International Journal of ChemTech Research
CODEN (USA): IJCRGG, ISSN: 0974-4290, ISSN(Online):2455-9555
Vol.10 No.9, pp 963-973, 2017

Comparison between cystatin C level and renal artery flow profile in normal pregnancy and preeclampsia

Cut Meurah Yeni[1]*, Noroyono Wibowo[2], Azen Salim[2], Prima Nanda Fauziah[3], Ani Melani Maskoen[4], Johannes Mose[5], Rovina Ruslami[6]

[1]Department of Obstetric and Gynecology, Faculty of Medicine, Universitas Syiah Kuala, Banda Aceh, Indonesia
[2]Department of Obstetric and Gynecology, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia
[3]Department of Medical Laboratory Technology, School of Health Sciences Jenderal Achmad Yani, Cimahi, Indonesia
[4]Department of Oral Biology, Faculty of Dentistry, Universitas Padjadjaran, Bandung, Indonesia
[5]Department of Obstetric and Gynecology, Faculty of Medicine, Universitas Padjadjaran, Bandung, Indonesia
[6]Department of Pharmacology and Therapy, Faculty of Medicine, Universitas Padjadjaran, Bandung, Indonesia

Abstract : Preeclampsia is a multisystem disease occured during pregnancy as indicated by endothelial dysfunction. During pregnancy, cystatin C level elevate in serum, mainly in preeclamptic pregnancy, which is associated with structural and functional alteration in kidney, which can be utilized as marker of transition from normal pregnancy to preeclampsia and its severity degree. This study aimed to observe the correlation between cystatin C level and profile of renal artery flow in normal pregnancy and preeclampsia. **Methods**: Samples were carried with consecutive sampling from normal patients at second and third trimester and pregnant patients with preeclampsia administred in *Poliklinik Kebidanan,* Dr. Zainoel Abidin Hospital, Banda Aceh, in February– May 2009. Measurement of doppler renal artery was performed with USG type Aloka 3500 Pro 3.5 MHz-5 MHz tranducer. After kindey and renal artery were identified, SD ratio, RI and PI were measured. **Results**: There was significant difference in cystatin C level between noremal group and preeclampsia. Cystatin level was higher in preeclampsia group compared to normal group. Renal artery flow was significant only in left PI. Cystatin and renal artery in preeclampsia showed left PI was strongly correlated. **Conclusion**: There was correlation between left PI and cystatin C in preclampsia in which the higher cystatin C, the higher left PI.
Keywords : preeclampsia, cystatin C, renal artery.

Introduction

Preeclampsia is a multisystem disease specifically occured during pregnancy as indicated by endothelial dysfunction due to inflammation. It has been a major cause in maternal and fetal morbidity and mortality as indicated by hypertension, proteinuria and broad ischemic vascularization [1,2]. Incidence of preeclampsia is

commonly 5-7 % of preeclampsia, although widely differ in parity, racial/genetic predisposition and environmetal factors. Survey carried in Indonesia in 1995 shows that number of maternal mortality due to eclampsia was 13%. Preeclampsia causes 16 % maternal mortality and 45 % perinatal mortality both direct or indirectly [3].

Clinically, preeclampsia is a manifestation of many pathological pathways. Further studies regarding its complex pathophysiology is encouraged for early detection, medication and prevention [3-5]. Cystatin C is common in human tissues such as kidney, liver, pancreas, colon, stomach, lung, and placenta in which cystatin C manifest in blood circulation both in normal pregnancy or preeclampsia [6-8]. Cystatin C is a strong extracellular inhibitor from cystein protease that possess regulatory effect on other protease [9]. Cystatin C has small molecular weight (13.3 kDa) in which its level represent glomerular filtration rate (GFR) in women [10]. During pregnancy, cystatin C level elevate in serum, mainly in preeclamptic pregnancy, which is associated with structural and functional alteration in kidney [11,12]. Therefore, cystatin C level in serum can be utilized as marker of transition from normal pregnancy to preeclampsia and its severity degree [12]. Elevated cystatin C in serum of pregnancy and preeclampsia is caused by change in regulation in kidney as well as increased protein synthesis [12,13]

Level of cystatin C in maternal plasma is a good indicator to early determine its severity. Study on renal biopsy found that cystatin level is correlated with alteration degree in distict renal structure (glomerular endotheliosis) in preeclampsia. Level of cystatin C is also correlated with GFR in normal pregnancy dan preeclampsia as well as further pregnancy [14,15].

