J Med Sci 2012;32(6):279-285
http://jms.ndmctsgh.edu.tw/3206279.pdf

The level of Nitric Oxide and Peak Expiration Flow Rate in Individuals with Tourette's Syndrome and Their Family Members

Chin-Bin Yeh[1,2*], Jia-Fwu Shyu[3], Chih-Hsing Hung[4], and Fan-Jung Wan[5]

[1]Division of Psychiatry; [3]Department of Biology & Anatomy;
[5]Institue of Undersea and Hyperbaric Medicine, National Defense Medical Center, Taipei;
[2]Department of Psychiatry, Tri-Service General Hospital, National Defense Medical Center, Taipei;
[4]Department of Pediatrics, Kaohsiung Medical University, Kaohsiung, Taiwan, Republic of China

Tourette's syndrome is a movement disorder that involves basal ganglia dysfunction. Since glutamatergic mechanisms have been implicated in the etiology and treatment of schizophrenia, we proposed that nitric oxide (NO), acting on the NMDA receptor as a neurotransmitter, is involved in the pathogenesis of basal ganglia in Tourette's syndrome. Twelve drug-free Tourette's syndrome subjects without a history of pediatric autoimmune disorders associated with streptococcal infection (PANDAS) or asthma were recruited. Expiration NO levels were measured and compared to 12 matched control subjects. Peak expiration flow rate was also monitored. We also measured NO levels from fathers, mothers and siblings of each of the subjects with Tourette's syndrome. There was no significant difference between the level of NO between the Tourette's syndrome subjects and the control group. However, the subjects with Tourette's syndrome had significantly higher peak expiratory flow rate than the control group. A significant correlation between NO levels in subjects with Tourette's syndrome and their mothers, but not their fathers or siblings, was also observed. Future studies are warranted to investigate the role of NO in the different phenotypes of Tourette's syndrome. The higher peak expiratory flow might be one of the pathophysiological effects with regard to the neuroanatomy of Tourette's syndrome.

Key words: nitric oxide, peak expiration flow rate, Tourette's syndrome

INTRODUCTION

Tourette's syndrome (TS) is a developmental neuropsychiatric disorder with onset in childhood, characterized by motor tics and phonic tic. The prevalence rate of TS is about 0.4% to 3.8% for children ages 5 to 18.[1,2]

The basal ganglia are considered a possible site of altered brain function in individuals with Tourette's syndrome (TS). Many studies have confirmed a critical role for dopamine (DA) in controlling basal ganglia output.[3,4] The most compelling evidence for the involvement of DA in TS comes from observations of the effects of pharmacological agents influencing the DA system.[5-8] A growing body of evidence suggests that glutamate regulates the release of DA in the central nervous system (CNS).[9], A number of recent studies have focused on glutamatergic mechanisms in the etiology and treatment of neuropsychiatric disorders.[11-14]

The potential actions of nitric oxide (NO) that have received attention from neuropsychiatric researchers include its role in relation to *N*-Methyl-D-aspartate (NMDA) receptors in the basal ganglia of individuals with movement disorders.[15] Glutamate, acting through its NMDA receptor, triples nitric oxide synthase (NOS) activity in a matter of seconds. The link between NMDA receptor activation and the generation of NO in the CNS was well established.[16] A number of studies have focused on the role of NO in movement disorders other than TS and have shown that NMDA-stimulated DA release in the nucleus accumbens can be reversed by pretreatment with NOS inhibitors.[17] A reduction of striatal NMDA receptor density has been reported in Huntington's disease.[18] The cellular protective effects for Parkinson's disease are believed to be based on the ability of amantadine to block the NMDA receptor.[19] The neuroprotective agent N-nitro-L-arginine, an NO inhibitor, is a candidate for the treatment of Parkinson's disease.[20] Recent studies have also shown that NO could represent a new form

