

Skinny**SCIENCE**®

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L-ARGININE M2®

FORMULATING SAFE L-ARGININE

Via

PROPER ISOFORM PATHWAYS

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PROPER ISOFORM PATHWAYS

Arginine and Nitric Oxide are Double Edge Swords; capable of providing powerful benefits to human health, and also of causing serious damage to the body. Since clinical studies have proven that oral L-arginine can possess severe side effects, including the destruction of sperm and impairment of fertility in males, and the generation of dangerous brain-free-radicals, it has become mandatory to inform the medical community and the public about the hidden dangers of L-arginine.

The clinically established cautions related to dietary supplementation with L-arginine include:

- Dangerous Isoform Pathways
- Reactivation of the herpes simplex virus and other viruses
- High glycemcic/insulin stimulation
- Free radical damage
- Mortality

L-arginine and Nitric Oxide (NO) can be beneficial or damaging to the human body, depending on the metabolic pathway utilized. Given the Isoform pathway taken by L-arginine and Nitric Oxide, they can act as a *Double-Edge Sword*, as they are extremely *unstable* molecules that converts to nitrate and nitrite within ten seconds.

L-arginine is a precursor for nitric oxide (NO) synthesis. NO is a mediator that is formed by a family of enzymes named NO synthases. In the brain, NO acts as a neurotransmitter; in the immune system, NO acts as a mediator of host defense; and in the cardiovascular system, NO mediates the protective effects of the intact endothelium, acting as a vasodilator and endogenous anti-atherogenic molecule.

Nitric oxide is a potentially dangerous free radical; a highly reactive molecule that can be harmful to living tissues and can cause brain-related free radical damage, but the key is to do so without sparking peroxynitrite, a biologically essential oxidant that is the most toxic free radical to the body.

The Isoform pathway accessed by L-Arginine and/or nitric oxide (NO) determine the metabolic outcome and potential dangers to human health. Thus, the Isoform pathway is essential in the formulation of *safe* NO and L-arginine products.

JOURNAL OF THE AMERICAN MEDICAL ASSOCIATION (JAMA)

Concerns regarding the long-term and short-term safety of L-arginine were validated in the 2006 Journal of the American Medical Association (JAMA), in which the dangers of L-arginine were clearly demonstrated.

The 2006 *Journal of the American Medical Association* (JAMA) entitled *L-Arginine Therapy in Acute Myocardial Infarction*, alerted physicians, scientists, and the public to the extreme potential toxicity of the amino acid L-arginine (in its free-form).

In response to the JAMA article, on January 4, 2006 the *Wall Street Journal* reported “Heart Patients Urged to Avoid L-Arginine.” The article stated that the dietary supplement L-arginine “May harm heart patients” and that “Heart attack patients should avoid the dietary supplement L-arginine, based on a study that was scuttled after six volunteers taking the over-the-counter supplement died.”

The L-arginine double-blind, placebo-controlled clinical trial was headed by Dr. Steven Schulman, M.D. of Johns Hopkins Medical Institutions, and involved administering various doses of the amino acid L-arginine orally to subjects who had experienced cardiac events (myocardial infarction).

The clinical trial was abruptly stopped when death occurred (within six months) in 6 patients (8.6%) in the L-arginine group and none (-0-) in the placebo group ($P = .01$). After six months of a planned two-year study, the researchers became alarmed at the mortality rate in the group taking L-arginine, as compared to those taking a placebo. None of the subjects in the placebo (non-arginine) group died.

Researchers conducting the clinical trial stated that instead of the expected benefits from L-arginine, increased risk of death was seen, leading to early termination of the study. As reported in JAMA, “Because of the safety concerns, the data and safety monitoring committee closed enrollment.”

L-ARGININE DISCREPANCIES

The JAMA article evidenced short-term use of L-arginine as related to mortalities, yet there are thousands of published clinical trials utilizing L-arginine that have not resulted in mortalities. The discrepancy can be explained by:

- 1) Length of use
- 2) Health status of the user
- 3) Forms of L-arginine
- 4) L-arginine Antagonists

- 5) Metabolic Mistakes
- 6) Isoform Pathway

In order to formulate safe L-arginine, one must possess intricate knowledge of L-arginine biochemistry as well as cardiovascular medicine, and clinical trial variables. If the correct Isoform pathway is not utilized in L-arginine formulas, mortalities and long-term health damage can result.