In normal pregnancy, there is hyperfiltration in renal glomerulus which elevate about 40-60 %. Hiperfiltration occurs due to decreased plasma oncotic tension plasma glomerulal capillary. Decreased oncotic pressure occurs due to two factors, hypervolemia due to hemodilution that reduce plasma protein level in glomerular microcirculation. Secondly, increased renal plasma flow (RPF). Moreover, renal size increase to 1 cm during pregnancy, whilst LFG in preeclampsia decreased from 149 mL/mnt 91 mL/mnt [15].

Experimental

Samples were carried with consecutive sampling from normal patients at second and third trimester and pregnant patients with preeclampsia administred in *Poliklinik Kebidanan,* Dr. Zainoel Abidin Hospital, Banda Aceh, in February 2009 – May 2009. Selection was in accordance with criteria including anamnesis, physical (general and obstetrics status) and laboratory test. Informed consent was obtained.

A 5 cc of vein blood was carried, and cystatin C was measured with Immunonephelometry. This technique used blood samples of 0,5 mL serum, heparin using reagen N latex cystatin C and Behring Nephelometer. Measurement of doppler renal artery was performed with USG type Aloka 3500 Pro 3.5 MHz-5 MHz tranducer. Subjects in left lateral decubitus held breath during sampling to display better profile. After kindey and renal artery were identified, SD ratio, RI and PI were measured .

Data analysis

Data were analyzed with SPSS 13.0. After fulfilling parametric condition, unpaired T-test was performed to compare value between two groups. If not, Man-Whutney test was then conducted. Correlation between cystatin C and arterial renalis flow profile was analyzed with Pearson test (in accordance to parametric) and Spearman (if not in accordance to parametric). Linear regresion was conducted between factors correlated to cystatin C.

Result

This study involved 64 respondents consisting of 32 both in normal pregnancy and preeclampsia. Respondents were predominently housewives (81,25%) and high school graduate (62,5%). During test, gestational age varied in range of 26-41 weeks iwth highest number at > 36 weeks (34,2% in preeclampsia and 25 % in normal pregnancy). Gestational age between normal pregnancy and preeclampsia showed no significant difference ($p > 0,05$).

There were no preclampsia history recorded from all respondents. As shown in Table 2 and 3, patients age average was 29 years old in range of 18-50 years old. Weight mass index was 28,28 in range of 18,47 – 42,97. Estimated fetal weight which found inhibited in in preeclampsia, was 56,3 % and significantly different compared to normal pregnancy (3,1 %), $p < 0.001$.

Table 1. Subject characteristics and comparison between groups

		GROUPS				Total		Equality test
		Preeclampsia		Normal				
		N	%	N	%	N	%	
Job								p>0,05
	Midwives	1	1.6%	0		1	1.56%	
	IRT	28	40.8%	24	37.6%	52	81.3%	
	Public	0		4	6.3%	4	6.2%	
	Private	3	4.7%	4	6.3%	7	10.9%	
Education								p>0,05
	Elementary school	3	4.7%	0	.0%	3	4.7%	
	Middle School	6	9.4%	4	6.3%	10	15.6%	
	High school	20	31.3%	20	31.3%	40	62.5%	
	Diploma	3	4.7%	3	4.7%	6	9.4%	
	Bachelor degree	0	.0%	5	7.8%	5	7.8%	
Gestational age (Weeks)								p>0,05
	< 32 mg	4	6.3%	11	17.2%	15	23.4%	
	32-36 mg	6	9.4%	5	7.8%	11	17.2%	
	> 36 mg	22	34.2%	17	25.00%	38	59,3%	
PJT/IUGR								p < 0,001
	Yes	18	56,3%	1	3,1%	19	19,7%	
	No	14	43,8%	31	96,9%	45	70,3%	

Table 2. Subject characteristics in accordance with maternal physic, fetal biometry, arterial renalis flow and cystatin C (n=64)

Characteristic	Median and distribution
Maternal physic	
Age	29 (18-50)
Weight mass (Kg)	68,73±12,38
Height (cm)	156±5
IMT	28,28±4,58
Fetal biometry	
BPD (mm)	87±7
AC (mm)	286±24
FL (mm)	67±6
Arterial renalis flow	
Right SD	2,57±0,48
Right RI	0,63±0,08
Right PI	1,11±0,32
Left SD	2,67±0,68
Left RI	0,65 (0,49-1,56)
Left PI	1,18 (0,73-3,01)
Cystatin	1,08 (0,62-2,25)