Received: June 29, 2012; Revised: August 24, 2012; Accepted: September 12, 2012
*Corresponding author: Chin-Bin Yeh, Department of Psychiatry, Tri-Service General Hospital, National Defense Medical Center, No. 325, Sec. 2, Cheng-gong Road, Taipei 114, Taiwan, Republic of China. Tel: +886-2-87927220; Fax: +886-2-87927221; E-mail: cyeh@ndmctsgh.edu.tw

of interneuronal communication—that is, a nonsynaptic interaction without receptors—and exert an inhibitory effect on DA transporters.[21] In recent years, the autoimmune model for NO in the pathogenesis of TS has been proposed from observations of Sydenham's chorea and other CNS illnesses.[22] These researchers hypothesize that the autoimmune etiology arises from targeted dysfunction of the basal ganglia in children with PANDAS (pediatric autoimmune disorders associated with streptococcal infection). Additionally, there are recent reports of dystonia, chorea encephalopathy, and dystonic choreoathetosis occurring as sequelae of streptococcal infection.[23] NO is present in neurons, as well as the vasculature, and causes cerebrovascular vasodilation.[24-26] The stimulation of microglia via toll-like receptor 9 (TLR9) and subsequent release of NO and TNF-alpha (tumor necrosis factor-alpha) is a major source of neurotoxicity in bacterial and autoimmune brain tissue injury.[27] It is also reasonable to consider that NO is an important factor for causing inflammation in neuronal cells.[28] We therefore propose that NO may take part in intercellular communication, including neurotransmission, in the underlying pathophysiology of TS. Meanwhile, the patients with neuropsychiatric symptoms were found to have poorer lung functions.[29-31] Besides, the poor pulmonary function might reflect the level of inflammation in different tissues.[32,33] Therefore, we will compare the peak expiratory flow rate of the patients with TS to controls to investigate the pulmonary function in TS patients.

METHODS

Subjects

Twelve children with TS, in a special child psychiatric clinic, were interviewed by an experienced child psychiatrist and met the criteria for DSM-IV diagnosis of TS. Exclusion criteria for this study included serious medical illness, major sensory handicaps, major neurological disease (including a seizure disorder), previous head trauma resulting in loss of consciousness, and any current other psychiatric disorder such as major depression, schizophrenia, pervasive developmental disorder or mental retardation. Parents of the TS subjects were interviewed with the Mini-International Neuropsychological Interview (MINI) to verify that they had no current neurological or psychiatric disorders. A child psychiatrist interviewed siblings of the TS subjects to check for DSM-IV diagnosis.

Twelve age-matched control children from the pediatric clinic in the same medical center were recruited to form a control group. Control subjects were without tic, TS, obsessive–compulsive disorder (OCD) or other exclusion criteria described above. The TS subjects and their families, as well as the control group, were also free from asthma and acute upper respiratory tract infection. All parents provided written informed consent.

Nitric oxide assay

The peak expiratory flow rate (PEFR) was measured three times in the morning about 10 A.M. using a peak flow meter (Astech Co, Port Washington, NY). The maximal value was analyzed. The eNO (exhaled nitric oxide) level was measured using a fast-response chemiluminescence analyzer (NOA 280; Seivers Instrument Inc, Boulder, Colo) with the validated, single-breath technique.[34] The nose was obstructed, and subjects were asked to inhale to total lung capacity and then exhale slowly through the Teflon side arm attached to the sampling port. Subjects exhaled against a fixed resistor at a constant mouth pressure (15 cm H_2O mmHg) corresponding to expiratory flow (75 mL/s), a maneuver that closes the velum of the posterior nasopharynx and avoids nasal NO contamination. To maintain a steady flow, mouth pressure was displayed on a computer screen as a prompt for subjects. The eNO values were recorded from the plateau at the end of exhalation. Measurements were only recorded when three measurements showed less than 10% variability. Most patients could complete three measurements without further trials. A mean was then calculated to represent these values.

Statistics

The independent t test was done for comparison of the age, height, body weight, the level of nitric oxide concentration, and the PEFR between groups. Pearson correlation analysis was done to investigate the correlationship of the level of nitric oxide between TS subjects and their family members. $P < 0.05$ was taken as significant.

RESULTS

Eight of the 12 TS subjects were comorbid for attention-deficit hyperactivity disorder (ADHD), and two of the TS subjects were comorbid for OCD. Seven fathers, 12 mothers and five siblings of TS subjects were interviewed and received an NO assay. None of the parents or the siblings met the criteria for DSM-IV diagnosis and all were without a current or past history of tic symptoms.