LENGTH OF USE

Many of the clinical trials conducted on L-arginine are “short-term” and therefore do not always manifest toxic side effects. Long-term side effects are evidenced in humans ingesting L-arginine over an extended period of time.

HEALTH STATUS OF USER

In the Johns Hopkins clinical trial (JAMA), within six months of first ingesting L-arginine, mortalities occurred. The human subjects in the trial had impaired cardiovascular function, and therefore were already compromised. In persons with compromised immune function, impaired overall health, disease states, and/or impaired cardiovascular function, certain Isoform Pathways and forms of L-arginine can trigger severe health events.

FORMS OF L-ARGININE

The L-arginine-nitric-oxide signaling pathway is probably the most complicated facet of biochemistry. In creating an efficacious formula, the form of L-arginine used will dictate the quality of the product. If the form of arginine used in a formula is not appropriate, the product will not work, and can exhibit side effects.

The L-form (left-handed) of arginine is acceptable for human use, while the D-form (right-handed) is not. The left-handed form of L-arginine is available in low-to-high grades and is priced as a raw material according to the grade and form.

There are many unacceptable forms of L-arginine. Some of the more commonly seen unacceptable forms of L-arginine are as follows:

ARGININE HCL

In studies where the HCL form of L-arginine has been used, metabolic acidosis and alterations in electrolytes have been documented. Not recommended for human use. Arginine HCL is the form that was used in the tragic Johns Hopkins/JAMA clinical trial.

ARGININE PYROGLUTAMATE

Whose action in the body is *entirely speculative* according to the Physician’s Desk Reference (PDR). Not recommended for human use.

ARGININE KETOGLUTARATE and ALPHA-KETOGLUTARATE (AKG)

The Physician’s Desk Reference (PDR) lists all known forms of L-arginine. The PDR has no report of any form of L-arginine called “Arginine alpha-ketoglutarate.” There are no

clinical studies showing the long-term safety or efficacy of Arginine alpha-ketoglutarate, which is inherently unstable. Clinical studies showing the stability of Arginine alpha-ketoglutarate are required before this form of arginine is used in humans.

L-glutamine cannot be metabolized via L-ornithine to L-citrulline and L-arginine, confirming the lack of an effect of extracellular L-glutamine on L-arginine levels. Further, L-glutamine reduces L-arginine synthesis in humans, and research has shown that Alpha-ketoglutarate can cause hypoglycemia. Researchers and physicians caution that Alpha-ketoglutarate is not recommended for human use.

L-ARGININE ANTAGONISTS

Antagonists are agents that directly compete with L-arginine to block its health benefits, prevent its metabolism, and negate its ability to cross the blood-brain-barrier (BBB).

It is well established in the scientific literature that L-arginine antagonists include Lysine, Citrulline, Ornithine, sucrose (sugar), maltodextrins, glucose polymers, dextrose, proteins, high glycemic ingredients, and amino acids.

Ingredients that directly block L-arginine, and should be excluded from L-arginine formulas are maltodextrins, glucose polymers, sugar (sucrose), high fructose corn syrup, high-fructose, fructose corn syrup, protein, competing amino acids, Lysine, Ornithine, flavors made with maltodextrins, sweeteners without caloric impact, and any high glycemic raw material.

Any of these ingredients block arginine transport and metabolism and are contraindicated in L-arginine formulas. Certain nutrients and supplements also block L-arginine. Aspirin is a known antagonist to arginine.

LYSINE ANTAGOLISM

One of the most prominent formulating mistakes related to L-arginine antagonism is the inclusion of Lysine (one of the *Blind Amino Acids*). L-lysine is a direct antagonist of L-arginine, though it is frequently seen in L-arginine formulas. L-lysine is added to L-arginine for the purpose of mitigating the herpes reaction of body sores (*Herpes Simplex Virus*), as L-arginine can reactivate the herpes simplex virus (HSV) if it is improperly formulated.

Formulas containing L-arginine with L-lysine *do not work*, and are the result of improper formulating. Dietary disproportions of amino acids can alter the flux of specific amino acids across the blood-brain-barrier (BBB), and the Lysine/Arginine antagonism is an example of this alteration.

