



## RESEARCH ARTICLE

# Genetic Polymorphism in the Promoter Region of Serotonin Transporter: Implications for Ethanol Abuse in Children and Adolescents

Carlos Eduardo Coral de Oliveira Ph.D.<sup>1</sup>; Julie Massayo Maeda Oda Ph.D.<sup>2</sup>;  
Carolina Batista Ariza Ph.D.<sup>1</sup>; Roberta Losi Guembarovski Ph.D.<sup>1</sup>;  
Bruna Karina Banin Hirata, M.S.<sup>1</sup>; Felipe Campos de Almeida, B.S.<sup>1</sup>;  
Nayara Delgado André Ph.D.<sup>3</sup>; Maria Helena Pelegrinelli Fungaro Ph.D.<sup>4</sup>;  
Maria Angelica Ehara Watanabe Ph.D.<sup>1</sup>

## Abstract

**Objectives:** To provide a review of published literature regarding genetic polymorphism of serotonin transporter gene, named as 5-HTTLPR, and its potential role as a susceptibility marker for ethanol abuse in childhood and adolescence. **Methods:** A literature review of several databases was conducted with the following keywords: 5-HTTLPR, children or adolescents or teenagers, susceptibility, alcohol or ethanol, abuse or misuse. **Results:** Alcohol interacts with serotonergic synaptic transmission in several ways, and the reduced availability of serotonin transporters might foster brain dysfunction, driving to alcohol abuse. The initial use of ethanol in children and adolescents is determined primarily by environmental influences, whereas the establishment of drinking patterns is strongly controlled by genetic factors. Functional polymorphic variants in the promoter region of the 5-HTTLPR gene have age-dependent effects in alcohol abuse. This polymorphism, mapped to the 5' region of the *SLC6A4*, is a variable number of tandem repeats (VNTR) and involves a direct repeat of 20-23 base pairs GC-rich sequences, comprising a short (S) allele, consisting of 14 repeats, and a long (L) allele, with 16 repeats. Additional variants have been described, although their influences on childhood and adolescence ethanol use are not clear. **Conclusion:** The influence of the 5-HTTLPR allelic variants in children and adolescent misuse of alcohol might be considered for clinical management, preventing long-term behavior problem. Identifying genetic markers associated to the potential alcohol misuse or abuse could be useful in guiding management and formulating effective coping strategies.

**Key Words:** alcohol abuse, serotonin transporter, genetic polymorphism.

## Résumé

**Objectifs:** Offrir une revue de la littérature publiée sur le polymorphisme génétique du gène transporteur de la sérotonine, nommé 5-HTTLPR, et son rôle potentiel de marqueur de la susceptibilité à l'abus d'éthanol dans l'enfance et l'adolescence. **Méthodes:** Une revue de la littérature dans plusieurs bases de données a été menée à l'aide des mots clés suivants: 5-HTTLPR, enfants ou adolescents ou teenagers, susceptibilité, alcool ou éthanol, abus ou excès. **Résultats:** L'alcool interagit de plusieurs façons avec la transmission synaptique sérotoninergique, et la disponibilité réduite des transporteurs de la sérotonine peut favoriser une dysfonction cérébrale, qui mène à l'abus d'alcool. L'utilisation initiale d'éthanol chez les enfants et les adolescents est déterminée principalement par des influences environnementales, alors que l'établissement

<sup>1</sup>State University of Londrina, Department of Pathological Sciences, Londrina, Brazil

<sup>2</sup>Federal University of Mato Grosso do Sul, Department of Medicine, Três Lagoas, Brazil

<sup>3</sup>Federal University of São João Del-Rei, Divinópolis, Brazil

<sup>4</sup>State University of Londrina, Department of General Biology, Londrina, Brazil

Corresponding E-Mail: maewatuel@gmail.com

Submitted: September 18, 2014; Accepted: September 24, 2015

de modèles de consommation d'alcool est fortement contrôlé par des facteurs génétiques. Les variantes polymorphiques fonctionnelles de la région promotrice du gène 5-HTTLPR ont des effets selon l'âge sur l'abus d'alcool. Ce polymorphisme, localisé à la région 5' de *SLC6A4*, est un nombre variable de répétitions en tandem (NVRT) et implique une répétition directe de séquences de 20-23 paires de base riches en GC, comprenant un allèle court (C), consistant en 14 répétitions, et un allèle long (L), avec 16 répétitions. Les variantes additionnelles ont été décrites, bien que leurs influences sur l'utilisation d'éthanol dans l'enfance et l'adolescence ne soient pas définies. **Conclusion:** L'influence des variantes alléliques de 5-HTTLPR sur l'excès d'alcool chez les enfants et les adolescents pourrait être considérée pour la prise en charge clinique, et la prévention de problèmes de comportement à long terme. L'identification des marqueurs génétiques associés à l'excès ou l'abus d'alcool potentiel pourrait être utile pour guider la prise en charge et formuler des stratégies d'adaptation efficaces.