The level of NO (pars per billion, p.p.b.) was not significantly different between TS subjects and control

Table 1 Comparison of the level of nitric oxide and peak expiratory flow rate (PEFR) between TS subjects and controls

	TS subjects (N=12)	Controls (N=12)	t	p
Age (year)	9.3±2.6	8.1±1.1	1.5	0.16
Height (cm)	137.3±17.3	132.5±14.0	–0.75	0.46
Body weight (kg)	35.6±13.3	30.3±9.8	–1.05	0.31
Nitric oxide (p.p.b)	10.2±3.8	9.4±4.0	0.46	0.65
PEFR (L/min)	261±87	187±40	2.68	0.014*

Data shown as mean ±SD. *P< 0.05 significance difference between groups.

Table 2 Correlation of nitric oxide levels of TS subjects and family members.

	Nitric oxide level (p.p.b)	Correlation (c.c.)	p
Fathers of TS cases (N = 7)	9.9±4.1	–0.051	0.914
Mothers of TS cases (N = 12)	9.1±4.2	0.795	0.002*
Siblings of TS cases (N = 5)	8.7±3.3	0.335	0.582

Data shown as mean ±SD. *P< 0.05 indicated significant correlation of the level of NO between the TS subjects and mothers of TS cases.

subjects. There was also no significant difference in NO levels between family members. However, the PEFR of the TS subjects was significantly higher than that of the control group (Table 1). The correlation between the level of NO of TS subjects and their family members was analyzed. Interestingly, there was a significant correlation between NO levels of TS subjects and their mothers (c.c. = 0.795, p = 0.002) but not their fathers (c.c. = –0.051, 0 = 0.914) or siblings (c.c. = 0.335, p = 0.582) (Table 2). However, no significant correlation was observed between the PEFRs of TS subjects and family members. Furthermore, there was no significant correlation between the PEFR and the level of NO among TS subjects (c.c. = 0.121, p = 0.708) and their father (c.c. = –0.298, p = 0.516), mother (c.c. = 0.207, p = 0.518), or sibling (c.c. = –0.254, p = 0.681).

DISCUSSION

In contrast to our hypothesis, we failed to identify NO as a biological marker for TS. We still consider this preliminary data worth reporting, as no studies have focused on NO levels in TS patients. It is difficult for most researchers to study the immunological functions of the subjects with neuropsychiatric disorders. The exhaled NO might provide another way to explore the immunological profile of children with neuropsychiatric disorders. As shown in our results, the NO level of TS patients was not significantly different from that in the control group. Thus, NO might not be a state marker for children with TS. Nevertheless, the role of NO as a trait marker in the dynamic interactions with stress, infection, environmental factors might warrant further studies. In addition, there are different isoforms of NO synthase in different localizations in the brain.[35] The NO in lung might not represent the level of NO in the CNS although there was the relationship between the level of exhaled NO and the symptoms severity of mental disorders in elder population but not in the children group.[36] Moreover, the role of NO in TS might through other neurotransmitters such as glutamate since animal studies indicate that glutamate enhances extracellular DA in the medial preoptic area of the hypothalamus, via NO activity, which facilitates male sexual behavior, and is proposed to be similar to the obsessive symptoms of TS.[37] Significantly reduced levels of glutamate have been observed in the medial globus pallidus of brains from four TS individuals.[38] Glutamatergic drugs exacerbate symptomatic behavior in a comorbid transgenic model of TS and OCD.[39] Another possible role for NO in the pathogenesis of TS is hypoxic-ischemia brain injury and oxidative stress. NO could combine with O_2– and H_2O_2 to produce peroxynitrite, which could react with protein tyrosine residues to produce nitrotyrosine. Peroxynitrite could also cause lipid peroxidation and DNA chain breaks, which lead to damage of the developing brain.[40,41] The involvement of NO in the pathogenesis of TS might be through the autoimmune mechanisms since many studies have shown that NO is involved in the immunological pathogenesis of neuropsychiatric disorders[42-45] and enhanced expression of the constitutive and inducible forms of NOS occurs in an animal model of autoimmune encephalomyelitis.[46] Although NO is stable in oxygen-free water, it is labile and lasts only a few seconds in biological fluids because of its inactivation by superoxides. NO dissolves in both aqueous and lipid media and readily diffuses from its site of synthesis across the cytosol or cell membrane, affecting targets in the same cell or in nearby neurons, glia, and vasculature.[47] Different inactivation and diffusion of NO in different individuals may account for the lack of significance difference between NO levels in TS and control subjects. We measured the downstream product in the NO production process, which may be affected by many factors, such as exercise, stress, smoking, and infection.[28,48-51] In the

future, we plan to collect more samples and control the above possible factors to further investigate the role of NO in TS.

As shown in Table 2, NO levels significantly correlated between TS subjects and their mothers. In contrast, this correlation was not found between TS subjects and fathers or siblings. Because of the small sample size, this result warrants further study although there were family studies found that the genetic factors play an important role in the transmission and expression of TS.[52,53]

Interestingly, we found that the PEFR was higher in TS subjects. The PEFR has been correlated to age, height and weight.[54,55] As shown in Table 1, there was no significant difference in age, height and weight between TS and control groups. Previous studies have shown that PEFR correlates with cognitive performance, including tests of similarities, naming, spatial recognition, memory, and figure drawing.[54] Measurement of the PEFR has been developed to assess airflow obstruction in patients with cerebral palsy or severe mental retardation, as well as in young children.[56] This suggests that the PEFR might be associated with the underlying neurophysiology of TS.[57-61] In contrast to other movement disorders, such as Huntington's disease and Parkinson's disease, physical examination in TS subjects did not provide direct clues to the neuroanatomic localization of the dysfunction. The mechanism for the increased PEFR in TS subjects is unknown, but we hypothesize that it involves neuroanatomic pathways implicated in the motor excitatory/inhibitory regulation. The abundance of basal ganglia projections to the frontal lobe emphasizes the role of the basal ganglia complex in premotor and prefrontal function. The basal ganglia also have considerable output to various brainstem structures, including the pedunculopontine nucleus. The descending projections from the basal ganglia signify a role for them in a number of basic aspects of motor control, such as muscle tone, posture and balance, as well as innate movement patterns such as locomotion, mastication, etc. Abnormalities of these brain structures may account for the significantly higher PEFR of the children with TS compared with the control group.[62]

In conclusion, the patients with TS have different PEFR but have no different eNO level when compared to controls. Future studies are warranted to investigate the role of other immunological or inflammatory-related factors other than eNO in the pathobiology of children with TS.

ACKNOWLEDGMENT

This study was supported by the NSC 100-2314-B-016-034-MY3, TSGH-C96-60 and B981126-2, Taipei, Taiwan, Republic of China.

DISCLOSURE

All authors declare that this study has no conflict of interest.

REFERENCES

1. Lin H,Yeh CB, Peterson BS, Scahill L, Grantz H, Findley DB, Katsovich L, Otka J, Lombroso PJ, King RA, Leckman JF. Assessment of symptom exacerbations in a longitudinal study of children with Tourette's syndrome or obsessive-compulsive disorder. J Am Acad Child Adolesc Psychiatry 2002;41:1070-1077.
2. Leckman JF,Yeh CB, Cohen DJ. Tic disorders: when habit forming neural systems form habits of their own? Zhonghua Yi Xue Za Zhi (Taipei) 2001;64:669-692.
3. Graybiel AM. Basal ganglia: new therapeutic approaches to Parkinson's disease. Curr Biol 1996;6:368-371.
4. Young AB, Penney JB. Neurochemical anatomy of movement disorders. Neurol Clin 1984;2:417-433.
5. Cheon KA, Ryu YH, Namkoong K, Kim CH, Kim JJ, Lee JD. Dopamine transporter density of the basal ganglia assessed with [123I]IPT SPECT in drug-naive children with Tourette's disorder. Psychiatry Res 2004;130:85-95.
6. Gilbert DL, Dure L, Sethuraman G, Raab D, Lane J, Sallee FR. Tic reduction with pergolide in a randomized controlled trial in children. Neurology 2003;60:606-611.
7. Golden GS. Gilles de la Tourette's syndrome following methylphenidate administration. Dev Med Child Neurol 1974;16:76-78.
8. Riddle MA, Leckman JF, Anderson GM, Hardin MT, Ort SI, Towbin KE, Shaywitz BA, Cohen DJ. Assessment of dopaminergic function in children and adults: long and brief debrisoquin administration combined with plasma homovanillic acid. Psychopharmacol Bull 1987;23:411-414.
9. Hallett PJ, Standaert DG. Rationale for and use of NMDA receptor antagonists in Parkinson's disease. Pharmacol Ther 2004;102:155-174.
10. Whitton PS. Glutamatergic control over brain dopamine release in vivo and in vitro. Neurosci Biobe-