Unlike L-arginine, L-Lysine does not improve whole muscle strength or size, and is therefore a *substandard amino acid* for athletes and persons desiring to increase size and power output from muscle tissue and muscle mitochondria.

A plethora of clinical studies have proven that L-arginine and L-Lysine are antagonistic.

A small sampling of these studies are listed herein:

- Cross-inhibition studies show that L-arginine transport is blocked by 10-fold in the presence of L-lysine or L-ornithine. *Physiological Reviews, Vol. 83, No. 1, January 2003, pp. 183-252; American Physiological Society. Regulation of Amino Acid and Glucose Transporters in Endothelial and Smooth Muscle Cells*
- The effects of inhibitors of nitric oxide (NO) synthase and other cationic amino acids on unidirectional L-arginine transport were studied, with the conclusion that “L-lysine and L-ornithine inhibit transport of L-arginine.” *British Journal of Pharmacology*

The arginine/lysine study conducted by *Johns Hopkins University* also confirmed the fact that arginine and lysine are antagonistic. The Hopkins study showed that arginine cannot effectively release growth hormone when in the presence of lysine.

- Dr. Robert Ronzio, Ph.D. in biochemistry from the *University of California at Berkeley*, post-doctoral fellow at *Tufts University Medical College*, states that “Lysine supplements antagonize arginine.”
- Competitive inhibition of arginine uptake by other naturally occurring amino acids, such as L-lysine and L-ornithine, reduces NO synthesis. *Inoue et al., 1993*

GH RESPONSE TO L-ARGININE & L-LYSINE

Reports of a growth hormone (GH) response using the combination of arginine and lysine are incorrect. Statistical analysis of the study showing use of lysine with arginine shows that significant amounts of arginine and lysine could not have been absorbed from the gut in the short period of time before the observed GH peak. The scientists were measuring a needle-stick injury-induced release of GH in teen-age experimental subjects. Since lysine is more of an insulin releaser than a GH releaser, and a large insulin release can counteract the effect of GH, this is further evidence that lysine should be considered an unacceptable co-factor in any arginine formulation.

LYSINE & HERPES SIMPLEX VIRUS

All forms of L-arginine can re-activate the Herpes Simplex Virus (HSV). Since formulators are concerned with selling products that actually *cause* mouth sores to appear, they routinely add Lysine to their formulas. As discussed, Lysine is a direct antagonist of L-arginine and cannot be used in its presence.

L-arginine does not *cause* herpes, but it does exacerbate latent herpes simplex viruses, and other viruses, causing symptoms to re-appear. The only known solution to this problem is the incorporation of specific technologies in formulating L-arginine compounds that mitigate or eliminate the aggravating biochemical factors by formulating L-arginine products appropriately.

L-lysine is routinely used by persons with herpes simplex virus to minimize episodes. In said cases, L-lysine must be taken 2-4 hours apart from orally ingested L-arginine, as their relationship is antagonistic.

In a certain substrate of the population, even minuscule amounts of supplemental arginine can cause symptoms to return. The herpes simplex virus reacts strongly to stress, and any mental or physical stress can reactivate symptoms.

In said cases, L-arginine products are contraindicated.

ORNITHINE ANTAGONISM

L-ornithine has been clinically proven to be an antagonist of L-arginine and disrupts L-arginine transport. Additionally, L-ornithine is contraindicated in L-arginine formulas and in products geared to the athlete.

L-ornithine should be avoided by bodybuilders, powerlifters, and all other athletes, as well as persons desiring to increase muscle mass, because ornithine is a non-protein amino acid. Ornithine is *not* anabolic and is not used to make protein in the human body, whereas arginine does make protein and is anabolic. Supplemental L-ornithine does not have a set metabolic pathway in the body, and as such is suspect as an orally ingested amino acid. L-arginine turns into ornithine in the body, so it would be metabolically repetitious to use L-ornithine in an L-arginine formula.

