level 3 consists of urogenital diaphragm muscles, and level 4 consists of external anal sphincter and perineal bodies.[6,18] Hargrove, el al. in 2011 reported that episiotomy increased the risk of three of four degree perineal tear, wound infection, and postpartum hemorrhage without decreasing the long term complication, such as perineal pain or SUI. In this study, we found no relationship between SUI and episiotomy ($p>0.05$) due to the lack of rupture degree data caused by episiotomy.

Based on perineum rupture, SUI was found mostly in those without rupture (66.67%). Perineum rupture causes damage to several muscles group in the hip region, particularly transversal perineum muscle.[5] Multivariate logistic regression test showed no significant relationship between SUI and perineum rupture. This finding was supported by Eason, et al, study which found no statistical significance between SUI and spontaneous perineum rupture.[19] Similarly, Ali HS, et al. found no significant relationship between SUI and perineum rupture in 3-month postpartum in a total of 141 women ($p = 0.197$).[13]

CONCLUSION

There is a relationship between stress urinary incontinence with women aging ≥ 35 years old and multiple parities.

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Research Article

Papsmear Examination for Diagnosing Pre-Cancer Lesion in Invisible Squamo-Columnar Junction

Pemeriksaan Papsmear dan IVA untuk Diagnosis Lesi Prakanker pada Tampilan Sambungan Skuamo-Kolumnar Tidak Tampak

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Abstract

Objective: To know the concealed pre-cancer lesion in women with invisible squamo-columnar junction (SCJ) by Papsmear examination.

Method: This study was a descriptive cross-sectional design starting from August 2014 to March 2015 at several Public Health Cares in Jakarta. A total of 1,682 subjects were screened by Acetoacetate Visual Inspection (AVI) examination. After the data was collected, the process was continued by verification, editing, and coding. The descriptive analysis showed the percentage of SCJ in age distribution, the percentage of AVI examination based on SCJ, and the percentage of Papsmear examination in invisible SCJ according to negative AVI result.

Result: There were 1,484 (88.2%) women with the visible SCJ and 198 (11.8%) women with invisible SCJ. The percentage of invisible SCJ in the menopausal women group was 122 (61,6%); meanwhile, in the non-menopausal women group, it was 76 (38.4%). Almost half of the percentage from visible SCJ was found in menopausal women group 45.8% (103/225 women). The positive AVI result was 4 (7.1%) in the menopausal women group and 52 (92.9%) in non-menopausal women group. The result of Papsmear examination with invisible SCJ were 197 (100%) normal.

Conclusion: Almost half of visible SCJ was found in menopausal women group. Most of positive AVI result was found in the non-menopausal women group. All women with the invisible SCJ have a normal Papsmear result.

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Keywords: acetoacetate visual inspection, papsmear, pre-cancer lesion, squamo-columnar junction

Abstrak

***Tujuan**: Untuk mengetahui kelainan yang tersembunyi pada keadaan sambungan skuamo-kolumnar (SSK) tidak tampak melalui pemeriksaan Papsmear.*

***Metode**: Penelitian ini merupakan deskriptif potong lintang. Penelitian dilakukan pada periode Agustus 2014 sampai Maret 2015 di beberapa puskesmas di Jakarta. Sebanyak 1.682 subjek yang dilakukan pemeriksaan IVA (Inspeksi Visual dengan Asam asetat). Setelah data dikumpulkan, akan dilakukan verifikasi data, editing, dan proses pengkodean. Analisis data deskriptif berupa variabel kategori yaitu persentase letak SSK berdasarkan distribusi usia, persentase hasil pemeriksaan IVA berdasarkan SSK, dan persentase hasil pemeriksaan Papsmear pada SSK yang tidak tampak dari hasil pemeriksaan IVA negatif.*

***Hasil**: Perempuan dengan SSK yang tampak 1.484 (88,2%), yang tidak tampak 198 (11,8%). Sambungan Skuamo-Kolumnar (SSK) yang tidak tampak pada perempuan yang sudah menopause sebanyak 122 (61,6%), sedangkan pada perempuan yang belum menopause sebanyak 76 (38,4%). Hampir setengahnya proporsi SSK yang tampak didapatkan pada kelompok perempuan yang sudah menopause 45,78% (103/225 perempuan). Hasil pemeriksaan IVA positif didapatkan 4 (7,1%) pada kelompok perempuan menopause dan 52 (92,9%) pada kelompok perempuan yang belum menopause. Pada pemeriksaan Papsmear dengan SSK yang tidak tampak, persentase kelainan lesi prakanker yaitu sebesar 197 (100%) normal.*

***Kesimpulan**: Hampir setengahnya SSK yang tampak ditemukan pada kelompok perempuan menopause. Sebagian besar IVA positif ditemukan pada kelompok perempuan yang belum menopause. Seluruh perempuan dengan SSK yang tidak tampak memiliki hasil pemeriksaan Papsmear normal.*

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***Kata kunci**: IVA, lesi prakanker, papsmear, sambungan skuamo-kolumnar*

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INTRODUCTION

Cervical cancer is the second most common cancer that affects women in the world.[1-4] Apart from that, it is the most common causes of death among cancer, especially for women in developing countries.[4] Based on the recent world's estimation, there are 493,000 new cervical cancer cases occurred each year, whereas there are 409,400 (83%) cases happened in women in developing countries and only 84,400 (17%) cases in developed countries.[2] Unfortunately, more than 80% of the cases are diagnosed at advanced stage when the 5-year survival rate is less than 40%.[2]

The main cause of high incidence of cervical cancer in developing countries is due to the lack of effective screening programs to detect and manage the early stage of cervical cancer or pre-cancer lesions of the stage.[4] Of the various modality screening which have been studied, Papsmear has ability to decrease 70% of cervical cancer cases; however, this examination is difficult to implement in developing countries like Indonesia. It seems that visual inspection of the cervix is the best techniques to apply, especially in areas with limitation of health facilities resources.[2,4-7] This technique is known as the VIA test (Visual Inspection with Acetic acid application). It is defined as a technique of cervical direct observation after being applied with acetic acid without the use of any magnifying tools.

Visual Inspection of Acetic acid examination depends on the Squamo-Columnar Junction (SCJ) condition in the cervical region. In childbearing age women and pregnant women, SCJ is located in the ecto-cervical that it can be seen on direct examination. Whereas in postmenopausal women, SCJ is often located in the endo-cervical canal so it cannot be seen on direct observation. However, the percentage of visible SCJ in menopausal women is 64.28% and invisible SCJ is 11.66%.[8] This rate is high for the menopausal women. Another study conducted by Dhaubhadel, et al. prospectively and descriptively in women aged 20-50 years, the result showed negative VIA Papsmear test from all 46-50 year of group women.[2] The study did not describe the visibility of SCJ in the investigation. Therefore, it could make the false negative results. If the SCJ is not visible, it suggests doing the Pap smear test. Unfortunately, the problem is in the first-line health care facility, such as primary health centers with limited facilities, the invisible SCJ (in the second examination), particularly in menopausal women is justified without doing the VIA examination. Therefore, this study aims to determine the percentage of hidden invisible SCJ in abnormal cervical pre-cancer lesions through the Pap smear examination.

