CHAPTER I

INTRODUCTION

1.1.Background

Mental retardation in Indonesia is not an uncommon disorder, with the National Survey report of 384,818 persons being mentally retarded in the year 2000, which counts for 0.19% of the country population at that time (National Survey, 2000). As mental retardation leads to stigma, discrimination, as well as social exclusion, underreporting is a likely phenomenon (Komardjaja, 2005). Genetic counseling services for families which have their members affected with mental retardation are essential due to the risk of recurrence and unavailability of treatment. There are still very few molecular genetics investigation in the Indonesian population, such as Fragile X screening as well as subtelomeric deletion and duplication studies (Faradz, 1998; Mundhofir, 2008).

Among the mentally retarded individuals, associated features are often found that may lead to specific diagnosis, such as abnormality of the head circumference. The head circumference measurement is a reflection of brain size (Cheong JL, 2008). Abnormalities in the measurement of head circumference may be caused by various underlying congenital, acquired or genetic problems (Stoll, 2001). Both microcephaly and macrocephaly, the presence of head circumference measurement below and above 2 standard deviations (SD) from the mean respectively, may occur in syndromic or non-syndromic disorders associated with mental retardation (Olney, 2007; Abuelo, 2007).

Mental retardation is commonly found together with abnormality of the head circumference, microcephaly or macrocephaly, as another main sign. In microcephaly, there is lack of normal growth of the brain cells, while in macrocephaly, there is too much growth of the brain cells. Both conditions

increases the risk for mental retardation, and there is a correlation between the degree of head circumference abnormality and the severity of cognitive impairment (Field, 2007; Ashwal, 2009). There is a high likelihood that mental retardation and abnormality of head circumference is caused by the same explanation, for example, a genetic mutation that is causing both mental retardation and microcephaly or macrocephaly (Abuelo, 2007; Olney, 2007).

Primary hereditary microcephaly (MCPH) is an autosomal recessive disorder characterized by microcephaly present at birth and mental retardation. The incidence of microcephaly at birth is between 1.3 and 150 per 100,000 live births (Kaindl, 2010). Mutations in the different genes, namely *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1* and *STIL* cause disruption of normal brain growth and leads to clinical manifestation of MCPH. The types of mutation that may occur in the MCPH genes are missense/nonsense, splicing, small deletions, small insertions, large deletions, large insertions/duplications, and translocations (Woods, 2005).

Macrocephaly is a known feature in several syndromic conditions. Several genes are known to cause syndromic macrocephaly. However, only one of these genes, the phosphatase and tensin homologue (*PTEN*) gene on 10q23.3, a tumor suppressor gene, is associated with the presence of macrocephaly and mental retardation (Varga, 2009). *PTEN* dysfunction has been associated with diseases characterised by overgrowth and also mental retardation. Even though some known genes causes macrocephaly, only *PTEN* gene cause macrocephaly and MR (Barker, 2003). Therefore this research will only focus on the investigation of *PTEN* gene in mentally retarded individuals with macrocephaly. The types of mutation known to affect *PTEN* gene and causing mental retardation with macrocephaly are deletions, insertions, missense, nonsense, splicing, and large deletions (Varga, 2009).

Little is known about the cause of microcephaly or macrocephaly which is associated with mental retardation in Indonesian population. This study looked for

and analysed the presence of mutations in the genes associated with MCPH: *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1* and *STIL* as well as in the *PTEN* gene for MR that is associated with macrocephaly by using DNA sequencing.

The understanding of possible genetic cause in the Indonesian mental retardation individuals with microcephaly or macrocephaly can assist in providing more appropriate diagnosis and genetic counseling in affected individuals and their family.

1.2.Research Questions

What are the mutation that can be found in the MCPH genes: *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1* and *STIL* gene of Indonesian mentally retarded individuals with microcephaly, and what are the mutations that can be found in the *PTEN* gene of Indonesian mentally retarded individuals with macrocephaly?