Table 3. Comparison between weight mass, cystatin C, and USG Doppler arterial renalis between preeclampsia and normal pregnancy

Characteristic	Groups					
	Preeclampsia			Normal		
	Med	Min	Max	Med	Min	Max
Age	30	20	50	27	18	36
Weight (Kg)	71.00	57.00	110.00	65.00	48.00	88.00
Height (cm)	156	145	170	156	145	164
IMT	29.12	25.08	42.97	25.34	19.47	35.00
BPD (mm)	90	74	97	87	67	96
AC (mm)	288	220	336	288	232	337
FL (mm)	70	55	76	66	50	76
Right SD	2.51	.98	4.22	2.50	2.25	2.90
Right RI	.64	.47	.85	.61	.55	.85
Right PI	1.06	.70	2.46	1.01	.88	1.15
Left SD	2.56	1.77	5.29	2.47	2.25	4.06
Left RI	.63	.49	1.56	.61	.50	.75
Left PI	1.10	.73	3.01	.99	.76	1.49
Cystatin	1.30	.89	2.25	.78	.62	.96

Table 4. Comparison between Cystatin C and arterial renalis between preeclampsia and normal prgenancy

	Groups										Mann Whitney Test
	Preeclampsia					Normal					
	Mean	Std	Med	Min	Max	Mean	Std	Med	Min	Max	
Right SD	2.58	.65	2.51	.98	4.22	2.55	.20	2.50	2.25	2.90	0,946
Right RI	.63	.10	.64	.47	.85	.62	.05	.61	.55	.85	0,544
Right PI	1.19	.44	1.06	.70	2.46	1.03	.08	1.01	.88	1.15	0,435
Left SD	2.75	.90	2.56	1.77	5.29	2.60	.34	2.47	2.25	4.06	0,643
Left RI	.69	.24	.63	.49	1.56	.61	.05	.61	.50	.75	0,216
Left PI	1.34	.55	1.10	.73	3.01	1.02	.14	.99	.76	1.49	0,042
Cystatin	1.36	.33	1.30	.89	2.25	.79	.10	.78	.62	.96	< 0,001

Bivariat Analysis

a. Comparison between Cystatin C level and arterial renalis flow in normal pregnancy and preeclampsia serum

Data between two groups were not normally distributed, thus requires non-parametric Mann Whitney test. Results showed Cystatin C and left PI showed significant difference between preeclampsia and normal pregnancy. Cystatin C and left PI was way higher than in preeclampsia than that in normal pregnancy. However, right PI and left/right SD arterial renalis showed no significant difference (Table 5 & 6)