METABOLIC MISTAKES

The complex mechanism that allows L-arginine to be accepted by the body does *not* allow for *metabolic mistakes* in formulating. Many of the current arginine products on the market today are perfect examples of very credible scientists attempting to produce an L-arginine product without benefit of expertise in arginine biochemistry. The ingredients in the majority of arginine products actually *block* L-arginine's entry into the body as a free form amino acid. If L-arginine is not properly formulated, it *will not* elevate serum levels of L-arginine, and *will not* provide health benefits. L-arginine products that are founded on *metabolic mistakes* do *not* elevate serum levels of L-arginine, and are therefore ineffective.

CITRULLINE

A prime example is the inclusion of L-citrulline in an L-arginine formulation. According to independent studies, L-citrulline is not an appropriate ingredient in an L-arginine formulation:

“Conversion of citrulline to arginine occurs primarily within the kidney. Increased mortality (death) risk ratio observed after citrulline is included in the Cox regression analysis reflects the effects of renal dysfunction on arginine bioavailability.”

“Citrulline levels trended higher in Sickle Cell Disease (SCD) patients with pulmonary hypertension and correlated with rising creatinine levels (Spearman $\rho = 0.51$; $P < .001$).”

Dysregulated Arginine Metabolism, Hemolysis-Associated Pulmonary Hypertension, and Mortality in Sickle Cell Disease. JAMA.2005;294:81-90

The use of Citrulline in an L-arginine formulation causes inherent problems, including depletion of L-arginine. According to *The Arginine Paradox (Folia Pharmacol. Japan Vol. 119 7-14:2002 Department of Pharmacology, Teikyo University School of Medicine)*:

- L-Arginine has attracted major interest because it has been identified as the natural substrate of nitric oxide synthase and is now recognized as a major player in the regulation of biological function.
- The arginine paradox refers to the phenomenon that exogenous L-arginine causes NO-mediated biological effects despite the fact that nitric oxide synthases (NOS) are theoretically saturated with the substrate L-arginine.
- There have been several explanations for this phenomenon, although none of them can explain the arginine paradox fully:
 - (1) L-arginine-induced insulin, which has vasodilatory actions.
 - (2) Neither extracellular nor intracellular concentration determines the NOS activity but rather the L-arginine amount transported across the plasma membrane may do so.
 - (3) Endogenous NOS inhibitors reduce the enzyme sensitivity to L-arginine. These inhibitors include, NG, NG-dimethyl-L-arginine, L-citrulline, argininosuccinic acid and agmatine.
 - (4) Intracellular L-citrulline, an NOS product, is a potent inhibitor of NOS so that the *cells may need extra L-arginine to compete with L-citrulline inhibition.*

Further evidence against the use of L-citrulline is the the 2005 *Alternative Medicine Review* reported that L-arginine, and *not* L-citrulline is the appropriate amino acid for oral supplementation:

- “The amino acid L-arginine is the preferred substance for oral supplementation to enhance nitric oxide synthesis.”
- “The mechanism by which L-arginine works is by providing the substrate for nitric oxide synthesis in vascular endothelial cells, which in turn creates cyclic GMP in the underlying vascular smooth muscle cells.”
- “L-arginine overcomes ADMA’s nitric oxide inhibition.”

- “Some supplement companies are marketing L-citrulline – a by-product of the arginine-to-nitric oxide pathway – as a substance to increase nitric oxide synthesis in vascular endothelial cells.”
- “Citrulline does not directly convert to nitric oxide, but instead is recycled to L-arginine (an ATP-dependent process), which then converts to nitric oxide.”

The Nobel Prize in Physiology/Medicine 1998 was awarded to doctors Robert F. Furchgott, Louis J. Ignarro, and Ferid Murad for their discoveries concerning "the nitric oxide as a signalling molecule in the cardiovascular system". As reported in the 2005 *Alternative Medicine Review*, “Ferid Murad, MD, PhD, Nobel-prize winner for his research on nitric oxide, has said the use of L-citrulline to increase nitric oxide is only *marginally effective*.” The *Alternative Medicine Review* conclusion is . . . toss out the citrulline, and utilize L-arginine. “Change bad medicine to good medicine and ignore the hype.”

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ISOFORM PATHWAYS

The most important aspect in selecting a safe L-arginine product is the addition of a “*Blind Amino Acid*” Rider. Without this essential facet, L-arginine cannot take a safe Isoform Pathway. A *Blind Amino Acid* is any amino acid that has been separated from its sister-amino-acids (SAA). All “L-form” amino acids are *Blind Amino Acids*.

L-arginine is a *Blind Amino Acid*, but arginine found in food, such as cheese, is not a *Blind Amino Acid*. Regular arginine, as found in protein foods, is always combined with all the other known amino acids, and is therefore not considered a *Blind Amino Acid*.

Blind Amino Acids, once separated from their sister-amino-acids, do not know where to go in the human body, and consequently wreak havoc. If they are directed to a specific Isoform Pathway via a “Rider” the formerly *Blind Amino Acid* is given metabolic direction, and becomes safe for long-term use. L-arginine can be engineered to enact specific cell signaling. Methodologies for developing target-specific amino acid formulas provides the ability to pre-select Isoform Pathways that are safe for use in humans.

Appropriately formulated L-arginine formulas will mediate the uptake of L-arginine into cells and tissues *in vivo*. This methodology allows non-synthetic molecules to reach their target site without causing severe side effects.

SAFE USE IN HUMANS

Part of the criteria for selecting an appropriate L-arginine product or formula is the issue of safe use in humans. This information is known as the *Historical Data* and/or the

History-of Use and refers to the length of time a product has been broadly used by humans.

The term “broadly used” is defined as *product-usage in large numbers of the human population*. Corporate records for any product sold to the public will show:

- How many people have used the product
- How long the product has been on the market
- FDA Regulatory sanctions; warning letters, reprimands, and fines levied against a product
- Federal Trade Commission (FTC) violations for false product claims

To determine the long-term safety of a product, its use in humans over a minimum of 5 years is analyzed. If an L-arginine product has *not* been used broadly by humans over a 5-year period, long-term safety has not been established.

- **5-YEARS of HISTORICAL DATA**
Use in humans for 5 years with no evidence of sanctioned side effects is considered the *minimum* criteria for determining safety.
- **10-YEARS of HISTORICAL DATA**
If an L-arginine product has been used broadly by the public for 10 + years with no sanctioned side effects, its safety and efficacy has been well established.
- **20-YEARS of HISTORICAL DATA**
If the product has been used broadly in humans for 20 + years, with no government sanctions or severe side effects, it is considered completely safe and efficacious by legal, epidemiologic, and evidential definition.

CONSUMER EDUCATION

Consumers have an absolute right to question the manufacturer of any L-arginine product and to be given the scientific justification and expertise behind the product. The History-of-Use related to a product should be revealed upon consumer request. L-arginine Nutraceutical products should always be accompanied by a web site that provides information on the product, including History-of-Use. If this information is not available to the public, the product’s safety and efficacy must be questioned. If an L-arginine product claims *drug status*, the pharmaceutical company that produces the product should provide documentation concerning the formula.

SUMMARY

Since the 2006 JAMA article was published, consumers have become extremely cautious about using L-arginine. It is unwise to use *any* L-arginine product without first ascertaining its Length of Use (history and use in humans), the formulator of the product, the Patent-status, the form of L-arginine used, the exclusion of L-arginine antagonists, and its ultimate Isoform Pathway via inclusion of a “*Blind Amino Acid Rider*.”

In terms of future L-arginine clinical trials, it is mandatory that the form of L-arginine be bound to a *Blind Amino Acid Rider*, and formulated by an L-arginine expert with a background in safe arginine Isoform Pathways.

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ADDITIONAL REFERENCES

P. Roy. Recent trends in the nitrenergic nervous system.2005. Indian Journal of Pharmacology; P. Roy, G. Venkat Ramana, M. Naidu, P. Usha Rani

Thomas G, Ramwell PW. Nitric oxide, donors and inhibitors. In: Bertram G Katzung, editor. Basic and Clinical Pharmacology. United States: McGraw Hill; 2004.p.313-8.

Chandran S, Sridhar N, Veeranjanyulu A. Nitric oxide: concepts, current perspectives and future therapeutic implications. Indian J Pharmacol 1998;30:351-66.

Lee J, Ryu H, Ferrante RJ, Morris SM Jr, Ratan RR. Translational control of inducible nitric oxide synthase expression by arginine can explain the arginine paradox. Proc Natl Acad Sci U S A 2003;100:4843-8.

Nakaki T, Hishikawa K.The arginine paradox [Article in Japanese]. Nippon Yakurigaku Zasshi 2002;119:7-14.

Lee J, Ryu H, Ferrante RJ, Morris SM, Ratan RR. Translational control of inducible nitric oxide synthase expression by arginine can explain the arginine paradox. Proc Natl Acad Sci U S A. 2003;100:4843-4848.

Böger RH, Ron ES. L-Arginine improves vascular function by overcoming the deleterious effects of ADMA, a novel cardiovascular risk factor. Altern Med Rev. 2005;10:14-23.

Cannon RO III. Oral L-arginine (and other active ingredients) for ischemic heart disease? J Am Coll Cardiol. 2002;39:46-48.

Mason RP, Walter MF, Jacob RF. Effects of HMG-CoA reductase inhibitors on endothelial function: role of microdomain and oxidative stress. Circulation. 2004;109(21 suppl 1):II-34-II-41.

Committee on the Use of Complementary and Alternative Medicine by the American Public. Complementary and Alternative Medicine (CAM) in the United States. Institute of Medicine of the National Academies. Washington, DC: National Academies Press; 2005.

Lakatta EG, Levy D. Arterial and cardiac aging: major shareholders in cardiovascular disease enterprises, I: aging arteries: a "set up" for vascular disease. Circulation. 2003;107:139-146.

Lakatta EG, Levy D. Arterial and cardiac aging: major shareholders in cardiovascular disease enterprises, II: the aging heart in health: links to heart disease. Circulation. 2003;107:346-354.

Taddei S, Virdis A, Mattei P, et al. Aging and endothelial function in normotensive subjects and patients with essential hypertension. Circulation. 1995;91:1981-1987.

Celermajer DS, Sorensen KE, Spiegelhalter DJ, Georgakopoulos D, Robinson J, Deanfield JE. Aging is associated with endothelial dysfunction in healthy men years before the age-related decline in women. J Am Coll Cardiol. 1994;24:471-476.

Sutton-Tyrrell K, Najjar SS, Boudreau RM, et al. Elevated aortic pulse wave velocity, a marker of arterial stiffness, predicts cardiovascular events in well-functioning older adults. Circulation. 2005;111:3384-3390.

Drexler H. Endothelial dysfunction. Prog Cardiovasc Dis. 1997;39:287-324.

Wilkinson IB, Qasem A, McEniery CM, Webb DJ, Avolio AP, Cockcroft JR. Nitric oxide regulates local arterial distensibility in vivo. Circulation. 2002;105:213-217.

Chauhan A, More RS, Mullins PA, Taylor G, Petch MC, Schofield PM. Aging-associated endothelial dysfunction in humans is reversed by L-arginine. J Am Coll Cardiol. 1996;28:1796-1804.

Rector TS, Bank AJ, Mullen KA, et al. Randomized, double-blind, placebo-controlled study of supplemental oral L-arginine in patients with heart failure. Circulation. 1996;93:2135-2141.

Mark DB, Nelson CL, Califf RM, et al. Continuing evolution of therapy for coronary artery disease: initial results from the era of coronary angioplasty. *Circulation*. 1994;89:2015-2025.

Schulman SP, Weiss JL, Becker LC, et al. Effect of early enalapril therapy on left ventricular function and structure in acute myocardial infarction. *Am J Cardiol*. 1995;76:764-770.

Chen CH, Nevo E, Retics B, et al. Estimation of central aortic pressure waveform by mathematical transformation of radial tonometry pressure. *Circulation*. 1997;95:1827-1836.

Avolio AP, Deng FQ, Li WQ, et al. Effects of aging on arterial distensibility in populations with high and low prevalence of hypertension: comparison between urban and rural communities in China. *Circulation*. 1985;71:202-210.

19. Kelly RP, Ting CT, Yang TM, et al. Effective arterial elastance as index of arterial load in humans. *Circulation*. 1992;86:513-521.

Adams MR, McCredie R, Jessup W, Robinson J, Sullivan D, Celermajer DS. Oral L-arginine improves endothelium-dependent dilatation and reduces monocyte adhesion to endothelial cells in young men with coronary artery disease. *Atherosclerosis*. 1997;129:261-269.

Lachin JM. Worst-rank score analysis with informatively missing observations in clinical trials. *Control Clin Trials*. 1999;20:408-422.

