INTRODUCTION

Attention deficit hyperactivity disorder is a psychiatric disorder of the neurodevelopmental type ***(Caroline and Clauss-Ehlers, 2014)*** in which there are significant problems of attention, hyperactivity, or acting impulsively that are not appropriate for a person's age ***(Childress and Berry, 2012)***. These symptoms must begin by age six to twelve and be present for more than six months for a diagnosis to be made ***(Dulcan et al., 2014)***.

It affects about 6–7% of children when diagnosed via the DSM-IV criteria ***(Willcutt, 2012)***.

ADHD is diagnosed approximately three times more in boys than in girls ***(Emond and Joyal, 2009)***. About 30–50% of people diagnosed in childhood continue to have symptoms into adulthood and between 2–5% of adults have the condition**.** The cause of most cases of ADHD is unknown; however, it is believed to involve interactions between genetic and environmental factors. Certain cases are related to previous infection or trauma to the brain ***(Thapar and Cooper, 2013)***.

A large majority of ADHD cases may arise from a combination of various heterogeneity genes, many of which

directly affect dopamine neurotransmission. Those involved with dopamine include: **Dopamine transporter**, **Dopamine receptor D_4**, **Dopamine receptor D_5**, **Trace amine-associated receptor 1**, **Monoamine oxidase A**, **Catechol-O-methyl transferase**, and **Dopamine beta-monooxygenase** Other genes associated with ADHD include: **serotonin transporter**, **Synaptosomal-associated protein 25** (**SNAP-25**), **alpha-2A adrenergic receptor**, **Tryptophan hydroxylase 2** ***(Kebir and Tabbane, 2009)***.

The dopamine hypothesis for ADHD's neurological mechanism is the most probable and studied theory. It is based on a malfunctioning or decreased functioning of the dopamine system in particular regions of the brain. It is specifically based on the malfunctioning of the D4, D2, and other particular dopamine receptors in the brain in addition to abnormally low or high levels of dopamine in the brain in general ***(Shaw, 2007)***.

Many studies reported the association between ADHD and Dopamine D4 receptor gene after finding the 7 –repeat alle of the DRD4 exon III 48bp variable number of tandem repeat (VNTR) polymorphism ***(Varga et al., 2012)***.

that's why we decided to carry out this piece of work to test this hypothesis on Egyptian children with ADHD.

AIM OF THE WORK

The present study aims at:

1- Identification of the possibly existing polymorphisms in DRD4 receptor gene involved in the study with ADHD.

2- Trying to do phenotype-genotype correlation as regard the polymorphism in Dopamine receptor gene in a sample of Egyptian children with ADHD disorder.

Chapter (1)

ATTENTION DEFICIT HYPERACTIVE DISORDER "ADHD"

1-1 Definition of ADHD:

The essential feature of attention-deficit/hyperactivity disorder (ADHD) is a persistent pattern of inattention and/or hyperactivity-impulsivity that interferes with functioning or development.

Inattention manifests behaviorally in ADHD as wandering off task, lacking persistence, having difficulty sustaining focus, and being disorganized and is not due to defiance or lack of comprehension.

Hyperactivity refers to excessive motor activity (such as a child running about) when it is not appropriate, or excessive fidgeting, tapping, or talkativeness. In adults, hyperactivity may manifest as extreme restlessness or wearing others out with their activity.

Impulsivity refers to hasty actions that occur in the moment without forethought and that have high potential for harm to the individual (e.g., darting into the street without looking). Impulsivity may reflect a desire for immediate rewards or an inability to delay gratification. Impulsive

behaviors may manifest as social intrusiveness (e.g., interrupting others excessively) and/or as making important decisions without consideration of long-term consequences (e.g., taking a job without adequate information) (DSM_V) (***American Psychiatric Association: Diagnostic and Statistical Manual of Mental Disorders 5th Edition, 2013)***.