THEORITICAL OVERVIEW

Invasive cervical cancer is usually preceded by a long phase of pre-invasive lesions, which are microscopically seen as precursor lesions developing from atypical cells to the various level of cervical intraepithelial neoplasia (CIN) before progression to invasive carcinoma. The epidemiological studies have identified several risk factors that contribute to the development of the CIN and cervical cancer. The risk factors are Human Papilloma Viral (HPV) infection, sexual contact in early age, changing sexual partners, multi-parity, long-term of oral contraceptive use, smoking, low socioeconomic status, infection with Chlamydia trachomatis, micronutrient deficiency described by less intake of vegetables and fruits. The types of HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59 and 68 are closely related to CIN and invasive cancer. Infection of one or more HPV types is suspected as the cause of cervical neoplasia. Infection of one or more of the oncogenic HPV types will contribute to the entrance of combined viral genome into the host cell genome; this formation will lead to cell neoplasia and turn into various level of CIN and finally, it will further develop into cervical cancer.[9-13]

Application of 5% acetic acid is believed to cause a reversible coagulation or precipitation of the cell protein. This application also causes swelling of the epithelial tissue and cell dehydration. The color of normal squamous epithelium is pink; while, the columnar epithelium will be in red color due to the reflection of light from the stroma in which its bottom is rich for blood vessels. If the epithelium contains a lot of protein cells, the acetic acid will coagulate this protein which will eliminate the color of the stroma. The result from this coagulation production is called as aceto-white area that can be seen with the naked eye and distinguished from the normal area which should be pink around it. Therefore, the effect of acetic acid depends on the amount of protein found in epithelial cells. Areas where the increased activity of the nucleus and DNA will change very clearly.[9,13]

When acetic acid is applied to normal squamous epithelium, the coagulation will occur in the superficial cell layer. Although the deeper part of the cell has more protein core, acetic acid cannot penetrate; thus, the result of precipitation is not sufficient enough to remove the color in the stroma. The CIN region and invasive cancer are an area where the protein coagulation will occur maximally because they contain of higher core protein and prevent light passing through the epithelium. As a result, the arrangement of blood vessels in the sub-epithelial disappears that the thick epithelium appears white. In the CIN, aceto-white area is limited to the transformation zone close to the SCJ; while, in cervical cancer, aceto-white area is seen around the cervix. Aceto-white area is not typical

for the CIN and early stage of cancer. This area can also be seen in other conditions due to the increase of core proteins, such as immature squamous metaplasia, healing or regenerating epithelium (associated with inflammation), leucoplakia (hyperkeratosis), and condyloma.[9,12,13] The symptoms of cervical cancer are vaginal bleeding, post-coital bleeding, vaginal discharge, lower abdominal pain, edema of lower extremity, obstructive uropathy, bowel obstruction, and also anemia.[12,14]

Aceto-white area in CIN and early stage of cancer are more whitish, thicker, and opaque as well as clear boundary; while, the aceto-white area in immature squamous metaplasia, the inflammation and regenerating epithelium will make less pale, thin, translucent, and ill defined. Aceto-white area caused by inflammation and epithelial healing process is usually spread to the cervix, not only in the transformation zone. Besides, this color will be disappeared within a minute. Leucoplakia and condyloma turn the color into grayish white after the application of acetic acid. The effect of acetic acid (aceto-white area) is slower in CIN lesions and early invasive cancer than immature squamous metaplasia and inflammation. This effect occurs after 3-5 minutes at CIN 2-3 and invasive cancer.[9,13]

METHODS

This cross-sectional design study is conducted in several primary health centers in Jakarta on the period of August 2014 to March 2015. The target population is all women who are married or have ever had sexual intercourse for VIA examination. We included women who did the VIA screening examination in primary health centers Jakarta in the period of August 2014 to March 2015. We excluded pregnant women and women with history of total hysterectomy. From the calculation formula, the total number of subjects needed in this study is 1,658 subjects. In this study, we performed VIA examination on 1,682 women.

All women that met the criteria of the study were labeled and subsequently did the Papsmear examination and the results were interpreted by expert pathologist. The result of Papsmear examination would be described with Bethesda system criteria in 2001. The data was analyzed where we conducted the descriptive statistics for categorical variables, namely the proportion of SCJ layout based on the age distribution, the proportion of the VIA examination results based on the location of the SCJ of the age distribution, and the percentage of the Papsmear examination results of the SCJ which was not visible from the IVA. All data were shown in frequency and percentage.

RESULTS

Of 1,682 subjects, only 9 (0.5%), 311 (18.5%), 638 (37.9), 492 (29.3), 232 (13.8%) women were less than 20, 20-29, 30-39, 40-49, and more than 50 years old; respectively.

Table 1. The Characteristics of the Subjects

Characteristics	n	%
Age		
< 20 yo	9	.5
20-29 yo	311	18.5
30-39 yo	638	37.9
40-49 yo	492	29.3
≥ 50 yo	232	13.8
Menopause state		
Not yet	1457	86.6
Menopause	225	13.4
Contraception state		
Not using contraception	354	21.0
Pill	127	7.6
Implant	43	2.6
Injection	603	35.9
IUD	470	27.9
MOW	27	1.6
Condom	58	3.4
SCJ		
Invisible SCJ	198	11.8
Visible SCJ	1484	88.2
Total	1682	100.0

In this study, according to the age group, we divided into group who were still menstruation (premenopausal state) and had stopped menstruation (menopausal state). There were 1,457 (86.6%) women who were still menstruation (premenopausal) and the others had stopped menstruation.

Table 1 showed that the majority of women (603 women (35.9%)) had ever used the injectable contraception. Meanwhile, the rate of permanent contraceptive acceptor (MOW) was only 27 (1.6%) women. There were 354 (21.0%) women had not ever used contraception. There were 1,484 (88.2%) women with visible SCJ and 198 (11.8%) women with invisible SCJ. The proportion of invi-

sible SCJ based on age group for more than 50, 40-49, 30-39, 20-29, less than 20 years old was 127 (64.1%), 35 (17.7%), 30 (15.2%), 6 (3.0%), and 0 (0.0%); consecutively.

Invisible SCJ proportion in women who had experienced menopause was 122 (61.6%) and 76 (38.4%) for premenopausal women. Almost half of visible SCJ obtained in the group of women who were menopausal (45.78% (103 women)).

Table 2 depicted the proportion of visible SCJ. For age group of 30-39, 40-49, 20-29, and less than 20 years old, there were 608 (41.0%), 457 (30.8%), 305 (20.6%), and 9 (0.6%); consecutively. The proportion of visible SCJ in menopausal women was 103 (6.9%) and 1,381 (93.1%) women in premenopausal state. However, in the premenopausal group, nearly half of 103 (45.77%) women had visible SCJ.

Table 2. The Proportion of Squamo-Columnar Junction (SCJ)

Characteristic	SCJ			
	Invisible SCJ	%	Visible SCJ	%
Age				
< 20 yo	0	0.0	9	0.6
20-29 yo	6	3.0	305	20.6
30-39 yo	30	15.2	608	41.0
40-49 yo	35	17.7	457	30.8
≥ 50 yo	127	64.1	105	7.1
Menopause State				
Not Yet	76	38.4	1381	93.1
Menopause	122	61.6	103	6.9

Table 3. Proportion of VIA Examination with Visible SCJ

Characteristic	VIA			
	Negative	%	Positive	%
Age				
< 20 yo	8	0.6	1	1.8
20-29 yo	291	20.4	14	25.0
30-39 yo	589	41.2	19	33.9
40-49 yo	439	30.7	18	32.1
≥ 50 yo	101	7.1	4	7.1
Menopause State				
Not Yet	1329	93.1	52	92.9
Menopause	99	6.9	4	7.1

In this study, the overall positive VIA test result was in 56 (3.98%) women. The proportion of positive VIA test result according to the age group were 19 (33.9%) for 30-39 years old, 18 (32.1%) for 40-49 years old, 14 (25.0%) for 20- 29 years old, 4 (7.1%) for more than 50 years old, and 1 (1.8%) for less than 20 years old. While, the proportion of positive VIA test result was obtained for 4 (7.1%) in menopausal women and 52 (92.9%) in menstruating women (Table 3).

In the Pap test with SCJ which was not visible, every test presented the normal result (100%) (Table 4). In this study, invisible SCJ was obtained in 198 subjects, but there was one subject that was not checked due to loss of data. However, this number still met the minimum of required sample (196 subjects).

