CHAPTER 1

INTRODUCTION

1.1 Background

PKD is one of factor which cause of End Stage Renal Disease (ESRD).[1,2] PKD has been contributed to 2.5% ESRD in Indonesia.[2] PKD is genetic disorder which cause numerous kidney cystic lesions.[3] The most common observed PKD is Autosomal Dominant Polycystic Kidney Disease (ADPKD).[4-6] ADPKD has been also considered to cause 2.5% ESRD in Japan, United States and Europe.[4] However, the genetic base of PKD in Indonesia is unknown.

The prevalence of ADPKD is 1 in 400-1000 live births.[4, 7, 8] It is multiracial disease and affected people worldwide.[9] The characteristics of ADPKD are massive cystic expansion and progressive renal enlargement lead to renal failure.[1] ADPKD is also the most common genetic disease which causes Chronic Kidney Disease (CKD).[10] Although its prevalence among another causal factor of ESRD is low, individual with ADPKD will be suffered from renal failure in 5^{th} to 6^{th} decade.[1]

The most prevalence mutation in ADPKD is *PKD1* gene mutation (85%).[5,7] *PKD1* gene is located on chromosome 16p13.3. Mutations in *PKD1* gene can be found at entire of the gene. The pathogenic mutations are out-of-frame deletions/insertions and nonsense mutations.[11]

The mutation data for APDKD is provided by The Polycystic Kidney Disease Mutation database. The variants of *PKD1* up to February 2011 were reported to be 864 changes. The mutations are 50.4% (436) pathogenic (including 35.6% definitely pathogenic, 11.3% highly likely pathogenic and 3.5% likely pathogenic), 0.5% (4) are hypomorphic, 7.8% (67) are indeterminate and 41.3% (357) are neutral. The polymorphism is accounted for 424 of 864 changes in *PKD1*. It suggests highly polymorphic and high new mutation rate in *PKD1* gene.[12]

qPCR High Resolution Melting (HRM) is molecular technique which fast and sensitive in mutation detection.[13] The purpose of this technique is to achieve fast and cost effective mutation detection.[14] The molecular analysis should be considered to establish a definite diagnosis especially in vague imaging results, negative family history, or in young potential individual who will be a kidney donor from patient's family.[15] Furthermore, it will be useful in genetic counseling process to patients and family. The aims of this research are giving information about family with polycystic kidney disease and the genetic mutation analysis.

1.2 Research Question

Is there pathogenic mutation of *PKD1* gene in patients and their families with suspected familial polycystic kidney disease?

1.3 Research Objectives

1.3.1 General Objectives

- To identify the pathogenic mutation of *PKD1* gene in patients and their families with suspected familial polycystic kidney disease.

1.3.2 Specific Objectives

- To identify the genotype and phenotype correlation in patients and their families with suspected familial PKD.
- To identify the benefit of qPCR-HRM technique in molecular diagnosis of ADPKD.

1.4 Research Advantages

- To provide early diagnosis in PKD patients and families with suspected familial PKD in efforts to prevent from renal failure.
- To provide fast and achievable molecular technique to establish definitive diagnosis of ADPKD.
- To provide data which help the genetic counselling process in PKD patients and families.
- As a preliminary study in Indonesia about molecular identification in familial PKD.

1.5 Originality

There were two publications about application of PCR HRM in mutation screening of ADPKD. This study applies the usage of qPCR-HRM technique for *PKD1* gene mutation detection in PKD patient. The potency

of fast and cost effectiveness of PCR HRM is promising to reduce the dependence of whole sequencing in mutation screening of ADPKD.[14]

Table 1. Research originality

No	Publications	Methods	Result
1	Bataille S, Berland Y, Fontes M, Burtey S. High resolution melt analysis for mutation screening in *PKD1* and *PKD2*. BMC Nephrol. 2011: 12;57.	Mutation screening in *PKD1* and *PKD2* with HRM in 37 unrelated patients with ADPKD.	There were 28 pathogenic mutations (25 in *PKD1* and 3 in *PKD2*) within 28 different patients. Furthermore, 52 new sequence variants were observed in *PKD1* and 2 in *PKD2*.
2.	Jas RM, Vasudevan R, Ismail P, Gafor AHA, Moin S, Eshkoor SA. Amplification of real-time high resolution melting analysis PCR method for polycystic kidney disease (PKD) gene mutations in autosomal dominant polycystic kidney disease patients. Afr. J. Biotechnol. 2012: 11(25); 6750-6757.	Eight common primers amplifying the respective genes in real-time high resolution melting analysis PCR (real-time *HRM* PCR) in terms of time, cost and sensitivity with respect to PCR-SSCP method	Case sample could be easily differentiated from control by differentiation of melting curve profile.