1.3. Research objectives

1.3.1. General research objectives

To identify and analyse the presence of underlying genetic mutation in the *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1*, *STIL* and *PTEN* genes of the Indonesian mentally retarded individuals with microcephaly or macrocephaly

1.3.2. Specific research objectives

1. To detect the presence of missense, nonsense, splicing, small deletions, and small insertions mutations of *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1* and *STIL* genes in the Indonesian mentally retarded individuals with microcephaly.

2. To analyse the potential pathogenic effect of these mutations that may be present in the *ASPM, WDR62, CENPJ, CDK5RAP2, MCPH1* and *STIL* genes in the Indonesian mentally retarded individuals with microcephaly.
3. To detect the presence of missense, nonsense, splicing, small deletions, small insertions, and large deletions mutations of the *PTEN* gene in the Indonesian mentally retarded individuals with macrocephaly.
4. To analyse the type of mutations and the potential pathogenic effect of the mutations that may be present in the *PTEN* gene in the Indonesian mentally retarded individuals with macrocephaly.

1.4.Research Benefits

1. To provide diagnosis for microcephaly or macrocephaly and mental retardation in the Indonesian population.
2. To provide appropriate genetic counselling for the family of Indonesian mental retardation individuals with microcephaly or macrocephaly.
3. To provide recommendation regarding genetic testing of MCPH or *PTEN* genes as early detection in Indonesian mental retardation individuals with microcephaly or macrocephaly.

1.5.Research Originality

Other studies in different populations have been done (table 1). However, this was the first research investigating the presence of genetic mutation in *MCPH* and *PTEN* gene in the Indonesian mental retardation individuals with microcephaly and macrocephaly. This study is a continuation of a previous study of Indonesian mental retardation individuals by Farmaditya Mundhofir and Alfi Afadiyanti (Mundhofir, 2008; Afadiyanti, 2011).

Table 1. Research Originality

No	Publications	Content
1	Woods CG, Bond J, Enard W. 2005. Autosomal recessive primary microcephaly (MCPH): a review of clinical, molecular, and evolutionary findings. *Am J Hum Genet*. 76(5):717-2	The first review on the clinical definition and molecular genetic basis of MCPH.
2.	Mahmood S, Ahmad W, Hassan MJ. 2011. Autosomal recessive primary microcephaly (MCPH): clinical manifestations, genetic heterogeneity and mutation continuum. *Orphanet J Rare Dis.* 6:39.	The review that describes various populations where MCPH genes have been investigated: Iranian, Turkish, Mexican, Arab, Indian, European, African, Brazilian families.
3.	Varga EA, Pastore M, Prior T, Herman GE, McBride KL. 2009. The prevalence of PTEN mutations in a clinical pediatric cohort with autism spectrum disorders, developmental delay, and macrocephaly. *Genet Med.* 11(2):111-7.	The first study to measure the prevalence of PTEN mutations in individuals with autism spectrum disorder, developmental delay, mental retardation, and macrocephaly.
4.	Goffin A, Hoefsloot LH, Bosgoed E, Swillen A, Fryns JP. 2001. *PTEN* mutation in a family with Cowden syndrome and autism. *Am J Med Genet*. 105(6):521-4.	The first study that describes *PTEN* mutations in subsets of individuals with Cowden syndrome and autism.
5.	Jackson AP, Eastwood H, Bell SM, Adu J, Toomes C, Carr IM, et al. 2002. Identification of microcephalin, a protein implicated in determining the size of the human brain. *Am J Med Genet*. 71(1):136-42.	The first study that discovers MCPH1 gene and its protein, microcephalin, and its role in the growth of the human brain.
6.	Hassan MJ, Khurshid M, Azeem Z, John P, Ali G, Chishti MS, et al. 2007. Previously described sequence variant in *CDK5RAP2* gene in a Pakistani family with autosomal recessive primary microcephaly. *BMC Med Genet.* 8:58.	The discovery of *CDK5RAP2* gene as causative for MCPH.