DISCUSSION

This study is based on the data from the examination in several health centers in Jakarta. In this study, the total of 1,682 women were analyzed

Table 4. Proportion of Papsmear Test Result in Invisible SCJ

	ASCUS	%	LSIL	%	HSIL	%	Normal	%
Age								
20-29 yo	0	0.0	0	0.0	0	0.0	6	3.0
30-39 yo	0	0.0	0	0.0	0	0.0	27	13.7
40-49 yo	0	0.0	0	0.0	0	0.0	36	18.3
≥ 50 yo	0	0.0	0	0.0	0	0.0	128	65.0
Menopause stase								
Not Yet	0	0.0	0	0.0	0	0.0	74	37.6
Menopause	0	0.0	0	0.0	0	0.0	123	62.4

based on their characteristics of age, history of contraception, menopausal status, state of the SCJ, the results of the VIA examination and Papsmear. This study began between August 2014 and March 2015. Women with positive VIA test result were evaluated on subsequent visits. While the women had cervical abnormalities, such as cervicitis, it would be treated with topical antiseptics. If the doctor found other abnormalities, such as cervical polyps and suspicious cancer, the women would be sent to higher level of health care facility.

This study has strengths and limitations. The strengths of this study were the sample was taken by doctors and health workers who have had the training and experience from the Female Cancer Program (FCP) in identifying the SCJ, the abnormalities in the cervical region, the procedures for VIA and conventional smear of Pap-smear examination sampling. Additionally, in this study, one pathologist performed Papsmear examination. There were some flaws in this study among others, namely in the Pap-smear examination, sampling errors might occur that could affect the results.

In this study, there were 225 (13.37%) menopausal women. The proportion of invisible SCJ was among 198 (11.7%) women, whereas almost half of postmenopausal women had visible SCJ. In the study conducted by Nuranna, et al. in October 2007 until December 2010, it showed that of 3,791 postmenopausal women (16.49%), the proportion of invisible SCJ was on 2,680 (70.69%) women. More than half of the menopausal women had visible SCJ (2,437 (64.28%) women).[8]

In this study, in Pap-smear with invisible SCJ, all results showed normal (100%). Shwe, et al. conducted a study of cervical cytology in Myanmar from 2010 to 2011. Of 1,771 women screened, 762 women (43.0%) resulted in abnormal smear, 866 (48.9%) and 87 (4.9%) were diagnosed as inflammation and Atypical Squamous Cells of Undetermined Significance (ASCUS). There were 42 (2.3%) and 11 (0.6%) cases of Low Grade Squamous Intraepithelial Lesion (LSIL) and High Grade Squamous Intraepithelial Lesion (HSIL). The cases of squamous cell carcinoma (SCC) occurred on 3 (0.2%) women.[15] While the study conducted by Sengul, et al., there were 32,578 cases of Pap-smear examination performed and analyzed between January 2001 and April 2010. From the investigation which had carried out, the results showed that 1.18% of ASCUS; 0.39% of LSIL; 0.16% of HSIL; 0.07% of Atypical Glandular Cells of Undetermined Significance (AGUS); 0.02% of squamous cell carcinoma, and 0.006% of adenocarcinoma 0.006%.[16] Abnormal cytology is more common in cases with older age, low parity, and period of perimenopause.[16] However, this study did not have data on SCJ picture.

In this study, all results were 100% normal cervical cytology at the invisible SCJ. However, this did not completely rule out pre-cancer lesions abnormalities. Study by Pan, et al. explained that overall, the level of sensitivity, specificity, positive predictive value, negative predictive value, and accuracy of cervical cytology for detecting cervical intraepithelial neoplasia (NIS 2+) were respectively 81.0 %, 95.4%, 38.3%, 99.3% and 94.9%.[17] A study held by Pak, et al. which aimed to compare the history of previous Papsmear test result in patients with cervical adenocarcinoma and squamous cell carcinoma of the cervix. In patients with cervical adenocarcinoma, the false negative results of Papsmear were very significant in the latest investigation. As already mentioned above, the normal Papsmear results did not guarantee not to develop the cervical cancer. In general, 157 patients (41.8%) carried out repeated screening within 2 years. Fifty-five (14.6%) carried out repeated screening within 2-5 years and 80 (22.3%) over 5 years. A total of 16.8% did not have documentation for Pap-smear examination results. The false negative results of Pap-smear for patients with cervical adenocarcinoma and squamous cell carcinoma were 9 (5.6%) and 2 (1.3%) patients.[18] Kirschner, et al. stated that the false negative of Pap-smear result was on 11 (9.8%) women.[19]

CONCLUSION

Almost half of invisible SCJ is found in the group of menopausal women and most visible SCJ is found in the group of women who have not menopausal yet. Positive VIA results are largely found in the group of women who have not menopausal yet. All the women with invisible SCJ have normal Papsmear examination results.

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Research Article

Postradical Hysterectomy Survival Rate in Early Stage Cervical Cancer Patients

Kesintasan Pasien Kanker Serviks Stadium Awal Pascahisterektomi

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Abstract

Objective: Determining the survival rate of early stage cervical cancer patients after radical hysterectomy.

Method: A retrospective cohort study was conducted on 24 early-stage cervical cancer patients who had performed radical hysterectomy in Prof. dr. R.D. Kandou Hospital Manado during the period between January 2008 and December 2010. We used Kaplan-Meier methods to observe the survival rate.

Result: The mean age of patients in this study was 47.8 (33-63) years old with a median of 45 years old. The largest proportion was less than 50 years old (66.7%) and stage IIA cervical cancer (66.7%) as the severity of cancer. Most histopathology type was the squamous cell carcinoma (50.0%). About 70.8% patients did not have lymph nodes metastasis and 62.5% patients did not receive adjuvant therapy. This study revealed that 1-year, 2-year, 3-year, 4-year, and 5-year survival rate were 100.0%, 100.0%, 95.8%, 83.0%, 70.8%; respectively.

Conclusion: The survival rate of early stage cervical cancer after radical hysterectomy in Manado is quite high.

[Indones J Obstet Gynecol 2016; 4-3: 164-169]

Keywords: cervical cancer, radical hysterectomy, survival rate

Abstrak

***Tujuan**: Mengetahui kesintasan pasien kanker serviks stadium awal yang dilakukan histerektomi radikal.*

***Metode**: Penelitian ini menggunakan desain penelitian kohort retrospektif yang menganalisa 24 pasien kanker serviks stadium awal yang dilakukan histerektomi di RSUP Prof. dr. R.D. Kandou Manado pada kurun waktu Januari 2008 hingga Desember 2010. Data yang diperoleh diolah menggunakan analisis kesintasan Kaplan-Meier.*

***Hasil**: Didapatkan rerata usia pasien adalah 47,75 tahun (rentang 33-63 tahun) dengan median 45 tahun. Proporsi terbanyak pada penelitian ini adalah pasien dengan usia < 50 tahun (66,7%) dan pasien kanker serviks stadium IIA (66,7%). Sebagian besar pasien memiliki jenis histopatologi karsinoma sel skuamosa (50,0%). Pada 70,8% pasien tidak terdapat penyebaran KGB dan 62,5% pasien tidak mendapatkan terapi adjuvan. Pada penelitian ini didapatkan kesintasan 1 tahun sebesar 100,0%, 2 tahun sebesar 100,0%, 3 tahun sebesar 95,8%, 4 tahun sebesar 83,0% dan kesintasan keseluruhan 5 tahun adalah sebesar 70,8%.*

***Kesimpulan**: Kesintasan pasien kanker serviks stadium awal pascahisterektomi radikal di Manado secara keseluruhan cukup tinggi.*