**Mots clés:** abus d'alcool, transporteur de la sérotonine, polymorphisme génétique.

## Introduction

Excessive alcohol use constitutes a serious public health problem in modern societies, and its effect reverberates both in physical and mental health. In almost all human cultures, where drinking has been considered a social activity, a minority cannot keep alcohol use within safe limits of consumption and some of them may abuse alcohol or become dependents. In Brazil, for instance, 35 million people less than 30 years old have problems related to psychoactive substance abuse, including alcohol (IBGE, 2013).

Concerning the etiology of addiction vulnerability, an interaction between genetic susceptibility and environmental stressors is assumed (Benjamin, 2002). Particularly, it is estimated that as much as 40% of variance in alcohol dependence may be resultant to genetic factors (Jentsch et al., 2014; Kenna et al., 2012; Z. Li & Zhang, 2013).

Several neurotransmitter systems, including the serotonergic and noradrenergic, are involved in substance abuse behavior (Goodman, 2008), and genes controlling these systems are therefore of interest in various aspects of drug use. Serotonin (5-hydroxytryptamine, 5-HT) is a monoamine neurotransmitter that regulates numerous important physiological processes, including sleep, food intake, pain, vascular tone, platelet function, and motor activity (Mohammad-Zadeh, Moses, & Gwaltney-Brant, 2008; Sanders-Bush, 2006). The serotonin transporter protein (5-HTT), a transporter located on the presynaptic neuron, regulates the serotonin reuptake from the synaptic cleft and therefore has a pivotal role within serotonergic neurotransmissions (Sanders-Bush, 2006).

A functional polymorphism of the serotonin transporter gene (5-HTTLPR; located on chromosome 17q12) has been considered within various studies. This gene-linked polymorphic region of 5-HTT is located at upstream of the transcription start site in 5' flanking region (Heils et al., 1996), and involves a direct variable number in tandem repeat (VNTR) of 20-23 base pairs (bp) GC-rich sequences, comprising a short (S) allele, consisting of 14 repeats, and a long (L) allele, with 16 repeats (Caspi et al., 2003).

Proposed factors that would increase vulnerability to alcohol dependence include the developmental stage, exposure to early life adversity (ranging from abuse, neglect, and

bullying), the socioeconomic background (higher family income), having an alcoholic parent (Mesic et al., 2013; Rehorcikova et al., 2013), life style without religion, and genetic predisposition; however, how these factors are involved separately or conjointly in alcohol consumption remain uncovered. In this context, this review focuses on the human 5-HTTLPR polymorphism and its implications for ethanol abuse in children and teenagers.

## Implications of Serotonin Transporter 5-HTT for ethanol abuse

Age-dependent changes in self-regulation and cognition are inversely related to impulsive behavior. Impulsivity is an important risk factor of a severe course of alcohol dependence (Jakubczyk et al., 2013), and it is evidenced in young children, gradually decreasing in a mostly linear manner into adulthood (Luna, Garver, Urban, Lazar, & Sweeney, 2004).

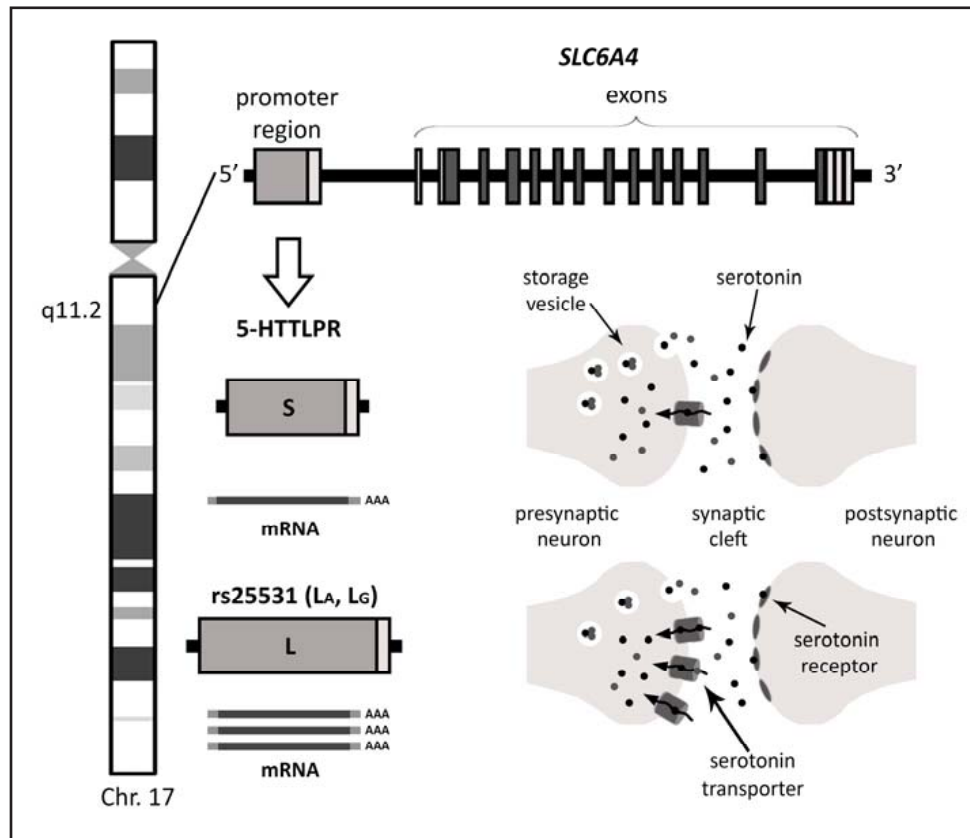
Moreover, reduced serotonergic neurotransmission is implicated in impulsive behavior, and 5-HTT is one of the most important modulators of serotonergic neurotransmission because it is responsible for the reuptake of 5-HT at nerve terminals, and thus determines the magnitude and duration of 5-HT signaling (Nomura et al., 2015). Particularly, the system of the 5-HTTLPR and acute manipulation of serotonin were independently and additively, associated with the elevated impulsive response style (Wedekind et al., 2010).