1-2 History of ADHD:

Early History of ADHD

Here is a brief overview of some of the early history of ADHD as reported by some authors:

Sir Alexander Crichton

The first example of a disorder that appears to be similar to ADHD was given by a Scottish physician Sir Alexander Crichton in 1798. In his publication, "An inquiry into the nature and origin of mental derangement", he mentioned "the incapacity of attending with a necessary degree of constancy to any one object". According to Crichton, the incapacity of attending, if not innate, can also be caused by nervous Disorders. This notion was later rediscovered in the concepts of minimal brain damage or dysfunction ***(Hodgkins et al., 2011).*** His work came after the German physician Melchior Adam Weikard who published a medical text book in 1775 which included a chapter called Attention Disorders.

Heinrich Hoffmann

In 1844, the German physician Heinrich Hoffmann created some illustrated children's stories including "Fidgety Phil". The description fits the hyperactive type of ADHD very well ***(Thome and Jacobs, 2004)***.

Sir George Frederic Still

The lectures of Sir George Frederic Still in 1902 are considered by many well known authors, such as Barkley and Conners, to be the scientific starting point of the history of ADHD. Still described 20 cases of children with a "defect of moral control as a morbid manifestation, without general impairment of intellect and without physical disease" ***(Still, 1902)***.

Alfred Frank Tredgold

Tredgold in 1908 discussed a correlation between early brain damage, for example, caused by birth defect or perinatal anoxia, and subsequent behavior problems or learning difficulties. At that time, epidemic encephalitis was spreading around the world; the residual effect of the encephalitis had many similarities to several ADHD symptoms. The concept of Post Encephalitic Behavior disorder was introduced then. Also, Tredgold was the first to discuss the importance of modifying the environment as part of the management of this disorder.

Franz Kramer and Hans Pollnow

In 1932, the German physicians Franz Kramer and Hans Pollnow reported on a hyperkinetic disease of infancy "the most characteristic symptom of affected children was a marked motor restlessness. The children are described as unable to stay still for a second, climb about preferring high furniture in particular." They also described several other features of ADHD.

Charles Bradley

In 1937, Charles Bradley from Rhode Island reported a positive effect of stimulant medication in children with various behavior disorders. This was the first description of the effect of stimulant treatment on ADHD. He used racemic amphetamine product ***(Findling, 2008)***.

However the first diagnostic description of what is now called ADHD occurred in The International Statistical Classification of Diseases Ninth Edition (ICD-9) and The Diagnostic and Statistical Manual of Mental Disorders, Second Edition (DSM-II) in 1965 and1968, respectively. ADHD was called hyperkinetic syndrome of childhood or hyperkinetic reaction. This term remains in the current ICD-10 as hyperkinetic disorder.

The DSM-III 1980 came with the first definition of attention deficit disorder (ADD) with or without hyperactivity ***(Lange et al., 2010)***.

In the DSM-III-R1987 the concept of two subtypes was removed and renamed the "Attention Deficit-Hyperactivity Disorder (ADHD) disorder." The symptoms of inattention, impulsivity, and hyperactivity were combined into a single list of symptoms with a single cutoff score, the term ADHD has persisted to the present ***(Lahey et al., 1994)***.

The DSM-IV (1994) added subtypes of predominantly hyperactive, predominantly inattentive, and combined ***(Lange et al., 2010)***.

The DSM-IV-TR (2000) was organized into a five-part axial system relating to different aspects of disorder or disability. ADHD was incorporated in The first axis. DSM-5 (2013), the diagnostic criteria for attention-deficit/ hyperactivity disorder (ADHD) in DSM-5 are similar to those in DSM-IV. The same 18 symptoms are used as in DSM-IV, and continue to be divided into two symptom domains (inattention and hyperactivity/ impulsivity), of which at least six symptoms in one domain are required for diagnosis **(*American Psychiatric Association, 2013)***.

1-3 Epidemiology of ADHD:

It is one of the most prevalent psychiatric disorders in childhood that is estimated to occur in 2–19% of children in the world ***(Skounti et al., 2007)***.