[Maj Obstet Ginekol Indones 2016; 4-3: 164-169]

***Kata kunci**: histerektomi radikal, kanker serviks, kesintasan*

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INTRODUCTION

Cervical cancer is the leading causes of death in gynecological cancers scope worldwide, which are nearly half a million cases diagnosed each year.[1,2] The current data estimated 493,243 women worldwide diagnosed with cervical cancer each year; while, 273,505 women died due to cervical cancer, and approximately 90% of these new cases and death was occurred in developing countries.[3,4] In Indonesia, based on data from 11 Pathology departments across Indonesia in 2002, cervical cancer was the top ten of most common cancers in men and women which was contributed as 24%.[5]

In North Sulawesi, there were 1,615 women diagnosed with cervical cancer in 2013. Based on registration data of patients in the Prof. dr. R.D. Kandou hospital from January 2014 to July 2015, there were 156 patients with cervical cancer who were treated.[6]

According to the increase of life expectancy around the world, especially for woman, it is essential to know the survival rate in patients with cervical cancer. Kim, et al. study in Korea in 2000 stated that through the 101-month follow up as the median, the 5-year survival rate in 292 patients diagnosed with stage Ib cervical cancer was 92%

and in 74 patients with stage IIa cervical cancer was 87%.[7] Study held by Bulk, et al. in 2003 reported that in Netherland, the 5-year survival rate in 1,441 patients with cervical cancer was 71%.[8] As well as the latest study in the United States by Brookfield, et al. in 2008 depicted that the median survival rate of the 5,367 cervical cancers was 43 months.[9] Meanwhile, according to study by Sirait, et al. in 1997, patients with cervical cancer in Dr. Cipto Mangunkusumo hospital had 5-year survival rate of 30% with a median survival of 213 patients was 934 days (31 months).[10]

Clinical classification system for cervical cancer by the International Federation of Gynecology and Obstetrics (FIGO) is considered effective in predicting the outcome of cervical cancer. Unfortunately, we usually observe the poor prognosis in cervical cancer patients with the same stage. To clarify this disparity, many studies have tried to identify the prognostic factors associated with cervical cancer. Several factors, such as pelvic lymph node metastasis, tumor size, depth of cervical stromal invasion, hematogenous or lymphatic dissemination, histopathologic type, and tumor extension to the corpus uteri, vagina, parametrial limit the surgical resection which were related to the prognostic value.[11,12] Therefore, this study aims to determine the survival rate of early stage cervical cancer patients after radical hysterectomy.

METHODS

The design of this study was a retrospective cohort study with Kaplan-Meier survival analysis. This study was conducted in Prof. dr. R.D. Kandou Hospital, Manado from July to December 2015. The population in this study were all patients with early stage cervical cancer who had performed radical hysterectomy in Prof. dr. R.D. Kandou Hospital, Manado over the period of 1 January 2008 to 31 December 2010.

We excluded the incomplete data on histopathology in medical record, cervical cancer as not the primary cancer, and unidentified medical records. The data were processed using Kaplan-Meier survival analysis to look for overall 1-year, 2-year, 3-year, 4-year, and also 5-year survival rate.

RESULTS

Based on the medical records in Prof. dr. R.D. Kandou hospital Manado and Pathology Department, Faculty of Medicine Universitas Sam Ratulangi, there were 24 patients recruited in this study. The mean age of patients was 47.8 (33-63) years old with the median of it was 45 years old. The highest proportion of patients was below 50 years old (66.7%) and stage II cervical cancer (66.7%). Most patients had histopathologic type of squamous cell carcinoma (50%). Of 70.8% patients, there were no lymph node metastasis and 62.5% patients did not receive adjuvant therapy.

The figure 1 described the analysis result of using the Kaplan-Meier on 24 subjects. We found that 1-year, 2-year, 3-year, 4-year, and 5-year survival rate were 100%, 100.0%, 95.8%, 83.0%, 70.8%; respectively.

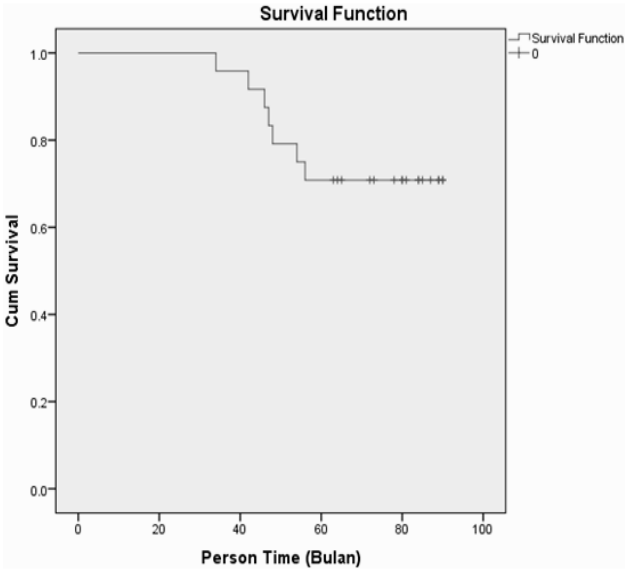

Figure 1. The 5-year Survival Rate of Early Stage Cervical Cancer Patients who had Performed Radical Hysterectomy

In addition, an analysis of the survival rate on some prognostic factors, it was found that 5-year survival rate of patients below 50 years old was 74.8% and the 5-year survival rate in stage II cervical cancer patients was 62.3% (Figure 2). Meanwhile, the 5-year survival rate of patients living with histopathological types of squamous cell carcinomas were 83.2% (Figure 3).

The 5-year survival rate of patients living without lymph node metastasis was 94%, whereas the 5-year survival rate of patients who received adjuvant therapy was 88.9% (Figure 4).

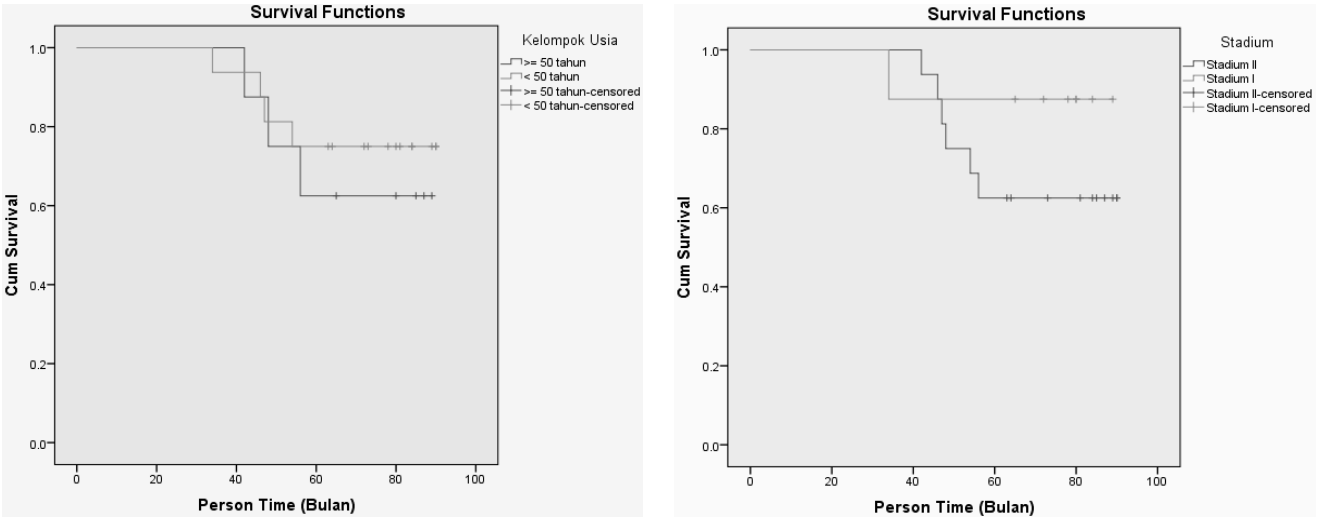

Figure 2. The 5-year Survival Rate Based on Age Group (left) and Stage of Cervical Cancer (right)