The low serotonin function is associated with alcoholism, leading to the speculation that increasing serotonin function could decrease ethanol consumption. Therefore, increasing extracellular serotonin might decrease ethanol intake. Extracellular serotonin can be pharmacologically increased by blocking the serotonin transporter using selective serotonin reuptake inhibitors (Sellers, Higgins, & Sobell, 1992).

Furthermore, variation in the clinical presentation of alcoholism, for example, in terms of age of onset, predisposing personalities, psychiatric comorbidity, severity of the disease and withdrawal symptoms (Moss, Chen, & Yi, 2007), suggests inter-individual differences in mechanisms of

### Figure 1. Schematic view of the polymorphisms associated with the serotonin transporter gene.

After a neuron stimulation, the 5-HT-contained vesicle fuses with the neuronal membrane and the neurotransmitter is released in the synaptic cleft. Serotonin can bind to receptors on neurons, both postsynaptic and presynaptic, and trigger the effector functions of serotonergic neurotransmission. The serotonin transporter (5-HTT) that activates neuronal reuptake of serotonin can modulate the concentration of serotonin in the cleft. The insertion/deletion in the promoter region, named 5-HTTLPR polymorphism is due to a direct VNTR of 20-23 bp deletion (S)/ insertion (L). LL genotype may express more membrane 5-HTT and relatively increase serotonin uptake when compared to S-allele carriers.



vulnerability including genetic risk factors (Enoch, 2013). Notably, variations in the 5-HTTLPR, a polymorphic region known as the serotonin-transporter-linked polymorphic region, can modulate serotonin transporter 5-HTT gene transcription, and thus, alter serotonergic signaling.

In fact, it is also known that both alcohol and nicotine stimulate the dopamine and the serotonin systems. And although a possible interaction between these neurotransmitter systems in substance use behavior have been suggested, it is not completely elucidated.

### Serotonin Transporter Linked Polymorphic Region (5-HTTLPR)

Solute carrier family 6 member 4 (SLC6A4) gene encoding 5-HTT resides on chromosome 17q11.1–17q12 in the human genome, consisting of 14 exons, and encoding a 630-amino acid protein (Jarrett et al., 2007). Three

polymorphisms were well described in distinct regions of this gene: a functional polymorphism with insertion or deletion of 20-23 bp-long repeat elements in the upstream regulatory region called 5-HTTLPR (Heils et al., 1996); a variable number of tandem repeats (VNTR) STin2, located in intron 2 and consisting of a variable number (usually 9, 10, or 12) of nearly identical 17 bp (5-HTTVNTR2) (Lesch et al., 1994); and a single nucleotide polymorphism (SNP I425V) in 3' untranslated region (UTR) (Battersby et al., 1999).

The promoter activity of the 5-HTT gene is modified by sequential elements within the proximal 5-HTTLPR (Caspi et al., 2003), and the polymorphism of the 5-HTTLPR (Figure 1) affects the expression of serotonin transporter (Heils et al., 1996). Reduced serotonin uptake and lower transcriptional efficiency of the promoter are associated with S-allele, comparing to more frequent L-allele (Lesch et al., 1996). The former has been related to suicidal behavior

(D. Li & He, 2007), depression (Lotrich & Pollock, 2004), neurotic personality trait (Lesch et al., 1996; Takano et al., 2007), cigarette smoking (Watanabe et al., 2011) and alcoholism (X. Hu et al., 2005).

A theoretical explanation for different effects of this variant allele is associated to its influence on expression levels of 5-HTT mRNA. Cell lines expressing the L-allele showed 1.4 to 1.7 fold increased mRNA levels in comparison to S-allele variant (Lesch et al., 1996). In addition, LL homozygotes lymphoblastoid cells expressed 30% higher 5-HTT mRNA as those carrying at least one S-allele (Hranilovic et al., 2004).