hav Rev 1997;21:481-488.
11. Carlsson ML, Carlsson A, Nilsson M. Schizophrenia: from dopamine to glutamate and back. Curr Med Chem 2004;11:267-277.
12. McCullumsmith RE, Clinton SM, Meador-Woodruff JH. Schizophrenia as a disorder of neuroplasticity. Int Rev Neurobiol 2004;59:19-45.
13. O'Neill MJ, Bleakman D, Zimmerman DM, Nisenbaum ES. AMPA receptor potentiators for the treatment of CNS disorders. Curr Drug Targets CNS Neurol Disord 2004;3:181-194.
14. Tsai G, Lane HY, Yang P, Chong MY, Lange N. Glycine transporter I inhibitor, N-methylglycine (sarcosine), added to antipsychotics for the treatment of schizophrenia. Biol Psychiatry 2004;55:452-456.
15. Karatinos J, Rosse RB, and Deutsch SI. The nitric oxide pathway: potential implications for treatment of neuropsychiatric disorders. Clin Neuropharmacol 1995;18:482-499.
16. Garthwaite J, Southam E, Boulton CL, Nielsen EB, Schmidt K, Mayer B. Potent and selective inhibition of nitric oxide-sensitive guanylyl cyclase by 1H-[1,2,4]oxadiazolo[4,3-a]quinoxalin-1-one. Mol Pharmacol 1995;48:184-188.
17. Segieth J, Fowler L, Whitton P, Pearce B. Nitric oxide-mediated regulation of dopamine release in the hippocampus in vivo. Neuropharmacology 2000;39:571-577.
18. Cepeda C, Ariano MA, Calvert CR, Flores-Hernandez J, Chandler SH, Leavitt BR, Hayden MR, Levine MS. NMDA receptor function in mouse models of Huntington's disease. J Neurosci Res 2001;66:525-539.
19. Crosby N, Deane KH, Clarke CE. Amantadine in Parkinson's disease. Cochrane Database Syst Rev 2003;1:CD003468.
20. Krzascik P, Kostowski W. Nitric oxide donors antagonize N-nitro-L-arginine and haloperidol catalepsy: potential implication for the treatment of Parkinsonism? Pol J Pharmacol 1997;49:263-266.
21. Kiss JP, Zsilla G, Vizi ES. Inhibitory effect of nitric oxide on dopamine transporters: interneuronal communication without receptors. Neurochem Int 2004;45:485-489.
22. Hoekstra PJ, Anderson GM, Limburg PC, Korf J, Kallenberg CG, Minderaa RB. Neurobiology and neuroimmunology of Tourette's syndrome: an update. Cell Mol Life Sci 2004;61:886-898.
23. Snider LA, Swedo SE. Post-streptococcal autoimmune disorders of the central nervous system. Curr Opin Neurol 2003;16:359-365.
24. Akerman S, Williamson DJ, Kaube H, Goadsby PJ. Nitric oxide synthase inhibitors can antagonize neurogenic and calcitonin gene-related peptide induced dilation of dural meningeal vessels. Br J Pharmacol 2002;137:62-68.
25. Dugas N, Delfraissy JF, Tardieu M. Immune regulatory role of nitric oxide within the central nervous system. Res Immunol 1995;146:707-710.
26. Toda N, Okamura T. The pharmacology of nitric oxide in the peripheral nervous system of blood vessels. Pharmacol Rev 2003;55:271-324.
27. Iliev AI, Stringaris AK, Nau R, Neumann H. Neuronal injury mediated via stimulation of microglial toll-like receptor-9 (TLR9). Faseb J 2004;18:412-414.
28. Evans SM, Whittle BJ. Role of bacteria and inducible nitric oxide synthase activity in the systemic inflammatory microvascular response provoked by indomethacin in the rat. Eur J Pharmacol 2003;461:63-71.
29. Katon WJ, Richardson L, Lozano P, McCauley E. The relationship of asthma and anxiety disorders. Psychosom Med 2004;66:349-355.
30. Allaire JC, Tamez E, Whitfield KE. Examining the association between lung functioning and cognitive performance in African American adults. J Aging Health 2007;19:106-122.
31. van Milligen BA, Lamers F, de Hoop GT, Smit JH, Penninx BW. Objective physical functioning in patients with depressive and/or anxiety disorders. J Affect Disord 2011;131:193-199.
32. Yoon SH, Choi NW, Yun SR. Pulmonary dysfunction is possibly a marker of malnutrition and inflammation but not mortality in patients with end-stage renal disease. Nephron Clin Pract 2009;111:c1-6.
33. Liu J, Wan L, Sheng CJ, Xie XL. Xi Bao Yu Fen Zi Mian Yi Xue Za Zhi. The correlative study on pulmonary function changes and Th1/Th2 cells & regulatory T cells in adjuvant arthritis rats. 2011;27:56-60.
34. Silkoff PE, McClean PA, Slutsky AS, Furlott HG, Hoffstein E, Wakita S, Chapman KR, Szalai JP, Zamel N. Marked flow-dependence of exhaled nitric oxide using a new technique to exclude nasal nitric oxide. Am J Respir Crit Care Med. 1997;155:260-267.
35. Billiar TR. Nitric oxide. Novel biology with clinical relevance. Ann Surg 1995;221:339-349.
36. Gramiccioni C, Carpagnano GE, Spanevello A, Turchiarelli V, Cagnazzo MG, Foschino Barbaro MP. Airways oxidative stress, lung function and cogni-