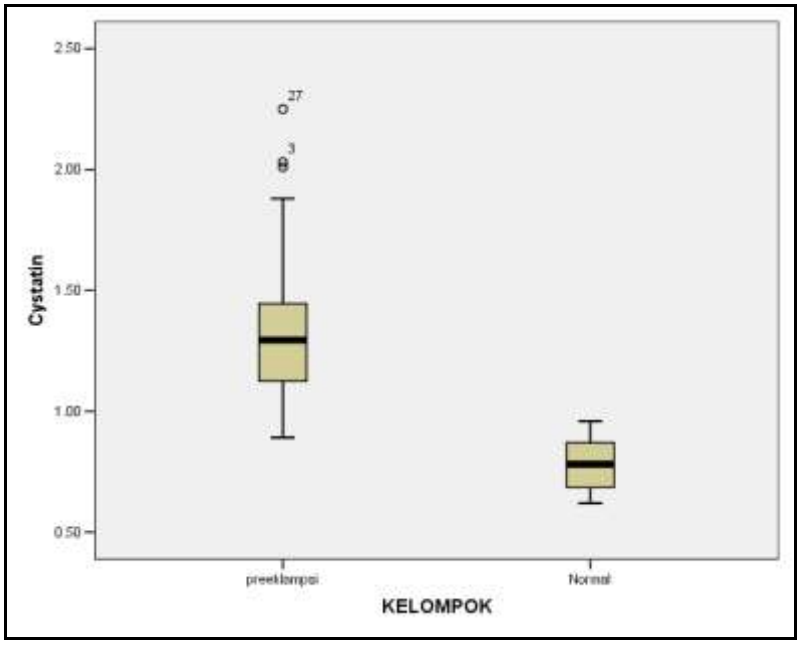

Figure 5 Boxplot of Cystatin between preeclampsia and normal pregnancy

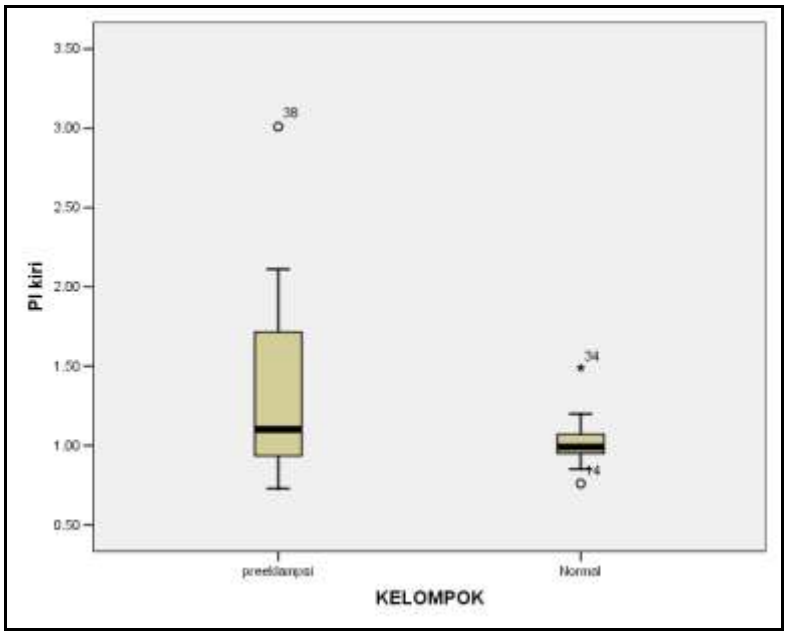

Figure 6. Boxplot of left PI between preeclampsia and normal pregnancy

b. Correlation between cystatin C and arterial renalis SD/PI/RI in preeclampsia

Correlation between cystatin C and arterial renalis flow was significantly different only in left PI which was Spearman test (p=0,016, r=0,422). However, these results was not in accordance with right RI which was not significantly different. Parameters such as SD, right PI, left RI) was not also significantly correlated (Fig 7-12).

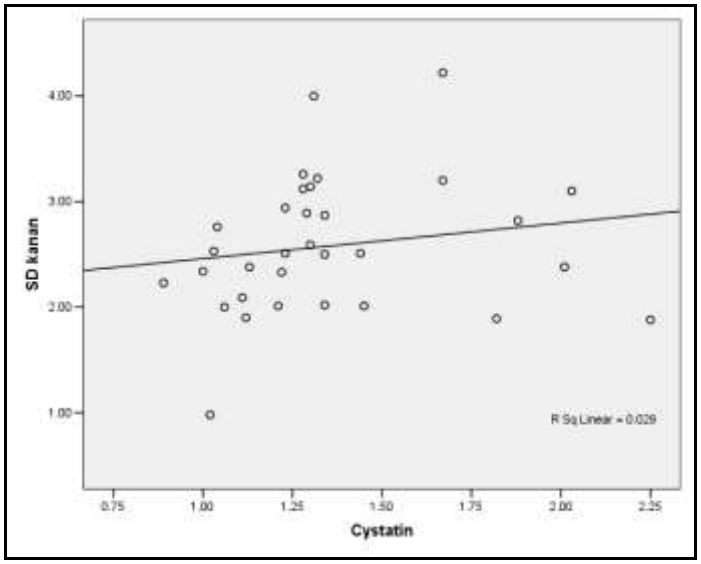

Figure 7. Scatter Plot of right SD with cystatin C with Spearman-correlation test (p=0,178 dan r=0,244)