Otherwise, 5-HTTLPR polymorphism may influence transporter protein synthesis. Lymphoblasts carrying LL genotype expressed 30-40% more membrane 5-HTT and relatively two fold increased serotonin uptake than S-allele carriers or homozygous cells (Heils, Mossner, & Lesch, 1997).

In this regard, it was shown that 5-HTTLPR variants might also alter serotonergic neurotransmission by modifying kinetic regulation of 5-HT receptors. *In vivo* human imaging study showed that 5-HTTLPR S-allele carriers or SS homozygous had lower 5-HT<sub>1A</sub> receptor binding potential values than those with LL genotypes, which possibly represents a desensitization and downregulation of this receptor as a result of a lifelong increase in 5-HT (David et al., 2005).

A third variant of 5-HTTLPR polymorphism, the L<sub>G</sub> allele has been described. A single nucleotide polymorphism (rs25531) changes an adenosine (L<sub>A</sub> allele) to a guanine (L<sub>G</sub> allele) (Nakamura, Ueno, Sano, & Tanabe, 2000), resulting in lower transcriptional activity of the gene (X. Z. Hu et al., 2006), similar to S-allele. The functional role of L<sub>A</sub> and L<sub>G</sub> alleles was not distinguished in some genetic studies and recently, Nardi et al. (2013) reported that it may not be polymorphic. However, it must imply in controversial results in association studies (Saiz et al., 2009).

In addition, uncommon alleles that are longer than the L variant have also been found. The VL and XL variants were firstly described by Gelernter, Kranzler, and Cubells (1997) study, and seems to be 40bp (18 repeats) and 81bp (20 repeats), respectively, longer than the L-allele. In fact, XL-alleles were revealed by sequence analysis, and arose through duplication of an internal segment composed of repeat elements VI to IX, comprising 85bp in total, and, most notably, includes the segments deleted in the S-allele (Delbruck et al., 1997).

While most of the studies demonstrated an association between 5-HTTLPR variants and alcohol dependence, the association with specific alleles is inconsistent, as well as on alcohol subtype, type of drinking behavior, ethnicity, comorbid diagnoses, age of onset (Kenna et al., 2012), and absence of evaluating other 5-HTTLPR alleles, such as L<sub>G</sub> and L<sub>A</sub>. The important question is whether all 5-HTTLPR

variants are functional and alter the serotonin transporter expression, and how their presence is important to change drinking behavior.

## Influence of 5-HTTLPR on ethanol abuse in children or teenagers

The study of alcohol use by children ages 12 and younger has been very limited (Donovan, 2014). Nevertheless, age of onset of alcohol use increases as a function of the age of the adolescents (Parra, O'Neill, & Sher, 2003). An American study estimated that 28.1 percent of 9th graders reported that they drank alcohol before age 13, compared with 14.2 percent of 12th graders (Eaton et al., 2010).

In this context, initiation of alcohol use before 14 years old is a potent predictor of later drinking problems, and is associated with a 40% risk for the development of alcohol dependence (Grant & Dawson, 1998). Indeed, early alcohol use was predicted by gene-environment interaction, as the presence of 5-HTTLPR S-allele in children who suffer from maltreatment was related to an increased risk of ethanol abuse (Martin, Volkmar, & Lewis, 2007).

A large cohort study using the Estonian Children Personality Behavior and Health Study highlighted the importance of investigating 5-HTTLPR genetic variants as a risk factor for drinking problems. Vaht, Merenakk, Maestu, Veidebaum, and Harro (2014) selected initially nine year-old (recalled at ages 15 and 18) and 15 year-old (recalled at ages 18 and 25) children, who provided self-reports on their alcohol use in all data collection waves (complete data available n=1,075). They verified that females with the SS genotype in the older cohort were the latest alcohol experimenters, while the SS females of younger cohort had tried alcohol earlier than any other group. In males, there was no significant cohort versus genotype interaction, but the 5-HTTLPR genotype was associated with alcohol use, and the SS genotype subjects reported the highest consumption.

These evidences suggest that genetic factors may provide clues about individual differences in overall environmental sensitivity (Simons et al., 2011). Indeed, social influences were partially dependent on 5-HTTLPR genotype, such as homozygous carriers of the S-allele were associated with stronger susceptibility to the influences of school-level smoking and drinking patterns. Hence, it has become reasonable that not only the differential susceptibility environment, but genetic factors may influence health behaviors, and possibly, vulnerability to drug use (Daw et al., 2013).

van der Zwaluw et al. (2010) tested whether 5-HTTLPR influenced the development over time of adolescent alcohol use. Adolescents with the 5-HTTLPR S-allele developed higher levels of alcohol consumption compared to L-allele carriers, revealing that the 5-HTTLPR polymorphism is associated with the development of alcohol use from early to late adolescence.