- ***Age:***

5 to 12 % in school-aged children ***(Wittchen et al., 2011)***. The course of symptoms from infancy to about the age of 3 years is variable, because of temperamental differences. By the age of 4 years, the diagnosis is more persistent into middle childhood ***(Taylor and Barke, 2008)***. Although its onset is usually before the age of 7 years, there is fewer data about prevalence of this disorder in preschoolers ***(Hakim shooshtary et al., 2010)***. Prevalence of ADHD in preschoolers is between 2% and 19.3% ***(Hakim shooshtary, 2010)***.

Roughly 80 % of children with ADHD will continue to meet diagnostic criteria for ADHD into their adolescent years and 60 % will maintain core symptoms into adulthood ***(Wittchen et al., 2011)***.

Previous studies have found that the percentage of parent reported (ever) ADHD diagnosed children below 17 years of age was 9.5% or about 5.4 million, which represents a 22% increase in four years from 2003 to 2007 ***(Boyle et al., 2011)***.

- ***Sex:***

Boys are more affected than girls with the ratio ranging from 2:1 to 9:1 ***(Skounti et al., 2007)***. This difference between genders may reflect either a difference in susceptibility or that females with ADHD are less likely to be diagnosed than males ***(Staller and Faraone, 2006)***.

In summary, sex differences in the prevalence and clinical presentation of ADHD are well-established but more research is required to explain these ***(Stergiakouli and Thapar, 2010)***.

- ***Geographical Distribution:***

Reports on the prevalence of ADHD have varied from 0.5% to 16%***;*** which is affected by many factors including the diagnostic criteria used, age and gender of the population, socioeconomic status and urban living ***(Meysamie et al., 2011)***.

Children in North America appear to have a higher rate of ADHD than children in Africa and the Middle East - however, this may be due to differing methods of diagnosis used in different areas of the world ***(Polanczyk et al., 2007)***.

It is estimated that ADHD affects between 5.4-8.7% of children in Africa **(Bakare, 2012)**. Data quality however is not high.

In the United States it is diagnosed in 2-16 percent of school children ***(Rader et al., 2009)***. The rates of diagnosis and treatment of ADHD are much higher on the east coast of the United States than on its west coast ***(Centers for Disease Control and Prevention, 2013)***.

In the UK an estimated 0.5 per 1,000 children had ADHD in the 1970s, while 3 per 1,000 received ADHD medications in the late 1990s. In the UK in 2003, 3.6 percent of male children and less than 1 percent in female children had the diagnosis ***(National Institute for Health and Clinical Excellence, 2008)***.

1-4 Etiology of ADHD:

The cause of most cases of ADHD is unknown; however, it is believed to involve interactions between genetic and environmental factors ***(Thapar, 2013)***. Certain cases are related to previous infection of or trauma to the brain ***(Millichap, 2010)***.

- ***The genetic basis for ADHD:***

Genetic factors are implicated in ADHD, but the mechanism of action is not completely understood. Twin,

family and adoption studies of ADHD have supported a strong genetic contribution to the disorder, with heritability ranging from 60-90% ***(Sharp et al., 2009)*** with evidence of shared familial/inherited risks for combined and inattentive type symptoms ***(Willcutt et al., 2012)***.

Genes regulating neurotransmitter systems have been implicated in ADHD. Candidate gene studies of ADHD have produced substantial evidence implicating several genes in the etiology of the disorder ***(Faraone and Mick, 2010)***.

Initial ADHD candidate gene studies were informed by an understanding of the actions of stimulant medications at the dopamine transporter and other targets related to alterations in catecholaminergic signaling ***(Faraone and Mick, 2010).***

Table (1): Candidate genes with the most consistent meta-analytic evidence for association with attention deficit hyperactivity disorder ***(Thapar et al., 2013)***.