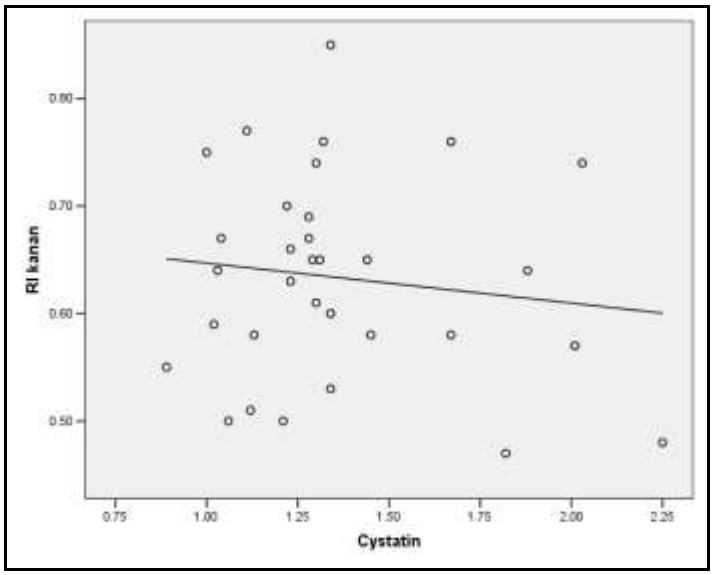

Figure 8. Scatter Plot of right RI and cystatin C with Spearman test (p=0,747 dan r=-0,059)

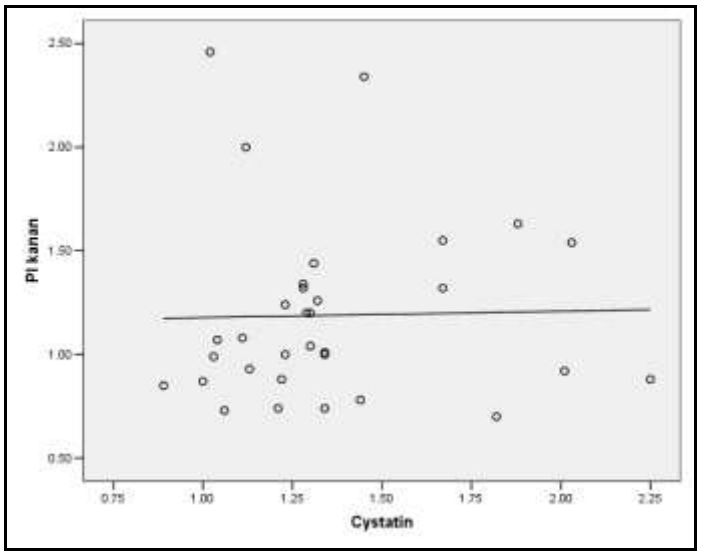

Figure 9. Scatter Plot of right PI and Cystatin with Spearman test (p=0,439 dan r=-0,142)

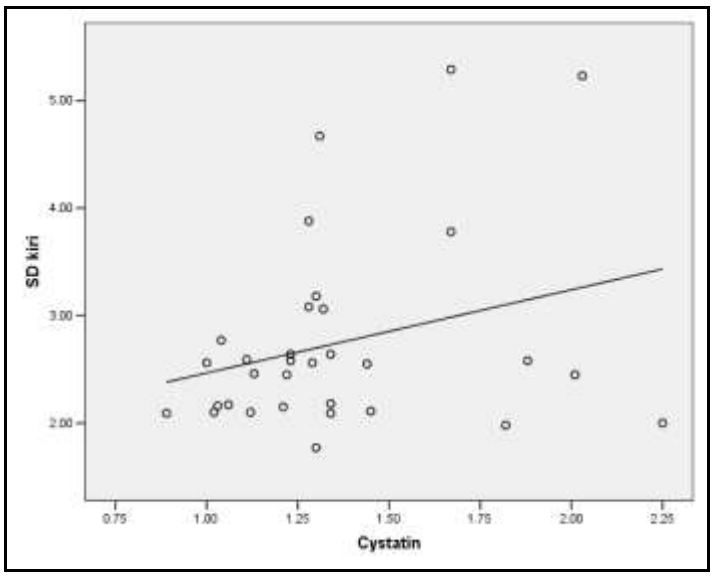

Figure 10. Scatter Plot of left SD and Cystatin with Spearman test (p=0,396 dan r=0,155)

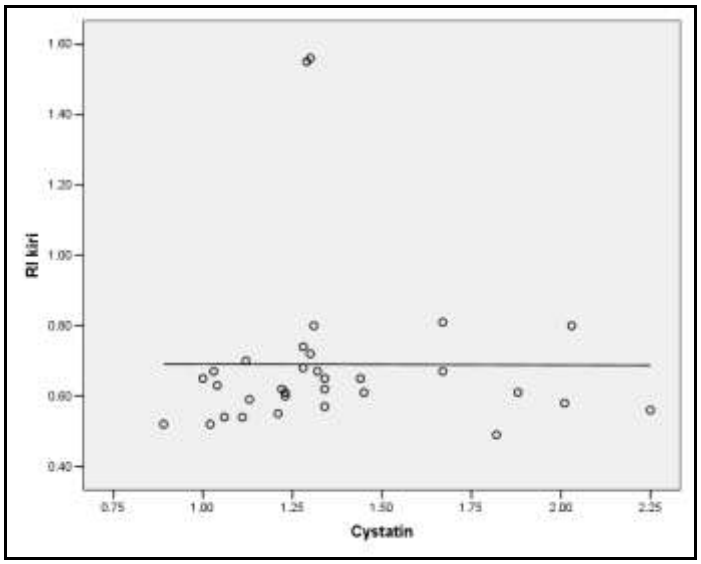

Figure 11. Scatter Plot of left RI and Cystatin with Spearman test (p=0,318 dan r=-0,182)

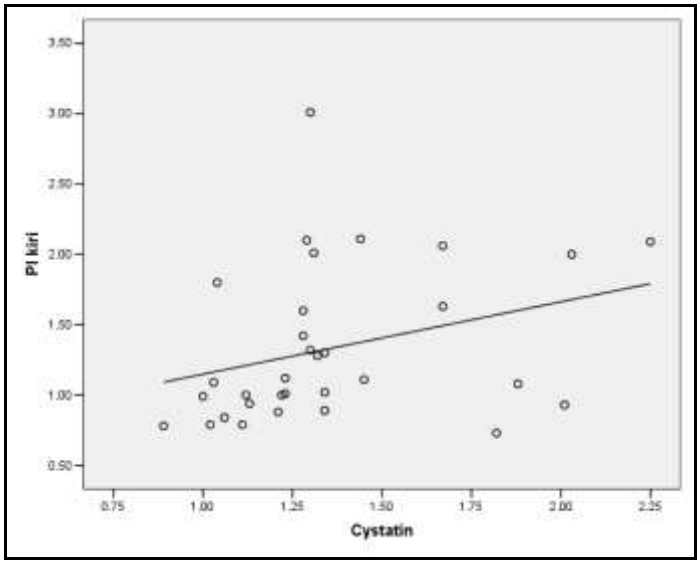

Figure 12. Scatter Plot of left PI and Cystatin with Spearman test (p=0,016 dan r=0,422)

Linear Regression

Results showed linear regression was sistatin = 0,81 + 0,24(PI Kiri), with accuracy (R Square) 4%. Linear regression performed separately in preeclampsia and normal will lower accuracy which was 0-3% (Fig. 13).