Moreover, drinking is very common among young adults, mainly those who are attending college, and the S-allele was significantly associated with higher levels of drinking in young adulthood (Guo, Wilhelmsen, & Hamilton, 2007). In addition, drinking to “get drunk” was more frequent in S-allele homozygous college students (Herman, Philbeck, Vasilopoulos, & Depetrillo, 2003).

The 5-HTTLPR S-allele was associated with increased drinking and drug use among European-American (Covault et al., 2007) and African-descent (Kranzler et al., 2012) college students who have experienced multiple negative life events. The S-allele carriers seemed to be at risk for a variety of adverse behavioral outcomes in response to stress.

Moreover, compelling evidences indicated that the S-allele carriers have an increased risk for early onset alcoholism (Hallikainen et al., 1999), have engaged more in heavy episodic drinking (Herman et al., 2003) and have a higher risk for alcohol dependence (Feinn, Nellissery, & Kranzler, 2005).

Gerra et al. (2010) investigated the involvement of 5-HTTLPR polymorphism, childhood parental neglect reported retrospectively and hypothalamus-pituitary-adrenal (HPA) axis function, with the susceptibility to use illicit drugs and alcohol abuse. They demonstrated significant association of perceived childhood neglect, 5-HTT gene variants and hypothalamus-pituitary-adrenal axis dysregulation, with substance abuse susceptibility. The higher frequency of the 5-HTTLPR SS genotype was associated with an increased susceptibility to use illegal psychotropic drugs among the adolescents.

Nobile et al. (2013) examined the moderating role of the 5-HTTLPR in socioeconomic status and family structure, and internalizing symptoms over adolescence. They reported that anxiety problems in the S-carriers seemed to be more stable and to predict a possible evolution towards the development of depressive problems. In the LL subjects, the anxiety trait was significantly less stable, and, in late-adolescence, seemed to be significantly predicted by socioeconomic status, suggesting a possible gene-environment interaction.

Altogether, the overview of the neurobiological development of addiction indicates that the diathesis develops as a result of some combination of genetic, prenatal, infancy, and childhood factors (Goodman, 2009). Increasing evidences suggest that genetic information can be useful in refining the choice of addiction treatment. However, as genetic testing becomes more common in the practice of medicine, a variety of ethical and practical challenges, some of which are unique to drug addiction, will also need to be considered (Ho et al., 2010).

It is possible that genetic variants are directly capable of altering reward, tolerance, and withdrawal, thereby predisposing individuals to alcohol dependence. Although

drinking behavior could be influenced by socioeconomic conditions and is therefore subject to cohort effects, the genetic polymorphism of 5-HTTLPR has been related to several aspects of alcohol abuse. All in all, 5-HTTLPR S-allele could emerge as a susceptibility marker of alcohol abuse in children and adolescents, although it is crucial to evaluate the different environmental interaction effects.

## Acknowledgments/Conflicts of Interest

The authors would like to acknowledge Fundação Araucária do Paraná, Secretaria da Ciência, Tecnologia e Ensino Superior (SETI), Fundo Estadual para a Infância e Adolescência (FIA/PR), Secretaria da Família e Desenvolvimento Social (SEDS) and Coordination of Undergraduate Studies of Londrina State University (PROPPG-UEL). The authors have no conflicts of interest to declare.

## References

- Battersby, S., Ogilvie, A. D., Blackwood, D. H., Shen, S., Muqit, M. M., Muir, W. J., . . . Harmar, A. J. (1999). Presence of multiple functional polyadenylation signals and a single nucleotide polymorphism in the 3' untranslated region of the human serotonin transporter gene. *J Neurochem*, 72(4), 1384-1388.
- Benjamin, L. S. (2002). *Interpersonal Diagnosis and Treatment of Personality Disorders*: Guilford Press.
- Caspi, A., Sugden, K., Moffitt, T. E., Taylor, A., Craig, I. W., Harrington, H., . . . Poulton, R. (2003). Influence of life stress on depression: moderation by a polymorphism in the 5-HTT gene. *Science*, 301(5631), 386-389. doi: 10.1126/science.1083968
- Covault, J., Tennen, H., Armeli, S., Conner, T. S., Herman, A. I., Cillessen, A. H., & Kranzler, H. R. (2007). Interactive effects of the serotonin transporter 5-HTTLPR polymorphism and stressful life events on college student drinking and drug use. *Biol Psychiatry*, 61(5), 609-616. doi: 10.1016/j.biopsych.2006.05.018
- David, S. P., Murthy, N. V., Rabiner, E. A., Munafo, M. R., Johnstone, E. C., Jacob, R., . . . Grasby, P. M. (2005). A functional genetic variation of the serotonin (5-HT) transporter affects 5-HT1A receptor binding in humans. *J Neurosci*, 25(10), 2586-2590. doi: 10.1523/JNEUROSCI.3769-04.2005
- Daw, J., Shanahan, M., Harris, K. M., Smolen, A., Haberstick, B., & Boardman, J. D. (2013). Genetic sensitivity to peer behaviors: 5HTTLPR, smoking, and alcohol consumption. *J Health Soc Behav*, 54(1), 92-108. doi: 10.1177/0022146512468591
- Delbruck, S. J., Wendel, B., Grunewald, I., Sander, T., Morris-Rosendahl, D., Crocq, M. A., . . . Hoehe, M. R. (1997). A novel allelic variant of the human serotonin transporter gene regulatory polymorphism. *Cytogenet Cell Genet*, 79(3-4), 214-220.
- Donovan, J. E. (2014). The Burden of Alcohol Use: Focus on Children and Preadolescents. *Alcohol Res Cur Rev*, 35(2), 6.
- Eaton, D. K., Kann, L., Kinchen, S., Shanklin, S., Ross, J., Hawkins, J., . . . Prevention. (2010). Youth risk behavior surveillance - United States, 2009. *MMWR Surveill Summ*, 59(5), 1-142.
- Enoch, M. A. (2013). Genetic influences on response to alcohol and response to pharmacotherapies for alcoholism. *Pharmacol Biochem Behav*. doi: 10.1016/j.pbb.2013.11.001
- Feinn, R., Nellissery, M., & Kranzler, H. R. (2005). Meta-analysis of the association of a functional serotonin transporter promoter polymorphism with alcohol dependence. *Am J Med Genet B Neuropsychiatr Genet*, 133B(1), 79-84. doi: 10.1002/ajmg.b.30132
- Gelernter, J., Kranzler, H., & Cubells, J. F. (1997). Serotonin transporter protein (SLC6A4) allele and haplotype frequencies and linkage disequilibrium in African- and European-American and Japanese populations and in alcohol-dependent subjects. *Hum Genet*, 101(2), 243-246.

- Gerra, G., Zaimovic, A., Castaldini, L., Garofano, L., Manfredini, M., Somaini, L., . . . Donnini, C. (2010). Relevance of perceived childhood neglect, 5-HTT gene variants and hypothalamus-pituitary-adrenal axis dysregulation to substance abuse susceptibility. *Am J Med Genet B Neuropsychiatr Genet*, *153B*(3), 715-722. doi: 10.1002/ajmg.b.31038
- Goodman, A. (2008). Neurobiology of addiction. An integrative review. *Biochem Pharmacol*, *75*(1), 266-322. doi: 10.1016/j.bcp.2007.07.030
- Goodman, A. (2009). The Neurobiological Development of Addiction - An Overview. *Psychiatric Times*, *26*(9), 14.
- Grant, B. F., & Dawson, D. A. (1998). Age of onset of drug use and its association with DSM-IV drug abuse and dependence: results from the National Longitudinal Alcohol Epidemiologic Survey. *J Subst Abuse*, *10*(2), 163-173.
- Guo, G., Wilhelmsen, K., & Hamilton, N. (2007). Gene-lifecourse interaction for alcohol consumption in adolescence and young adulthood: five monoamine genes. *Am J Med Genet B Neuropsychiatr Genet*, *144B*(4), 417-423. doi: 10.1002/ajmg.b.30340
- Hallikainen, T., Saito, T., Lachman, H. M., Volavka, J., Pohjalainen, T., Ryyanen, O. P., . . . Tiihonen, J. (1999). Association between low activity serotonin transporter promoter genotype and early onset alcoholism with habitual impulsive violent behavior. *Mol Psychiatry*, *4*(4), 385-388.
- Heils, A., Mossner, R., & Lesch, K. P. (1997). The human serotonin transporter gene polymorphism--basic research and clinical implications. *J Neural Transm*, *104*(10), 1005-1014.
- Heils, A., Teufel, A., Petri, S., Stober, G., Riederer, P., Bengel, D., & Lesch, K. P. (1996). Allelic variation of human serotonin transporter gene expression. *J Neurochem*, *66*(6), 2621-2624.
- Herman, A. I., Philbeck, J. W., Vasilopoulos, N. L., & Depetrillo, P. B. (2003). Serotonin transporter promoter polymorphism and differences in alcohol consumption behaviour in a college student population. *Alcohol Alcohol*, *38*(5), 446-449.
- Ho, M. K., Goldman, D., Heinz, A., Kaprio, J., Kreek, M. J., Li, M. D., . . . Tyndale, R. F. (2010). Breaking barriers in the genomics and pharmacogenetics of drug addiction. *Clin Pharmacol Ther*, *88*(6), 779-791. doi: 10.1038/clpt.2010.175
- Hranilovic, D., Stefulj, J., Schwab, S., Borrmann-Hassenbach, M., Albus, M., Jernej, B., & Wildenauer, D. (2004). Serotonin transporter promoter and intron 2 polymorphisms: relationship between allelic variants and gene expression. *Biol Psychiatry*, *55*(11), 1090-1094. doi: 10.1016/j.biopsych.2004.01.029
- Hu, X., Oroszi, G., Chun, J., Smith, T. L., Goldman, D., & Schuckit, M. A. (2005). An expanded evaluation of the relationship of four alleles to the level of response to alcohol and the alcoholism risk. *Alcohol Clin Exp Res*, *29*(1), 8-16.
- Hu, X. Z., Lipsky, R. H., Zhu, G., Akhtar, L. A., Taubman, J., Greenberg, B. D., . . . Goldman, D. (2006). Serotonin transporter promoter gain-of-function genotypes are linked to obsessive-compulsive disorder. *Am J Hum Genet*, *78*(5), 815-826. doi: 10.1086/503850
- IBGE, I. B. d. G. e. E.-. (2013). *Projeção da população do Brasil por sexo e idade para o período de 2000/2060*. Rio de Janeiro: Instituto Brasileiro de Geografia e Estatística.
- Jakubczyk, A., Klimkiewicz, A., Mika, K., Bugaj, M., Konopa, A., Podgórska, A., . . . Wojnar, M. (2013). Psychosocial predictors of impulsivity in alcohol-dependent patients. *J Nerv Ment Dis*, *201*(1), 43-47. doi: 10.1097/NMD.0b013e31827aaf9d
- Jarrett, M. E., Kohen, R., Cain, K. C., Burr, R. L., Poppe, A., Navaja, G. P., & Heitkemper, M. M. (2007). Relationship of SERT polymorphisms to depressive and anxiety symptoms in irritable bowel syndrome. *Biol Res Nurs*, *9*(2), 161-169. doi: 10.1177/1099800407307822
- Jentsch, J. D., Ashenhurst, J. R., Cervantes, M. C., Groman, S. M., James, A. S., & Pennington, Z. T. (2014). Dissecting impulsivity and its relationships to drug addictions. *Ann N Y Acad Sci*. doi: 10.1111/nyas.12388
- Kenna, G. A., Roder-Hanna, N., Leggio, L., Zywiak, W. H., Clifford, J., Edwards, S., . . . Swift, R. M. (2012). Association of the 5-HTT gene-linked promoter region (5-HTTLPR) polymorphism with psychiatric disorders: review of psychopathology and pharmacotherapy. *Pharmacogenomics Pers Med*, *5*, 19-35. doi: 10.2147/PGPM.S23462
- Kranzler, H. R., Scott, D., Tennen, H., Feinn, R., Williams, C., Armeli, S., . . . Covault, J. (2012). The 5-HTTLPR polymorphism moderates the effect of stressful life events on drinking behavior in college students of African descent. *Am J Med Genet B Neuropsychiatr Genet*, *159B*(5), 484-490. doi: 10.1002/ajmg.b.32051
- Lesch, K. P., Balling, U., Gross, J., Strauss, K., Wolozin, B. L., Murphy, D. L., & Riederer, P. (1994). Organization of the human serotonin transporter gene. *J Neural Transm Gen Sect*, *95*(2), 157-162.
- Lesch, K. P., Bengel, D., Heils, A., Sabol, S. Z., Greenberg, B. D., Petri, S., . . . Murphy, D. L. (1996). Association of anxiety-related traits with a polymorphism in the serotonin transporter gene regulatory region. *Science*, *274*(5292), 1527-1531.
- Li, D., & He, L. (2007). Meta-analysis supports association between serotonin transporter (5-HTT) and suicidal behavior. *Mol Psychiatry*, *12*(1), 47-54. doi: 10.1038/sj.mp.4001890
- Li, Z., & Zhang, H. (2013). Analyzing Interaction of mu-, delta- and kappa-opioid Receptor Gene Variants on Alcohol or Drug Dependence Using a Pattern Discovery-based Method. *J Addict Res Ther, Suppl* *7*, 007.
- Lotrich, F. E., & Pollock, B. G. (2004). Meta-analysis of serotonin transporter polymorphisms and affective disorders. *Psychiatr Genet*, *14*(3), 121-129.
- Luna, B., Garver, K. E., Urban, T. A., Lazar, N. A., & Sweeney, J. A. (2004). Maturation of cognitive processes from late childhood to adulthood. *Child Dev*, *75*(5), 1357-1372. doi: 10.1111/j.1467-8624.2004.00745.x
- Martin, A., Volkmar, F. R., & Lewis, M. (2007). *Lewis's Child and Adolescent Psychiatry: A Comprehensive Textbook*: Wolters Kluwer Health/Lippincott Williams & Wilkins.
- Mesic, S., Ramadani, S., Zunic, L., Skopljak, A., Pasagic, A., & Masic, I. (2013). Frequency of Substance Abuse Among Adolescents. *Mater Sociomed*, *25*(4), 265-269. doi: 10.5455/msm.2013.25.265-269
- Mohammad-Zadeh, L. F., Moses, L., & Gwaltney-Brant, S. M. (2008). Serotonin: a review. *J Vet Pharmacol Ther*, *31*(3), 187-199. doi: 10.1111/j.1365-2885.2008.00944.x
- Moss, H. B., Chen, C. M., & Yi, H. Y. (2007). Subtypes of alcohol dependence in a nationally representative sample. *Drug Alcohol Depend*, *91*(2-3), 149-158. doi: 10.1016/j.drugaldep.2007.05.016
- Nakamura, M., Ueno, S., Sano, A., & Tanabe, H. (2000). The human serotonin transporter gene linked polymorphism (5-HTTLPR) shows ten novel allelic variants. *Mol Psychiatry*, *5*(1), 32-38.
- Nardi, B., Marini, A., Turchi, C., Arimatea, E., Tagliabracchi, A., & Bellantuono, C. (2013). Role of 5-HTTLPR Polymorphism in the Development of the Inward/Outward Personality Organization: A Genetic Association Study. *PLoS One*, *8*(12), e82192. doi: 10.1371/journal.pone.0082192
- Nobile, M., Greco, A., Perna, G., Colombo, P., Bianchi, V., Bellina, M., . . . Molteni, M. (2013). Effect of the serotonin transporter gene and of environment on the continuity of anxiety and depression traits throughout adolescence. *Epidemiol Psychiatr Sci*, 1-11. doi: 10.1017/S2045796013000565
- Nomura, M., Kaneko, M., Okuma, Y., Nomura, J., Kusumi, I., Koyama, T., & Nomura, Y. (2015). Involvement of serotonin transporter gene polymorphisms (5-HTT) in impulsive behavior in the Japanese population. *PLoS One*, *10*(3), e0119743. doi: 10.1371/journal.pone.0119743
- Parra, G. R., O'Neill, S. E., & Sher, K. J. (2003). Reliability of self-reported age of substance involvement onset. *Psychol Addict Behav*, *17*(3), 211-218. doi: 10.1037/0893-164X.17.3.211
- Rehorcikova, V., Nemcovska, E., Sklenarova, Z., Kallay, A., Kallayova, D., Brazinova, A., & Slana, M. (2013). Impact of family level factors on alcohol drinking in primary school children. *Cent Eur J Public Health*, *21*(4), 202-206.

- Saiz, P. A., Garcia-Portilla, M. P., Florez, G., Arango, C., Corcoran, P., Morales, B., . . . Bobes, J. (2009). Differential role of serotonergic polymorphisms in alcohol and heroin dependence. *Prog Neuropsychopharmacol Biol Psychiatry, 33*(4), 695-700. doi: 10.1016/j.pnpbp.2009.03.016
- Sanders-Bush, E. M., S.E. (2006). 5-Hydroxytryptamine (serotonin): receptor agonists and antagonists. In L. L. Brunton, J.S.; Parker, K.L. (Ed.), *Goodman and Gilman's: The Pharmacological Basis of Therapeutics* (Vol. 11, pp. 2021). New York: McGraw-Hill.
- Sellers, E. M., Higgins, G. A., & Sobell, M. B. (1992). 5-HT and alcohol abuse. *Trends Pharmacol Sci, 13*(2), 69-75.
- Simons, R. L., Lei, M. K., Beach, S. R., Brody, G. H., Philibert, R. A., & Gibbons, F. X. (2011). Social Environmental Variation, Plasticity Genes, and Aggression: Evidence for the Differential Susceptibility Hypothesis. *American Sociological Review, 76*(6), 833-912. doi: 10.1177/0003122411427580
- Takano, A., Arakawa, R., Hayashi, M., Takahashi, H., Ito, H., & Suhara, T. (2007). Relationship between neuroticism personality trait and serotonin transporter binding. *Biol Psychiatry, 62*(6), 588-592. doi: 10.1016/j.biopsych.2006.11.007
- Vaht, M., Merenakk, L., Maestu, J., Veidebaum, T., & Harro, J. (2014). Serotonin transporter gene promoter polymorphism (5-HTTLPR) and alcohol use in general population: interaction effect with birth cohort. *Psychopharmacology (Berl)*. doi: 10.1007/s00213-013-3427-8
- van der Zwaluw, C. S., Engels, R. C., Vermulst, A. A., Rose, R. J., Verkes, R. J., Buitelaar, J., . . . Scholte, R. H. (2010). A serotonin transporter polymorphism (5-HTTLPR) predicts the development of adolescent alcohol use. *Drug Alcohol Depend, 112*(1-2), 134-139. doi: 10.1016/j.drugalcdep.2010.06.001
- Watanabe, M. A., Nunes, S. O., Amarante, M. K., Guembarovski, R. L., Oda, J. M., Lima, K. W., & Fungaro, M. H. (2011). Genetic polymorphism of serotonin transporter 5-HTTLPR: involvement in smoking behaviour. *J Genet, 90*(1), 179-185.
- Wedekind, D., Herchenhein, T., Kirchhainer, J., Bandelow, B., Falkai, P., Engel, K., . . . Havemann-Reinecke, U. (2010). Serotonergic function, substance craving, and psychopathology in detoxified alcohol-addicted males undergoing tryptophan depletion. *J Psychiatr Res, 44*(16), 1163-1169. doi: 10.1016/j.jpsychires.2010.04.002