Gene name	Codes for	Variant	Risk allele	Pooled odds ratio
Dopaminergic genes				
DRD4	Dopamine D4 receptor	VNTR in exon 3	7-repeat	1.33 (Smith, 2010)
		Polymorphism in promoter region	5-repeat	1.68 (Li et al., 2006)
			T allele	1.21 (Gizer et al., 2009)
DRD5	Dopamine D5 receptor	Dinucleotide repeat in 5′ flank	148-bp allele	1.23 (Gizer et al., 2009)
DAT1	Carrier protein involved in dopamine reuptake	VNTR in intron 8	3 repeat	1.25 (Gizer et al., 2009)
		Polymorphism in 3′UTR	G allele	1.20 (Gizer et al., 2009)
		VNTR in 3′UTR	10 repeat	1.17 (Yang et al., 2007)
Serotonergic genes				
5HTT	Carrier protein involved in serotonin reuptake	5HTTLPR polymorphism in promoter region	Long allele	1.31 (Faraone et al., 2005)
HTR1B	Serotonin 1B receptor	Polymorphism in exon 1	G allele	1.11 (Gizer et al., 2009)
Other				
SNAP-25	Protein involved in neurotransmitter release, synaptic plasticity and axonal growth	Polymorphism in 3′UTR	Not known	1.19 (Faraone et al., 2005)

Dopaminergic system:

The effectiveness of methylphenidate, which acts by blocking the dopamine transporter, in ADHD treatment as well as the association of ADHD with those executive neuropsychological functions and fronto striatal pathways, that are dependent on an intact dopaminergic neurotransmission make the dopaminergic system the most intensively analyzed neurotransmitter system in ADHD ***(Swanson et al., 2007)***.

❖ ***DRD4:***

The dopamine D4 receptor gene, DRD4, is the most replicated gene in the field with over 20 studies examining an association between DRD4 and ADHD ***(Faraone et al., 2005).*** DRD4 encodes a 7-transmembrane G-protein-coupled dopamine receptor and is expressed in regions implicated in ADHD, such as the limbic system, frontal cortex, and globus pallidus. DRD4 mediates postsynaptic actions of dopamine.

The DRD4 7-repeat allele increases the risk for ADHD as it seems to alter the function of the encoded receptor by making it less sensitive to dopamine than the alternative alleles ***(Banaschewski et al., 2010)***.

❖ ***DRD5 gene:***

Another dopaminergic gene, the *DRD5* has been investigated in relation to ADHD and the gene was found to be significantly associated with ADHD in four meta-analytic studies ***(Stergiakouli and Thapar, 2010)***.

The most recent meta-analysis reported a significant association with moderate heterogeneity in reported effect sizes ***(Gizer et al., 2009)***.

❖ ***DAT1:***

The dopamine transporter gene, DAT1, mediates the active reuptake of dopamine from the synapse and is a principal

regulator of dopaminergic neurotransmission. Interestingly, many individuals with ADHD respond well to medications such as methylphenidate that block DAT1 leading to increased amount and duration of dopamine in the synapse, in addition the increased density of the dopamine transporter in ADHD brains is reported to normalize with methylphenidate treatment furthermore, mice that lack the dopamine transporter gene are hyperactive ***(Wallis et al., 2008)***.

- **Serotonergic system:**

Serotonin dysregulation has been hypothesized to play a causal role in ADHD ***(Oades, 2008)***.

The main candidate genes studied within the serotonergic system are those coding for the serotonin transporter (5-HTT), the 1B and 2A serotonin receptors (HTR1B) and (HTR2A) and the dopamine decarboxylase (DDC) and tryptophan hydroxylase (TPH2) genes. Several other serotonin receptor genes have also been studied much less extensively ***(Banaschewski et al., 2010)***.

The environmental basis of ADHD

1. Pre-natal environmental factor:-

Play an important role in the pathogenesis of ADHD. Prenatal factors are associated with maternal lifestyle during pregnancy as: