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Protecting Circulation with Enzyme Nutrition

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ABSTRACT

Cardiokinase is a potent fibrinolytic enzyme with the potential for fighting cardiovascular diseases, hence referred so often. It is extracted and highly purified from a traditional Japanese food called Natto which is made from fermented soybeans and is also called nattokinase. Improper blood clotting or failure of clots to properly dissolve can lead to serious health consequences. Thrombotic diseases typically include cerebral hemorrhage, cerebral infarction, cardiac infarction and angina pectoris, and also include diseases caused by blood vessels with lowered flexibility, including senile dementia and diabetes. Pharmacologically Cardiokinase has a safe profile as the key agent for restoration of vasculature health and cure of all thrombotic diseases. Cardiokinase also termed "super health food" has caused world wide attention not only for its unique flavor and rich nutrition, but also due to a variety of health functions.

Key words: Cardiokinase, nattokinase, natto, fibrin, blood clots.

INTRODUCTION

Nattokinase was discovered in 1980 by Dr Hiroyuki Sumi, researcher at Chicago University after testing over 173 natural foods as potential thrombolytic agents, searching for a natural agent that could effectively dissolve thrombus allied with cardiac and cerebral infarction[43-45]. Nattokinase was discovered in Natto, a fermented cheese-like food that has been used in Japan for over 1000 year. Natto is a traditional Japanese food made of soybeans . To prepare the beans are cooked and then by the action of the bacterium *Bacillus subtilis ssp. natto* fermented[9, 17] . During this process is formed a slimy, stringy substance to the beans. In the traditional method of preparation are the bacteria from rice straw , into which the beans are wrapped[3]. In the modern manufacturing process, the bacteria cultures inoculated with beans, so that the use of rice straw is no longer necessary[21]. The botanical source for Nattokinase is Glycine max(L.)Merr. It appears as a yellow-white fine powder[22].

Cardiokinase is a particularly potent treatment because it enhances the body's natural ability to fight blood clots in several different ways and has many benefits including convenience of oral administration, confirmed efficacy, prolonged effects, cost effectiveness, and can be used preventatively[1,3,4]. It is a naturally occurring, food dietary supplement that has demonstrated stability in the gastrointestinal tract. The properties of Cardiokinase closely resemble those properties of plasmin so it dissolves fibrin directly[7,8]. More importantly, it also enhances the body's production of both plasmin and other clot dissolving agents.

Like other proteases, nattokinase has potent anti-inflammatory activity as well as several other beneficial properties. However, probably the most important aspect of nattokinase is its fibrinolytic activity. To understand fibrinolytic activity, first one must understand the basic process of coagulation or clotting of blood. In damaged tissue, the broken blood vessel releases a compound called

thromboplastin. At the same time platelets adhere to the broken edges of the vessel and disintegrate, releasing platelet factor 3. Both of these react with protein factors and calcium ions to form prothrombin activator. Once the prothrombin activator is formed, the process from prothrombin to a clot follows.[11] Of course, clotting forms an important function in tissue repair. However, a clot in the blood stream (a thrombus) can be very dangerous, even fatal.

Research with nattokinase demonstrates that it may help avoid or reduce the likelihood of deep vein thrombosis, cardiac infarction, pulmonary emboli and stroke. It appears to accomplish this via its fibrinolytic, anti-inflammatory and modulating effect on blood pressure. Studies on hypertension demonstrate an average drop of10.9%in Systolic Blood Pressure and a 9.7 percent drop in Diastolic Blood Pressure.[6,7,21]

In vitro and in vivo studies have consistently demonstrated the potent fibrinolytic effect of Nattokinase The fibrinolytic activity of nattokinase, the various medicinal uses and case studies are described herewith.

Prevention and Treatment of Heart Conditions

In the interest of preventive medicine, proteolytic enzymes can be used as interventional medicines that serve to inhibit over activity of fibrin[15,16]. One particular enzyme, known as nattokinase, has demonstrable fibrinolytic activity. Nattokinase is derived from a Japanese food known as natto, a preparation of soybeans that has undergone fermentation with a bacterium known as *Bacillus subtilis natto*[5,6]. Thought to be produced specifically from this process of fermentation, nattokinase is not derived directly from other soy-based foods[23].