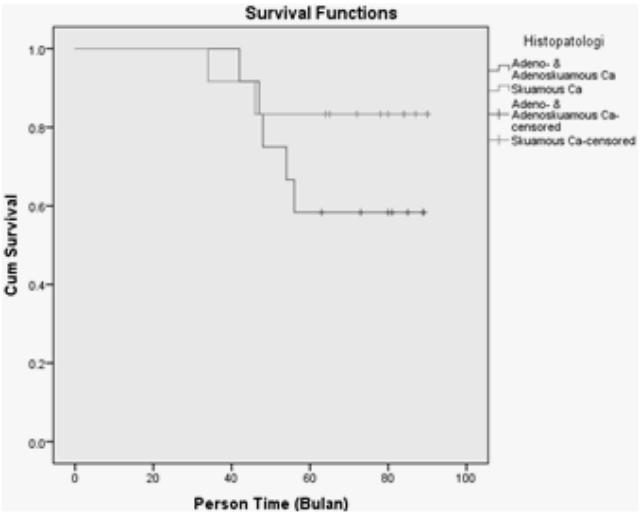

Figure 3. The 5-year Survival Rate Based on Histopathological Type

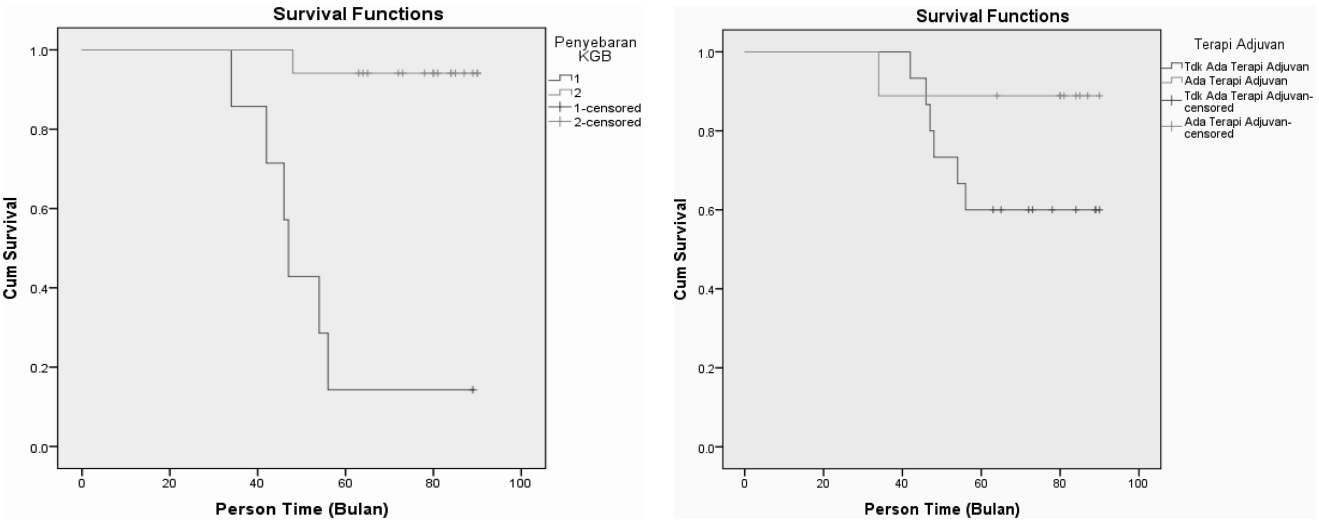

Figure 4. The 5-year Survival Rate Based on Lymph Node Metastasis (left) and Adjuvant Therapy (right)

DISCUSSION

In this study, we analyzed 24 cases of early stage cervical cancer who had performed radical hysterectomy in Prof. dr. R.D. Kandou Hospital Manado on the period of 1 January 2008 to 31 December 2010. The occurrence of information bias in this study was because the design of the study used a retrospective cohort, in which all the relevant events that had occurred at the commencement of the study; for example, the inaccurate data of patients' death remembered by family. Apart from that, there were also several missing cases (loss to follow up). We optimized the observation follow up less than 5 years through several ways, such as appropriate phone number listed in medical records or home visit. This primary data were obtained from medical records; therefore, the incomplete data would affect the overall result.

Overall, of 24 women in this study, there were 7 patients (29.2%) died so that the survival rate was 70.8%. In this study, there were 5 women (20.8%) who regularly undertook the treatment at the hospital for up to 5 years or until the patient dies. Unfortunately, most patients did not have good adherence to the treatment.

The survival rate of early stage cervical cancer patients who had performed radical hysterectomy showed the overall 1-year, 2-year, 3-year, 4-year, and 5-year survival rate were 100.0%, 100.0%, 95.8%, 83.0%, and 70.8%. Actually, the 5-year survival rate in this study was lower than other studies conducted in some developed and developing countries. The reasons for the difference in the survival rate are due to large number of patients who lost to follow-up for 5 years and the divergence among prognostic factors in every center.

In this study, the average age of patients was 47.75 years old with a median of 45 years old. This result was in accordance with study conducted by Ho, et al. They stated that the median age of patients with early stage cervical cancer was 47 years old.[1] Similarly, study held by Kim, et al. explained that the mean age of patients with early stage cervical cancer was 48.5 years old with a range of 22-71 years old.[7] Data from Canada also showed the mean age of 49 years old with the median age of 46 years old.[1]

Based on previous studies, there were relationship between age and survival rate of patients with cervical cancer. However, this study revealed the survival rate difference was found in both age groups.

According to study by Rijker, et al., the 5-year survival rate in patients with stage IB-IIA cervical cancer was not significantly different between age groups, but it had meaningful survival rate difference in patients with advanced stage cervical cancer.[13]

The results showed a tendency early stage cervical cancer patients with age below 50 years old had a 5-year survival higher than cervical cancer patients with age above 50 years old. Indeed, the study conducted by Tone Bjorge, et al. in Norway on 7,429 women with cervical cancer concluded that there was a tendency of worse prognosis in patients with cervical cancer in young age.[14] Atahan, et al. also got significant 5-year survival rate differences ($p = 0.001$) in women with cervical cancer aged less than 40 years old compared to patients with cervical cancer at age more than 40 years old.[15]

In this study, patients with cervical cancer diagnosed with stage IA, IB, and IIA were 4 (16.7%), 4 (16.7%), and 16 (66.7%) patients; contributively. Every region exhibited the various rates of it. Study conducted in Korea showed that patients with stage IB was 79.8% and patients with stage IIA was 20.2% in 2000.[7] Data from Taiwan in 2004 showed that the number of patients with stage IB and IIA were 81.7% and 18.3%.[11] Meanwhile, according to study by Chyong, et al. in 2007, the number of cervical cancer patients stage IB and IIA were 88.1% and 11.9%.[16] This distinction may be due to the differences in the number of samples and low coverage of Pap smear in Indonesia as an early detection of cervical cancer.

The 5-year survival rate of cervical cancer patients with stage I and II in this study were 87.5% and 62.2%; respectively. This finding was similar to other studies.

In this study, the number of cases with histopathological type of squamous cell carcinoma (SCC) were 12 patients (50.0%), adeno-squamous carcinoma were 4 patients (16.7%) and adenocarcinoma were 8 patients (33.3%). The proportion of the cases was opposite with several other studies. Based on the report by FIGO, the majority of cervical cancers are squamous cell carcinoma types histopathology (80%) followed by adenocarcinoma (11%) and adeno-squamous carcinoma (6%).