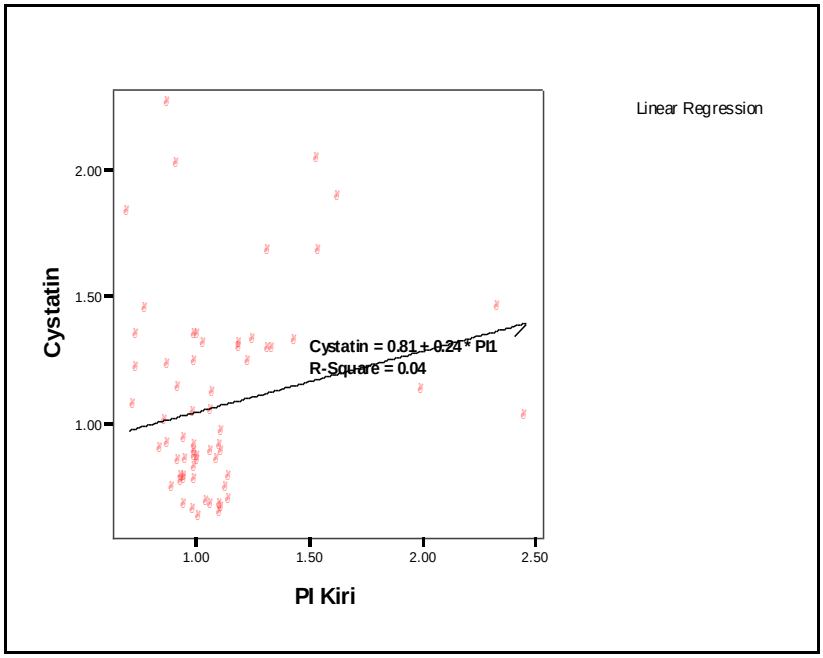

Figure 13. Scatter Plot of left PI and cystatin C with linear regression

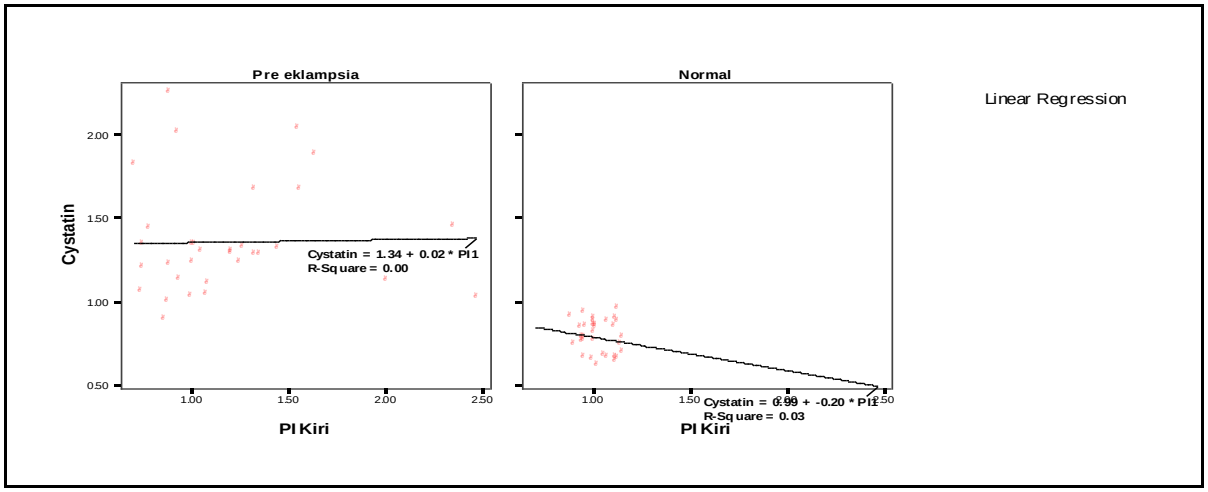

Figure 14. Scatter Plot of left PI and cystatin C with linear regression in preeclampsia and normal pregnancy

Discussion

Pregnant women that underwent the test were predominently 29 years old. This is in accordance with *Survey Kesehatan Nasional* that healthy reproduction age is 20-35 years old. Pregnant women above 40 years old, or even 50 years old. Respondents were mostly high school graduate and housewives. There was no significant difference in age, eduaction, job, gestational age, and nutritional status between normal pregnancy and severe preeclampsia.

There was significant difference in cystatin C level between normal group and preeclampsia. Cystatin level was higher in preeclampsia group compared to normal group. These findings are in accordance with previous studies [11-13]. Several studies showed increase in cystatin level in normal pregnancy that resulted 0.79 mg/L (normal cystatin C in women is 0.52 – 0.98 mg/L) [12-14]. High cystatin C level in preeclampsia is correlated with renal structural and functional alteration [11,12] as well as increase in protein regulation [12,13]

The result of present study showed that renal artery flow was significant in left PI. This is in accordance with study done by Kurjak et al that found significant difference yet it was only on arterial resistivity index among normal pregnancy [40]. According to Khyse-Anderson, Rosenthal and Mares, renal artery flow reduced in preeclampsia yet it remains unclear which parameter that reduced [31-33]. In this study, significant increased PI in severe preeclampsia occurred due to slow renal artery flow. Resistivity index also increased yet it was not significant. This is due to in preeclamptic patients in this study did not undergo renal damage (renal endothelial).

Cystatin and renal artery in preeclampsia showed left PI was strogly correlated. For our best knowledge, there are no studies regarding this. Thus, measurement of left PI can be used as predictor of cystatin C qualitatively in preeclampsia.

Conclusion

Cystatin C was significantly higher in preeclampsia compared to normal. Left PI was higher in preeclampsia. There was correlation between left PI and cystatin C in preecclampsia in which the higher cystatin C, the higher left PI.

Acknowledgment

This research was funded by Internal Grant from Universitas Syiah Kuala, Banda Aceh.

Interest conflict

Authors declare there is no conflict of interest

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