Nattokinase causes mild enhancement of fibrinolysis in plasma, as evidenced by its effect on fibrinolytic parameters and production of tissue plasminogen activator, a potent thrombolytic agent that causes fibrinolysis at the site of a blood clot. Therapeutic Enzymes using the Body's Helpers as Healers Nattokinase is thought to work by stopping plasminogen activator inhibitor[26-29]. That is, nattokinase works by preventing the blockage of the formation of plasminogen activator, thereby allowing for the degradation of the clotting process. The fibrinolytic activity of nattokinase is fourfold that of plasmin, a main fibrinolytic enzyme found in the body.

Nattokinase is particularly effective because it enhances the body's natural ability to fight blood clots in several different ways. It dissolves fibrin directly and appears to enhance the body's natural production of both plasmin and other clot-dissolving enzymes like urokinase. An in vivo study was undertaken to demonstrate the thrombolytic activity of nattokinase, plasmin and elastase on an induced clot in the common carotid artery of laboratory rats. The results indicate that the thrombolytic activity of nattokinase is stronger than that of plasmin or elastase in vivo in this model.

In animal studies, nattokinase can reduce markedly the thickening of blood vessel walls that normally occurs following an injury to the endothelium. In addition, nattokinase leads to dissolution of clots that build inside vessel walls as responses to injuries. These actions suggest that nattokinase can be used to treat and prevent atherosclerosis because of its fibrinolytic activity at the blood-vessel wall[31,43].

As a result of scientific research, nattokinase is available as a dietary supplement for human use. Because nattokinase is considered a systemic enzyme, it is important to demonstrate its absorption from the GI tract. An in vivo study of the duodenal absorption of nattokinase in rats showed intact absorption of the enzyme. This was based upon the subsequent degradation of plasma fibrinogen clearly demonstrating transport of nattokinase across the intestinal tract. The action of nattokinase on the cleavage of fibrinogen in the plasma was remarkably prolonged being present in plasma samples drawn 3 to 5 hours after administration of the enzyme.When taken orally in humans, nattokinase retains its activity, by escaping degradation during the digestive process and has been shown to raise the level of fibrinolytic activity significantly for several hours after dosing[41-45].

Other applications of nattokinase include treating cardiovascular diseases, such as stroke, angina, deep-vein thrombosis, atherosclerosis,

venous stasis, peripheral vascular disease, and claudication[44]. Arteriosclerosis, excessive clotting, and inflammation are routine in developing arterial plaques. Enzyme therapy digests the fibrin and reverses the inflammation, which decreases the size of the artery-obstructing plaques[40,47].

We have noted that symptoms of angina, impaired blood flow to the brain, and poor circulation to the legs often disappear with enzymatic treatment for cardiovascular conditions. The gentle, yet effective use of nattokinase for preventing cardiovascular diseases makes this an optimal choice from preventive and treatment perspectives ditions[33-36,50]. Combination with other anti coagulative therapies or drugs should be approached with great caution, however. Nattokinase is widely available today; one particular version of this enzyme is marketed as a preventive treatment for deep-vein thrombosis on long flights[45-46,49].

Case Studies

Throughout the 1990s, several researchers reported laboratory findings on various methods of isolating and purifying nattokinase and documented its fibrinolytic activity in-vitro. In 1990, Sumi et al., reported the effectiveness of nattokinase capsules in dissolving experimentally induced thrombi in dogs.[12] There have been at least three studies demonstratingthe activity of nattokinase in rats.[13–15]

The process of forming a clot is complex and involves several enzymes. However, the body mainly produces one central enzyme for dissolving a clot, plasmin. It happens that the properties of nattokinase are very similar to plasmin. An in vivo study was undertaken to demonstrate the thrombolytic activity of nattokinase, plasmin and elastase on an induced clot in the common carotid artery of laboratory rats. The results indicate that the thrombolytic activity of nattokinase is stronger than that of plasmin or elastase in vivo in this model[47,50].

A study conducted with natto on 12 healthy adults (6 men and 6 women, between the ages of 21 and 55) sought to demonstrate fibrinolytic activity. The volunteers were given 200 grams of natto (the food) before breakfast, then their fibrinolytic activity was tested over time. The results indicate natto generates an increased ability to dissolve blood clots. As a control, researchers later fed the same amount of boiled soybeans to the same volunteers and tracked their fibrinolytic activity. The tests showed no significant change.[2]The accumulation of fibrin in blood vessels significantly increases the likelihood of thrombosis formation resulting in a cardiovascular event. For thrombolytic therapy, microbial fibrinolytic enzymes are now much more accepted. The physiochemical properties of this enzyme is becoming well characterized and its effectiveness in thrombolysis in vivo has been furtheridentified.[12, 13]

A fascinating study was conducted to measure the effect of nattokinase in the prevention of deep vein thrombosis and superficial vein thrombosis on extended flights of 7-8 hours on high-risk individuals. The nattokinase had no thromboses. The placebo group had 5 deep vein thromboses and 2 superficial vein thromboses or 7.6% of 94 individuals. After the flight, the degree of edema was increased by 12% in the placebo group and decreased and the nattokinase group decreased by 15%. The authors conclude that nattokinase was effective in reducing thrombotic events and in controlling edema in high-risk subjects on long flights.[14]In a study following 238 high-risk patients for stroke, the authors demonstrated a significant relationship between increased common carotid artery intima-media thickness (CCA-IMT) and the occurrence of myocardial infarction and stroke. For each increment of 0.1 m min CCA-IMT the probability of experiencing recurrent stroke increased by 18.0%.[15]

A study with rat femoral artery investigating the effect of dietary supplementation with natto extracts on the intimal thickening of arteries was conducted. It was shown that dietary natto extract supplementation suppressed intimal thickening (0.06 +/- 0.01; $P < 0.05$) compared with the control group. These findings suggest that natto extracts, because of their thrombolytic activity, suppress intimal thickening after vascular injury as a result of the inhibition of thrombi formation.[16,18]

In another study by the same group, they demonstrated that nattokinase (NK) inactivates

plasminogen activator inhibitor type 1, which in turn potentiates fibrinolytic activity. Further, they investigated the effect on neo intima formation and on thrombolysis at the site of endothelial injury. They conclude that dietary natto-extracts supplementation suppresses intimal thickening produced by endothelial injury in rat femoral artery. These effects are at least partially attributable to NK, which showed enhanced thrombolysis near the vessel wall.[17,18,19]At least 20 or more proteins form amyloid fibrils, which are causative agents in Alzheimer's disease, prion disease, and systematic amyloidosis. Improving amyloid clearance is a major target of the therapy for these diseases. Degradation of amyloids may help prevent or at least alleviate these diseases. In this study, the authors demonstrated the amyloid-degrading ability of nattokinase. They also determined that this ability is shared by proteinase K and subtilisin, but not by trypsin or plasmin.[20]

Furthermore, animal studies have shown that nattokinase is completely safe and nontoxic.[2, 25] According to unpublished studies, acute oral toxicity studies show that extremely high doses of nattokinase are not lethal to rats. In addition, a 90-day trial at a wide range of doses demonstrated that nattokinase does not cause gross histopathologic changes.

While more research is needed, it seems clear that nattokinase offers significant benefits to those who are at risk for a cardiovascular event. Indeed, nattokinase may reduce the risk of deep vein thrombosis, myocardial infarction, pulmonary emboli and stroke. Further, its effect to reduce the thickness of the intima-media of arterial walls may result in reduced blood pressure as well as other risk factors like atherosclerosis. The demonstrated reduction of the aggregation of red blood cells and lowered whole blood viscosity leads one to conclude that nattokinase helps provide vascular conditions that are less likely to produce blood clots. Additional research is needed in the area of amyloid degradation. Nattokinase or related compounds may offer hope for the prevention and treatment of amyloid-related diseases.

One of the more compelling human trials involved 12 healthy Japanese volunteers. 12 each participant was initially given 200 g of natto once per day before break fast, followed by periodic plasma blood testing. After a 2-week interval, the same individuals were given 200 g of boiled soybeans once per day before break fast. Again, plasma blood was collected. There was just a slight are ability in the results of the control groups who were given boiled soybean. Conversely, the natto-treated group showed a clear shortening of euglobulin lysis time and elevations of euglobulin fibrinolytic activity after a single administration of natto.

CONCLUSION

The traditional Japanese food Natto has been used safely for over 1000 years. The potent fibrinolytic enzyme nattokinase appears to be safe based upon the long-term traditional use of this food. Medical uses include prevention of heart attacks, strokes, senility, osteoporosis, cancer; antibiotic effects, anti ageing effects and many others. Nattokinase has many other benefits including convenience of oral administration, confirmed efficacy, prolonged effects, cost effectiveness, and can be used preventatively. It is a naturally occurring, food based dietary supplement that has demonstrated stability in the gastrointestinal tract, as well as to changes in pH and temperature.

Stressful era of modernization has led to high rates of cardiovascular diseases; thence it would then seem prudent to add this effective natural product to our heart health preventive arsenal as more recently, both clinical and non-clinical studies have demonstrated that Nattokinase supports heart health and promotes healthy circulation. More recent uses include meal replacements, medical foods, tablets, capsules, cold extruded bars, dry mixes and yoghurt drinks and cheese manufacture.

All prior epidemiologic and clinical research points to nattokinase's effectiveness and safety for managing a wide range of diseases, including hypertension, atherosclerosis, coronary artery disease (such as angina), stroke, and peripheral vascular disease. Evidence from long-term use at high doses in Japanese people points to nattokinase as a safe nutrient that acts as a very powerful

fibrinolytic agent. However, more research is needed on humans to verify the predicted safety of formulated extracts that deliver high concentrations of nattokinase while eliminating naturally occurring vitamin K. Hereby, this paper paraphrases the assorted therapeutic and medicinal uses of nattokinase.

REFERENCES

1. Tai M W, Sweet B V, Nattokinase for prevention of thrombosis. *Am J Health Syst Pharm*, **63**(12): 1121-1123 (2006).
2. Sumi, H., Hamada, H., Tsushima, H.,Mihara, H., Muraki, H. A novel fibrinolytic enzyme (nattokinase) in the vegetable cheese natto: A typical and popular soybean food in the Japanese diet. *Experientia* **43(**10): 1110-1111 (1987).
3. Sumi , H . Accumulation of Vitamin K (Menaquinone-7) in Plasma after Ingestion of Natto and Natto Bacilli (*B. subtilis natto*), *Kurashiki*, 1–5 (1998).
4. Yong Peng, Xiaojuan Yang,and Yizheng Zhang, Microbial fibrinolytic enzymes: an overview of source, production, properties, and thrombolytic activity in vivo, *Applied Microbiology and Biotechnology*, **69**(2): 126-132 (2005).
5. Osborn T M, Intravenous thrombolytic therapy for stroke: A review of recent studies and controversies. *Ann Emerg Med* **34**: 244-255 (1999).
6. Yasuhiro Suzuki, Kazunao Kondo, Dietary supplementation with fermented soybeans suppresses intimal thickening, *Nutrition*, **19**(3): 261-264 (2003).
7. Chien-Hsun Hsia, Ming-Ching Shen, Jen-Shiou Lin, Yao-Ke Wen, Kai-Lin Hwang, Thau-Ming Cham, Nae-Cherng Yang, Nattokinase decreases plasma levels of fibrinogen, factor VII, and factor VIII in human subjects, *Nutrient Research*, **29**(3): 145-220 (2009).
8. Collins, R, Aspirin, heparin and fibrinolytic therapy in suspected acute myocardial infarction. *N Engl J Med* **336,** 847–846, (1997).
9. Yoshinori, M.; Wong, A. H. K.; Jiang, B. Fibrinolytic enzymes in Asian traditional fermented foods, *Food Res. Int.*,**38**, 243–250 (2005).
10. Harenberg, J., Giese, C., Zimmermann, R. Effect of dried garlic on blood coagulation, fibrinolysis, platelet aggregation and serum cholesterol levels in patients with hyperlipoproteinemia. *Atherosclerosis* **74**,3,247–249 (1988).
11. C H Hsia, Nattokinase decreases plasma levels of fibrinogen, factor VII, and factor VIII in human subjects, *Nutr. Res.* **29**,190–196 (2009).
12. Sumi, H., Hamada, H., Nakanishi, K.,Hirantani, H. Enhancement of the fibrinolytic activity in plasma by oral administration of nattokinase. *Acta Haematol* **84**: 139-143 (1990).
13. Ohkuro, I., Komatsuzaki, T, The level of serum lysozyme activity in animals fed a diet containing natto bacilli [in Japanese],*MedBio* **102**: 335-337 (1981).
14. Fujita, M., Hong, K., Ito, Y., Fujii, R., Kariya,K., Nishimuro, S. Thrombolytic effect of nattokinase on a chemically induced thrombosis model in rat. *Biol Pharm Bull* **18**(101): 387-1391 (1995).
15. Fujita, M., Hong, K., Ito, Y., Misawa, S.,Takeuchi, N., Kariya, K., Nishimuro, S, Transport of nattokinase across the rat intestinal tract. *Biol Pharm Bull* **18**(9): 1194-1196 (1995).
16. Okamoto, A., Hanagata, H., Kawamura, Y.,Yanagida, F. Anti-hypertensive substances in fermented soybean, natto. *Plant Foods Hum Nutr* **47**(1): 39-47 (1995).
17. Murayama, M., Sumi, H. *Basic and Clinical Aspects of Japanese Traditional Food Natto,*Miyazaki, Japan: Department of Physiology, Miyazaki Medical College, (1998).
18. Maruyama M, Sumi, H. Effect of natto diet on blood pressure,*Basic and Clinical Aspects*

of Japanese Traditional Food Natto. (vol. II) Tokyo: Japan Technology Transfer Association, 1–3 (1995).

19. Loehry, C.A., Permeability of the small intestine to substances of different molecular weight. *Gut,* 466-470 (1970).
20. Gardener, M.L.G. Gastrointestinal absorption of intact proteins. *Ann Rev Nutr* **8**: 329-350 (1988).
21. Murray, M. Thymus extracts, *J NaturopathicMed,* **5**(1): 77-79 (1994).
22. Sasaki, K., Moryama, S., Tanaka, Y., Sumi, H., Toki, N., Robbins, K.C. The transport of 125I-labeled human high molecular weight urokinase across the intestinal tract in a dog model with stimulation of synthesis and/or release of plasminogen activators. *Blood* **66**(12): 69-75 (1985).
23. Toki, N., Sumi, H., Sasaki, K., Boreisha, I.,Robbins, K.C, Transport of urokinase across the intestinal tract of normal human subjects with stimulation of synthesis and/or release of urokinase-type proteins. *J C l i n l n v e s t,* **75(**4): 1212-1222 (1985).
24. Sjum, H., Sasaki, K., Toki, N., Robbins, K.C. Oral administration of urokinase. *Thromb Res Suppl,* **20**(5-6): 711-714 (1980).
25. D J Wu *et al*., Lipid-lowering effect of nattokinase in patients with primary hypercholesterolemia, *Acta Cardiol. Sinica* **25**: 26-30 (2009).
26. Sumi H., Hamada, H., Nakanishi K., & Hiratani H. ,Enhancement of the fibrinolytic activity in plasma by oral administration of nattolinase. *Ata, Haematol*. **84**: 139-143 (1990).
27. Urranos, T et al. The profibrinolytic enzyme subtilisin NAT purified from *Bacillus subtilus* cleaves and inactivates plasminogen activator inhibitor. *J. Biol. Chem.* **276**: 24690-24696 (2001).
28. Ruei-Lin Hsu, Kung-Ta Lee, Jung-Hao Wang, Lily Y.L. Lee and Rita P.Y. Chen, Amyloid-Degrading Ability of Nattokinase from *Bacillus subtilis Natto*, *J. Agric. Food Chem.,* **57**(2): 503-508 (2009).
29. Ji Young Kim, Si Nae Gum, Jean Kyung Paik, Effects of Nattokinase on Blood Pressure: A Randomized, Controlled Trial. *Hypertension Research,* **31**: 1583-1588 (2008).
30. Cesarone, M.R.,Prevention of venous thrombosis in long-haul flights with FliteTabs: the Lonflit-Flite randomized, controlled trial. Angiology **54**,531-539 (2003).
31. Hayashi U.,Relationship between food containing “Natto” and the blood pressure of SHR, *Japanese Heart Journal*, **17**: 343-344 (1976).
32. Calvino, N.,The enzyme of enzymes—Nattokinase, *Townsend Letter for Doctors and Patients*, November, (2002).
33. Ding, W.J., Hypotensive and physiological effect of angiotens in converting enzyme inhibitory peptides from soy protein on spontaneously hypertensive rats. *Journal of Agriculture and Food Chemistry* **49**: 501-506 (2001).
34. Otsuguro K, Maeda T, Suzuki Y, Kondo K, Matsumoto Y, Zhao B-Q, Tsukamoto Y, Urano T, Umemura K,Dietary supplementation of fermented soybean, natto, suppresses intimal thickening and modulates the lysis of mural thrombi after endothelial injury in rat femoral artery. *Life Sci* **73**: 1289-1298 (2003).
35. Berg, D. Berg, L. Couvaras, J. Harrison, H. Chronic Fatigue Syndrome and/or Fibromyalgia as a Variation of Antiphopholipid Antibody Syndrome: an Explanatory Model and Approach to Laboratory Diagnosis. *Blood Coag Fibrinol,* **10**: 435-438 (1999).
36. MacLenna A,Antiphospholipid syndrome: Clinical and immunologic manifestations and patterns of disease expressionin a cohort of 1,000 patients, *Arthritis & Rheumatism* **46**(4): 1019-1027 (2002).
37. Harrison H, Berg L, Berg D, Polygenic Contributions of Procoagulant Genetic Factors in Chronic Fatigue Syndrome and Related Chronic Illnesses, *Genetics in Medicine*,**11**,2 (2002).
38. Fox P.F., Law J., Mc Sweeney P.L.H., Wallace J., Biochemistry of cheese ripening,*Cheese: Chemistry, Physics and Microbiology*,**1**(2): 389-438 (1993).
39. Fox P.F., O’Connor T.P., Mc Sweeney P.L.H., Guinee T.P., O’Brien N.M., Cheese: Physical, chemical, biochemical and nutritional aspects, Adv. *Food Nutr. Res.*,163-328 (1996).
40. Fox P.F., Wallace J.M., Morgan S., Lynch

C.M., Niland E.J., Tobin J., Acceleration of cheese ripening, *Anton. Leeuwenhoek*, 70271-70297 (1996).

41. Fox P.F., Guinee T.P., Cogan T.M., Mc Sweeney P.L.H., Fundamentals of Cheese Science, Aspen Publishers, Gaithersburg, (2000).
42. Fujita M., Nomura K., Hong K., Ito Y., Asada A., Nishimuro S., Purification and characterization of a strong fibrinolytic enzyme (nattokinase) in the vegetable cheese natto, a popular soybean fermented food in Japan, *Biochem. Biophys. Res. Comm.,* **197**: 34-1347 (1993).
43. Sumi Hiroyuki, Sasaki Tomohiro, Yatagai Chieko, Kozaki Yasutaka,Determination and Properties of the Fibrinolysis Accelerating Substance (FAS) in Japanese Fermented Soybean "Natto",*Nippon Nogeikagaku Kaishi*, **74(**11)**:** 1259-1264 (2000).
44. Ku TW, Tsai RL, Pan TM,A simple and cost-saving approach to optimize the production of subtilisin NAT by submerged cultivation of *Bacillus subtilis natto*,*J Agric Food Chem*; **57**(1): 292-296 (2009).
45. Maruyama M, Sumi H ,Effect of Natto Diet on Blood Pressure. *JTTAS,* (1995).
46. Pais E, Alexy T, Holsworth RE, Meiselman HJ, Effects of nattokinase a pro-fibrinolytic enzyme on red blood cell aggregation and whole blood viscosity, *Clin Hemorheol Microcirc*, **35**(1-2): 139-142 (2006).
47. Mitsugu Fujita, Kyongsu Hong, Yae Ito, Satoru Misawa, Naoto Takeuchi, Kimio Kariya ,Satoshi Nishimuro, Transport of Nattokinase across the Rat Intestinal Tract, *Biological & Pharmaceutical Bulletin*,**18**(9): 1194-1196 (1995).
48. Mitsugu Fujita1,Kyongsu Hong, Yae Ito, Rie Fujii, Kimio Kariya ,Satoshi Nishimuro, Thrombolytic Effect of Nattokinase on a Chemically Induced Thrombosis Model in Rat, *Biological & Pharmaceutical Bulletin*, **18**(10): 1387-1391(1995).
49. M R Cesarone, Prevention of Venous Thrombosis in Long-Haul Flights with Flite Tabs: The lonflit-flite randomized, *Controlled trial angiology*, **54**(5): 531-539 (2003).
50. Georgios Tsivgoulis, M D,Konstantinos Vemmos,M D,Common Carotid Artery Intima-Media Thickness and the Risk of Stroke Recurrence, *Stroke*. **37**: 1913 (2006).