Based on study in Taiwan on early stage cervical cancer, they obtained the majority of cases were squamous carcinoma (83.2%) and the remaining were 16.8% of adenocarcinoma and adeno-squamous carcinoma.[16] This different distribution may be caused by distinction in the number of samples in this study and the risk factors according to different places and cultures; for example, the number of oral contraception which is used long term.

In this study, the 5-year survival rate of cervical cancer patients with a histopathologic type of squamous cell carcinoma was higher than other types of histopathology. The study by Chen, et al. concluded that the histopathologic type of adenocarcinoma had worse survival rate in patients with stage I and II after radical hysterectomy.[17] The histopathological type of adeno-squamous carcinoma showed lower survival rate than squamous cell carcinoma in the early stage.[18] According to FIGO, this only applies significantly in stage IV, other than the histopathologic type of squamous cell carcinoma and adenocarcinoma, it has worse survival, yet.[19] Contrary, study held by Look, et al. and Lee, et al. did not get the difference in survival rate between squamous cell carcinoma and adenocarcinoma.[20,21] Reis, et al. in their study also concluded that there was no evidence that the type of histopathology would affect the survival rate.[22]

In this study, the number of cases with lymph node metastasis were 7 patients (29.2%) and the number of cases without metastasis were 17 patients (70.8%). There were differences in survival rate of the presence and absence of lymph nodes metastasis in this study. These results were similar to a lot of study that the most dependent variables associated with survival rate was the status of lymph node metastasis. Patients without lymph node metastasis had 5-90% of survival rate, whereas if there were positive lymph node metastasis, the survival rate became 20-74% based on lymph nodes metastasis location and size.[12] Likewise, this study found the 5-year survival rate of cervical cancer patients without lymph node metastasis was 94% differed greatly in patients with lymph node metastasis (14.4%).

In this study, a total of 9 patients with cervical cancer (37.5%) received adjuvant therapy and 15 patients with cervical cancer (62.5%) cases did not get the adjuvant therapy. Giving adjuvant chemoradiation may improve survival rate up to 43 months compared with patients with similar characteristics who receiving radiotherapy adjuvant.[12] According to the study by Lahousen, et al., adjuvant chemotherapy or radiation did not improve survival rate or recurrence rate in patients with high risk of cervical cancer after radical hysterectomy.[23] In this study, the group of patients who received adjuvant therapy had a 5-year survival rate of 88.9%; while, the group of patients who did not get adjuvant therapy was around 60%.

CONCLUSION

The survival rate of early stage cervical cancer after radical hysterectomy in Manado is quite high. Several prognostic factors, such as age, stage of cervical cancer, the type of histopathology, lymph node metastasis and adjuvant therapy affect 5- year survival rate of cervical cancer patients who performing radical hysterectomy.

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Research Article

Phosphatase Regenerating Liver-3 and E-Cadherin Expression in Epithelial Ovarian Cancer

Ekspresi Phosphatase Regenerating Liver-3 dan E-Cadherin pada Kanker Ovarium Epitel

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Abstract

Objective: To determine the expression of Phosphatase Regenerating Liver-3 (PRL-3) and E-Cadherin in the epithelial ovarian cancer on various stages and differentiation grades.

Method: This was a cross-sectional study design conducted at Obstetrics and Gynecology Department of several teaching hospitals, Faculty of Medicine Universitas Hasanuddin from January to June 2015. The expression of PRL-3 and E-cadherin was assessed immunohistochemically in 40 patients with epithelial ovarian cancer including 15 patients in early stage and 25 patients in advanced stage. We used the Fisher's exact test with the significance of $p<0.05$ for the statistical analysis. The Spearman correlation test was used to analyze the correlation between PRL-3 and E-cadherin expression.

Result: Our study showed no statistically significant difference of PRL-3 expression to the stage and the differentiation degree of epithelial ovarian cancer ($p>0.05$). The significant difference was found in the expression of E-cadherin whereas the high expression was shown at early stage than advanced stage ($p<0.05$). There was no significance difference between degree of differentiation and E-cadherin expression ($p>0.05$). This study also pointed out no correlation between the expression of PRL-3 and E-cadherin in epithelial ovarian cancer ($p>0.05$).

Conclusion: PRL-3 overexpression does not decrease E-cadherin expression in epithelial ovarian cancer.

[Indones J Obstet Gynecol 2016; 4-3: 170-175]

Keywords: E-cadherin, epithelial ovarian cancer, PRL-3

Abstrak

***Tujuan**: Menilai ekspresi Phophastase Regenerating Liver-3 (PRL-3) dan E-cadherin dihubungkan dengan stadium dan derajat differensiasi kanker ovarium epitel.*

***Metode**: Penelitian potong lintang yang dilakukan pada beberapa rumah sakit pendidikan pada Departemen Obstetri dan Ginekologi Fakultas Kedokteran Universitas Hasanuddin dari Januari sampai Juni 2015. Penilaian ekspresi PRL-3 dan E-cadherin secara imunohistokimia dilakukan pada 40 pasien kanker ovarium epitel (15 pasien stadium awal, 25 pasien stadium lanjut). Data dianalisis menggunakan uji Fisher's exact dengan tingkat kemaknaan $p<0,05$. Uji korelasi Spearman digunakan untuk menilai korelasi antara ekpresi PRL-3 dan E-cadherin pada kanker ovarium epitel.*

***Hasil**: Penelitian kami menunjukkan tidak ada perbedaan bermakna ekspresi PRL-3 terhadap stadium dan derajat diferensiasi kanker ovarium epitel ($p>0,05$). Perbedaan bermakna ditemukan pada ekspresi E-cadherin di mana ekspresi lebih tinggi pada stadium awal dibandingkan stadium lanjut ($p<0,05$), sedangkan tidak ada perbedaan bermakna antara tingkat diferensiasi dan ekspresi E-cadherin ($p>0,05$). Penelitian ini juga menemukan ekspresi PRL-3 tidak berkorelasi dengan E-cadherin pada kanker ovarium epitel ($p>0,05$).*

***Kesimpulan**: Ekpresi berlebihan PRL-3 tidak menurunkan ekspresi E-cadherin pada kanker ovarium epitel.*

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***Kata kunci**: E-cadherin, kanker ovarium epitel, PRL-3*

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INTRODUCTION

Ovarian cancer is the sixth common disease which contributes to higher death incidence in women worlwide.[1] More than 200.000 women died because of ovarian cancer, especially in women living in low income among developed and developing countries.[2] The highest rate of death is located in sub-Saharan Africa, including South Africa (40/100.000 people). In Africa, most of the patients with ovarian cancer are diagnosed in advanced stage disease (59.3% is diagnosed in stage III). Whereas the decrease of ovarian cancer incidence and mortality is documented in developed countries, such as USA, Canada, and Scandinavia; conversely, this documentation is not completed in developing countries due to the lack of efficiency from the screening programme.[3]

In recent years, there is the development of molecular biology in ovarian carcinoma to discover number of therapeutic targets and new molecular

prognostic factors, such as phosphatase regenerating liver-3 (PRL-3) and E-cadherin. Phosphatase Regenerating Liver-3 is a part of the protein tyrosine phosphatase that has a role in regulating the intracellular growth pathway. Some studies suggest that PRL phosphatase, especially PRL-3, plays important function in growth, proliferation, cell motility and invasion regulation.[4] The PRL-3 also has a role in the process of carcinogenesis. Specifically, PRL-3 is involved in the reconstruction of the cytoskeleton, adhesion and cancer cell cycle regulation, also epithelial-mesenchymal transition. Through this mechanism, PRL-3 is involved in invasion, migration, metastasis, and angiogenesis. Higher expression of PRL-3 is associated with tumor progression and severity of the disease. The over expression of PRL-3 is varied based on the type of tumor; many studies showed that tumor metastases both in lymph nodes and other organs would increase the PRL-3 expression extremely.[5]

E-cadherin (E-cad), the cell adhesion molecule plays an important role in maintaining tissue integrity. Impaired function of E-cadherin has often been associated with tumor formation and invasion in vivo and in vitro.[6] E-cadherin is expressed on epithelial cells and it is important for epithelial cells differentiation and cells adhesion during embryogenesis.[7] Abnormal expression of E-cadherin is associated with higher invasiveness and poor differentiation from several types of epithelial carcinomas.[8]

According to the theory above, we have not ever reported the association between PRL-3 and E-cadherin in epithelial ovarian cancer in Indonesia, especially Makassar. Therefore, this study aims to determine the role of PRL-3 and E-cadherin in epithelial ovarian cancer progressivity. Further more, we would like to assess the relationship between the expression of PRL-3 and E-cadherin through clinicopathological factors, as potential target for epithelial ovarian cancer therapy.