tive impairment in aging. Monaldi Arch Chest Dis 2010;73:5-11.
37. Dominguez JM, Muschamp JW, Schmich JM, Hull EM. Nitric oxide mediates glutamate-evoked dopamine release in the medial preoptic area. Neuroscience 2004;125:203-210.
38. Anderson GM, Pollak ES, Chatterjee D, Leckman JF, Riddle MA, Cohen DJ. Brain monoamines and amino acids in Gilles de la Tourette's syndrome: a preliminary study of subcortical regions. Arch Gen Psychiatry 1992;49:584-586.
39. McGrath MJ, Campbell KM, Parks CR, Burton FH. Glutamatergic drugs exacerbate symptomatic behavior in a transgenic model of comorbid Tourette's syndrome and obsessive-compulsive disorder. Brain Res 2000;877:23-30.
40. Hensley K, Tabatabaie T, Stewart CA, Pye O, Floyd RA. Nitric oxide and derived species as toxic agents in stroke, AIDS dementia, and chronic neurodegenerative disorders. Chem Res Toxicol 1997;10:527-532.
41. Veltkamp R, Rajapakse N, Robins G, Puskar M, Shimizu K, Busija D. Transient focal ischemia increases endothelial nitric oxide synthase in cerebral blood vessels. Stroke 2002;33:2704-2710.
42. Chrapko WE, Jurasz P, Radomski MW, Lara N, Archer SL, Le Melledo JM. Decreased platelet nitric oxide synthase activity and plasma nitric oxide metabolites in major depressive disorder. Biol Psychiatry 2004;56:129-134.
43. Ozcan ME, Gulec M, Ozerol E, Polat R, Akyol O. Antioxidant enzyme activities and oxidative stress in affective disorders. Int Clin Psychopharmacol 2004;19:89-95.
44. Sogut S, Zoroglu SS, Ozyurt H, Yilmaz HR, Ozugurlu F, Sivasli E, Yetkin O, Yanik M, Tutkun H, Savaş HA, Tarakçioğlu M, Akyol O. Changes in nitric oxide levels and antioxidant enzyme activities may have a role in the pathophysiological mechanisms involved in autism. Clin Chim Acta 2003;331:111-117.
45. Yanik M, Vural H, Kocyigit A, Tutkun H, Zoroglu SS, Herken H, Savas HA, Köylü A, Akyol O. Is the arginine-nitric oxide pathway involved in the pathogenesis of schizophrenia? Neuropsychobiology 2003;47:61-65.
46. Kim S, Moon C, Wie MB, Kim H, Tanuma N, Matsumoto Y, Shin T. Enhanced expression of constitutive and inducible forms of nitric oxide synthase in autoimmune encephalomyelitis. J Vet Sci 2000;1:11-17.
47. Baranano DE, Ferris CD, Snyder SH. Atypical neural messengers. Trends Neurosci 2001;24:99-106.
48. Higashi Y, Yoshizumi M. Exercise and endothelial function: role of endothelium-derived nitric oxide and oxidative stress in healthy subjects and hypertensive patients. Pharmacol Ther 2004;102:87-96.
49. Hoyt JC, Robbins RA, Habib M, Springall DR, Buttery LD, Polak JM, Barnes PJ. Cigarette smoke decreases inducible nitric oxide synthase in lung epithelial cells. Exp Lung Res 2003;29:17-28.
50. Martin MC, Gomez-Jimenez J, Esteban F, Sauri R, Mourelle MI, Salgado A. Cytokines and nitric oxide in streptococcal toxic shock syndrome. Med Clin (Barc) 1995;104:458-460.
51. Masood A, Banerji B, Vijayan VK, Ray A. Pharmacological and biochemical studies on the possible role of nitric oxide in stress adaptation in rats. Eur J Pharmacol 2004;493:111-115.
52. Kano Y, Ohta M, Nagai Y, Pauls DL, Leckman JF. A family study of Tourette syndrome in Japan. Am J Med Genet 2001;105: 414-421.
53. Pauls DL. An update on the genetics of Gilles de la Tourette syndrome. J Psychosom Res 2003;55:7-12
54. Cook NR, Albert MS, Berkman LF, Blazer D, Taylor JOP, Hennekens CH. Interrelationships of peak expiratory flow rate with physical and cognitive function in the elderly: MacArthur Foundation studies of aging. J Gerontol A Biol Sci Med Sci 1995;50:317-323.
55. Host A, Host AH, Ibsen T. Peak expiratory flow rate in healthy children aged 6–17 years. Acta Paediatr 1994;83:1255-1257.
56. Vink GR, Arets HG, van der Laag J, van der Ent CK. Impulse oscillometry: a measure for airway obstruction. Pediatr Pulmonol 2003;35:214-219.
57. Lombroso PJ, Mack G, Scahill L, King RA, Leckman JF. Exacerbation of Gilles de la Tourette's syndrome associated with thermal stress: a family study. Neurology 1991;41:1984-1987.
58. Nomura Y, Fukuda H, Terao Y, Hikosaka O, Segawa M. Abnormalities of voluntary saccades in Gilles de la Tourette's syndrome: pathophysiological consideration. Brain Dev 2003;25 Suppl 1:48-54.
59. Saka E, Graybiel AM. Pathophysiology of Tourette's syndrome: striatal pathways revisited. Brain Dev 2003;25 1:15-19
60. Scahill L, Lombroso PJ, Mack G, Van Wattum PJ, Zhang H, Vitale A, Leckman JF. Thermal sensitivity in Tourette's syndrome: preliminary report. Percept Mot Skills 2001;92:419-32.
61. Segawa M. Neurophysiology of Tourette's syn-

drome: pathophysiological considerations. Brain Dev 2003;25 Suppl 1:62-69.
62. Peterson BS, Gore JC, Riddle MA, Cohen DJ, Leckman JF. Abnormal magnetic resonance imaging T2 relaxation time asymmetries in Tourette's syndrome. Psychiatry Res 1994;55:205-221.