METHODS

This cross-sectional study was conducted from January to June 2015. Tumor tissues were collected from 40 patients who had been undergone surgery for epithelial ovarian carcinoma at Obstetrics and Gynecology Department of several teaching hospitals, Faculty of Medicine Universitas Hasanuddin. The analysis of immunohistochemical was performed at Anatomical Pathology Laboratory, Department of Anatomical Pathology Faculty of Medicine Universitas Hasanuddin and Pathology Laboratory Dr. Wahidin Sudirohusodo Hospital Makassar. We got the written informed consent from the patients before performing this study.

Tumor tissues taken during surgery were fixed in buffered and paraffin-embedded formalin. The tissue blocks were cut using a microtome into 4-μm-thickness sections on silanized glasses. After that, the tissue sections were immersed in hot water at 40°C and dried on hot plate at 60°C for 1 hour. We would store the sections at room temperature.

We performed the immunohistochemical analysis using streptavidin-biotin-peroxidase labeled streptavidin-biotin (Dako, Carpinteria, USA). Before staining, the tissue sections were deparaffinized in xylenes for 15 minutes. Then, the tissue sections were washed with dH_2O two times for 5 minutes each and incubated with Phosphate-buffered saline (PBS) for 5 minutes. In order to exhibit antigen, the tissue sections were heated in an autoclave for 15 minutes in citrate buffer. The tissue sections were cooled at room temperature for 1 hour and after drying, they were demarcated using a pap pen. After that, we repeated the washing with dH_2O for 5 minutes and PBS for 5 minutes before incubating them into 0.3% hydrogen peroxidase for 15 minutes. After endogenous peroxidase blocked, the tissue sections were incubated with blocking solution for 30 minutes to block avidin contained in the sections. The tissue sections were incubated overnight at -4°C with primary PRL-3 antibody (Santa Cruz) diluted 1 : 100 and E-cadherin monoclonal antibody (Biocare) diluted 1 : 100. The tissue sections would be washed three times with dH_2O before incubating with secondary antibodies and streptavidin for 30 minutes. The 3,3'-diamino-benzidine tetrahydrocloride was used for staining approximately 10 minutes to obtain the staining reaction that could be detected by microscopic examination. Subsequently, the sections were stained with hematoxylin eosin to clarify the nucleus of a cell for 30 seconds and they were washed with running water for 5 minutes. The tissue sections would be dehydrated in alcohol gradually from 70%, 80%, 90% to 100% for every 2 minutes. Then, the sections were dipped in xylene for 5 minutes and covered with cover slip after malinol application. The staining degree of PRL-3 and E-cadherin expression could be seen

from the percentage of cell group stained. The percentage was obtained from the sum of positive cells in the entire field of tissue sections view examination using a light microscope.

Immunohistochemical scoring system for protein E-cadherin and PRL on epithelial ovarian cancer uses three parameters consisting of intensity of expression, the percentage of positive cells, and expression patterns. The expression of E-cadherin is as follows: grade 1 = score 1-3; grade 2 = score 4-6; grade 3 = score 7-9; grade 4 = score ≥10. The grade 1 and 2 are classified as low level of expression; while, grade 3 and 4 are classified as high level of expression.

The Fisher's exact test was used to analyze the correlation between PRL-3 and E-cadherin expression with clinicopathological parameters. The correlation of PRL-3 and E-cadherin expression level with epithelial ovarian cancer was analyzed with Pearson's test. We used $p<0.05$ as statistically significant. Data were analyzed using SPSS for windows version 20.

RESULTS

We included 40 women who experienced epithelial ovarian cancer. The youngest age was 24 and 59 years old for the oldest patients. Most of the patients were between 31-40 years old (35%) and 41-50 years old (35%). Most of them were jobless (65%) and they had 1 to 4 children. In this study, 15 women were at early stage (37.5%) and 25 women (62.5%) were at advanced stage of the disease. Poor differentiation degree was mostly found in 47.5% patients (Table 1).

Table 1. Subject Characteristics

Characteristics	n	%
Age (years old)		
≤ 30	2	5.0
31-40	14	35.0
41-50	14	35.0
>60	10	25.0
Occupation		
Jobless	26	65.0
Governmental officer	7	17.5
Private officer	7	17.5
Parity		
Nulliparous	13	32.5
1-4	21	52.5
≥5	6	15.0
Stages		
Early	15	37.5
Advanced	25	62.5
Differentiation degree		
Good	11	27.5
Mild	10	25.0
Poor	9	47.5

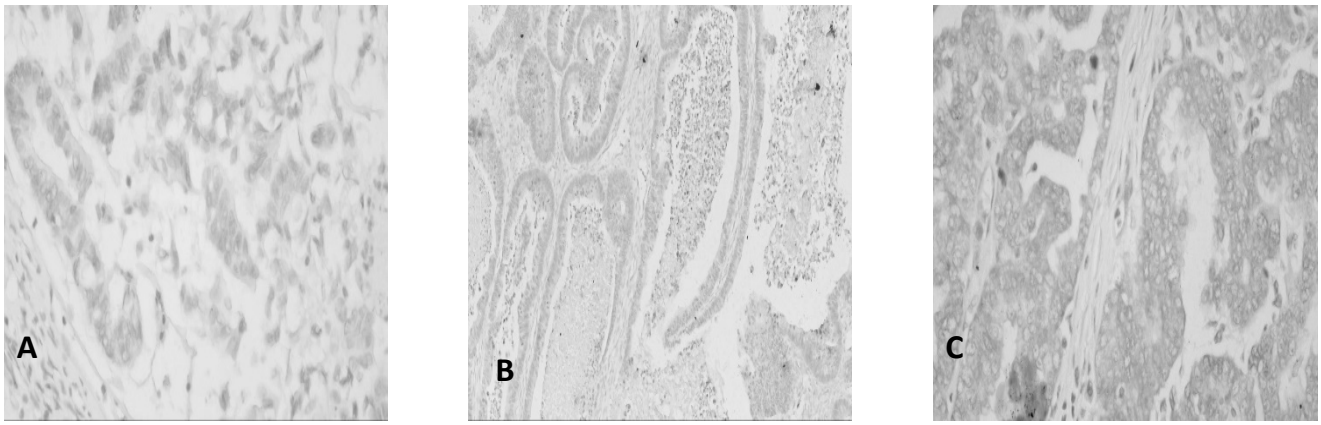

Figure 1. Immunohistochemical Staining of PRL-3 Expression in Epithelial Ovarian Cancer.
A = Grade I, **B** = Grade II, **C** = Grade III.

Figure 1 showed the expression of PRL-3 in epithelial ovarian cancer using immunohistochemical staining. The PRL-3 expression at various stages and degrees of differentiation showed no significant difference between the expression of PRL-3 at early stage compared to advanced stage. Apart from that, it pointed out the similar result to various degrees of epithelial ovarian cancer differentiation (p> 0.05) (Table 2).

The expression of E-cadherin in epithelial ovarian cancer using immunohistochemical staining was showed in Figure 2. E-cadherin expression at various stages and degrees of differentiation was found significantly different. High expression of E-cadherin was observed in early stage; while, low expression was seen in advanced stage (p<0.05). Conversely, there was no significant difference found between E-cadherin expression and degree of differentiation (p>0.05) (Table 2).

Table 3. Correlation PRL-3 and E-cadherin

	E-cadherin
PRL-3	rs = 0.161*
	p = 0.320*

**Spearman correlation test*

The statistical analysis showed no significant association between the expression of PRL-3 and E-cadherin in epithelial ovarian cancer (p>0.05). Therefore, it could be inferred that the expression of PRL-3 was not associated with the decreased expression (down-regulation) of E-cadherin in epithelial ovarian cancer.

DISCUSSION

This study showed the PRL-3 expression on the primary tumor tissue from 40 samples of epithelial ovarian cancer using the immunohistochemical

Table 2. PRL-3 and E-cadherin Expression on Stage and Differentiation Degree of Epithelial Ovarian Cancer

Variables	PRL-3		*p-value**	E-cadherin		*p-value**
	Low	High		Low	High	
Stage						
Early	13 (86.7)	2 (13.3)	1.000	2 (13.3)	13 (86.7)	<0.001
Advanced	21 (84.0)	4 (16.0)		19 (76.0)	6 (24.0)	
Differentiation degree						
Good	9 (81.8)	2 (18.2)	1.000	5 (45.5)	6 (54.5)	0.727
Poor	25 (86.2)	4 (13.8)		16 (55.2)	13 (44.8)	

**Fisher's Exact*

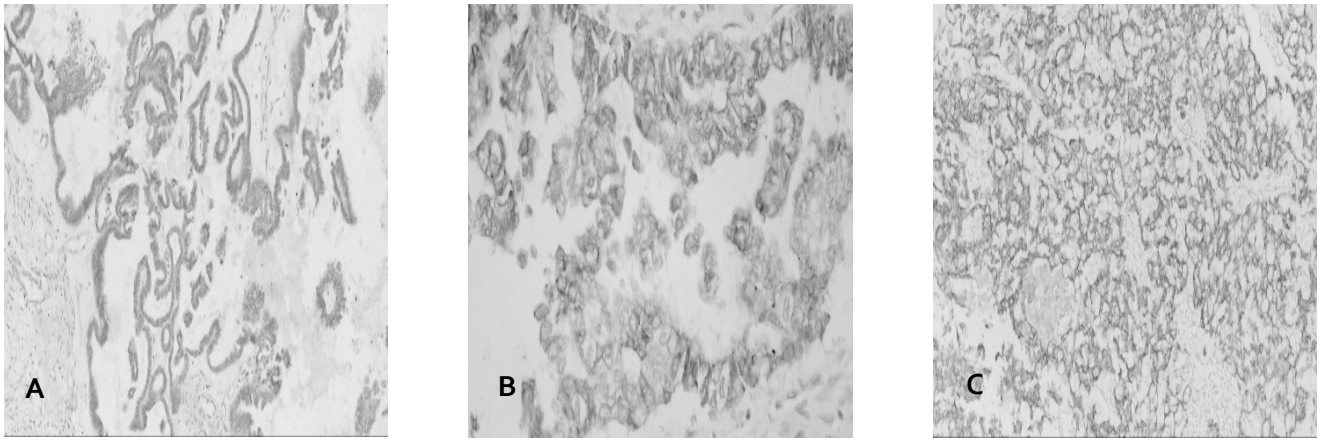

Figure 2. Immunohistochemical Staining of E-cadherin Expression in Epithelial Ovarian Cancer.
A = Grade I, **B** = Grade II, **C** = Grade III.

method. Positive expression of PRL-3 was found in 34 samples of primary ovarian tumors. This study found no significant association between PRL-3 expression in primary tumor and epithelial ovarian cancer stage. The similar result reported by Reich, et al. stated that there was no significant difference between the expression of PRL-3 and ovarian cancer stage.[9] In contrast, Polato, et al. found PRL-3 expression was high at an advanced stage than early stage.[10] It could be due to the sample size which involved 61 samples in stage I and 23 samples in stage III. Meanwhile, Reich, et al. study indicated fewer sample for early stage than advanced stage.

The PRL-3 expression was not significantly different in degrees of differentiation. Similar finding was reported by Reich, et al. and Polato, et al. studies which stated that there was no association between the expression of PRL-3 and degree of differentiation. Interestingly, Liu, et al. found up-regulation of PRL-3 ovarian cancer only occurred in differentiated cancer cells.[11]

E-cadherin plays a role in maintaining cell adhesion to prevent tumor cell invasion. E-cadherin mediated intercelullar adhesion that connects to cytoskeleton and cathenin as an intracellular binding protein. Low expression of E-cadherin will lead to detach tumor cells and attach to the matrix component resulting in migration and metastasis. E-cadherin tumor suppression gene has been widely studied in tumorigenesis. The interaction between E-cadherin molecules is crucial to the formation of adhesion among cells and maintain the cells adhesion. The loss of E-cadherin is associated with the transition from benign lesions to the invasive and metastatic lesions.[12]

In this study, the protein E-cadherin was expressed in all primary epithelial ovarian cancer. Although, there was no significant difference of E-cadherin expression in various degrees of differentiation, there were statistically significant difference of the E-cadherin expression in epithelial ovarian cancer stage. High expression of E-cadherin was found in early stage than advanced stage. Study by Faleiro-Rodrigues, et al. revealed a significant decreased expression of E-cadherin based on FIGO staging and they showed no correlation to tumor differentiation.[13] In another study on the expression of E-cadherin and its relationship with ovarian cancer showed that a decreased expression of E-cadherin was associated with the severity of ovarian cancer based on FIGO staging, lymph node metastasis, and degree of tumor differentiation. This result would like to reveal that the expression of E-cadherin tended to decrease with the increasing of stage disease and poorly differentiated cancer cells.[14]

This is the first study that reported association between the expression of PRL-3 and E-cadherin. We revealed that the increased levels of PRL-3 did not affect the levels of E-cadherin and also vice versa. Our result was contrary with the result by Liu, et al. Liu found that the PRL-3 increased the epithelial mesenchymal transition (EMT) by down-regulated expression of E-cadherin. The EMT enhancement mechanism caused by PRL-3 was still unknown. Liu, et al. found that PRL-3 might improve the EMT in colorectal cancer cells SW480 models deficient in the expression of E-cadherin in vivo and in vitro. The overexpression of PRL-3 or SW480 cell solution was injected subcutaneously in nude mice. The result of immunohistochemical analysis of tumor samples from nude mice showed that the PRL-3 increased the up-regulation of mesenchymal marker vimentin and down-regulation epithelial marker of E-cadherin and cytokeratin.[11] Guo, et al. study on the ability of PRL-3 triggering tumor angiogenesis reported that subcutaneous injection of PRL-3 in nude mice led to increase formation of endothelial cells in the tumor mass.[5] In contrast, Sundfeldt, et al. revealed that the down-regulation or abnormal expression of E-cadherin was associated with highly invasive capacity and poorly differentiated epithelial ovarian carcinoma.[6]

CONCLUSION

Our study has shown that E-cadherin plays an important role in the progression of epithelial ovarian cancer. This suggests that E-cadherin might have therapeutic value and a potential biomarker in the progression of ovarian cancer. The PRL-3 and E-cadherin also plays a role in ovarian cancer metastasis. Therefore, we need to investigate the role of PRL-3 and E-cadherin in the process of ovarian cancer metastases for future studies.

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