Berger VW. Improving the information content of categorical clinical trial endpoints. *Control Clin Trials*. 2002;23:502-514.

Böger RH, Bode-Böger SM. The clinical pharmacology of L-arginine. *Annu Rev Pharmacol Toxicol*. 2001;41:79-99.

Maggioni AP, Maseri A, Fresco C, et al, the investigators of the Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico (GISSI-II). Age-related increase in mortality among patients with first myocardial infarctions treated with thrombolysis. *N Engl J Med*. 1993;329:1442-1448.

White HD, Barbash GI, Califf RM, et al. Age and outcome with contemporary thrombolytic therapy: results from the GUSTO-I trial. *Circulation*. 1996;94:1826-1833.

Aguirre FV, McMahon RP, Mueller H, et al. Impact of age on clinical outcome and postlytic management strategies in patients treated with intravenous thrombolytic therapy: results from the TIMI II study. *Circulation*. 1994;90:78-86.

Ishihara H, Yokota M, Sobue T, Saito H. Relation between ventriculoarterial coupling and myocardial energetics in patients with idiopathic dilated cardiomyopathy. *J Am Coll Cardiol.* 1994;23:406-416.

Asanoi H, Sasayama S, Kameyama T. Ventriculararterial coupling in normal and failing hearts. *Circ Res.* 1989;65:483-493.

Pfeffer MA, Braunwald E. Ventricular remodeling after myocardial infarction: experimental observations and clinical implications. *Circulation.* 1990;81:1161-1172.

Mitchell GF, Moya LA, Braunwald E, et al. Sphygmomanometrically determined pulse pressure is a powerful independent predictor of recurrent events after myocardial infarction in patients with impaired left ventricular function. *Circulation.* 1997;96:4254-4260.

St. John Sutton M, Pfeffer MA, Plappert T, et al. Quantitative two-dimensional echocardiographic measurements are major predictors of adverse cardiovascular events after myocardial infarction. *Circulation.* 1994;89:68-75.

Ludmer PL, Selwyn AP, Shook TL, et al. Paradoxical vasoconstriction induced by acetylcholine in atherosclerotic coronary arteries. *N Engl J Med.* 1986;315:1046-1051.

Vallance P, Collier J, Moncada S. Effects of endothelium-derived nitric oxide on peripheral arteriolar tone in man. *Lancet.* 1989;2:997-1000.

Bank AJ, Wilson RF, Kubo SH, Holte JE, Dresing TJ, Wang H. Direct effects of smooth muscle relaxation and contraction on in vivo human brachial artery elastic properties. *Circ Res.* 1995;77:1008-1016.

Cooke JP, Dzau VJ. Derangements of the nitric oxide synthase pathway, L-arginine, and cardiovascular diseases. *Circulation.* 1997;96:379-382.

Böger RH, Bode-Böger SM, Szuba A, et al. Asymmetric dimethylarginine (ADMA): a novel risk factor for endothelial dysfunction. *Circulation.* 1998;98:1842-1847.

Loscalzo J. What we know and don't know about L-arginine and NO. *Circulation.* 2000;101:2126-2129.

Moncada S, Higgs A. The L-arginine-nitric oxide pathway. *N Engl J Med.* 1993;329:2002-2012.

Lerman A, Burnett JC, Higano ST, McKinley LJ, Holmes DR. Long-term L-arginine supplementation improves small-vessel coronary endothelial function in humans. *Circulation.* 1998;97:2123-2128.

Clarkson P, Adams MR, Powe AJ, et al. Oral L-arginine improves endothelial-dependent dilation in hypercholesterolemic young adults. *J Clin Invest.* 1996;97:1989-1994.

Cheng JW, Baldwin SN. L-arginine in the management of cardiovascular diseases. *Ann Pharmacother.* 2001;35:755-764.

Morris SM Jr. Regulation of enzymes of urea and arginine synthesis. *Annu Rev Nutr.* 1992;12:81-101.

Castillo L, Sanchez M, Chapman TE, Ajami A, Burke JF, Young VR. The plasma flux and oxidation rate of ornithine adaptively decline with restricted arginine intake. *Proc Natl Acad Sci U S A.* 1994;91:6393-6397.

Berkowitz DE, White R, Li D, et al. Arginase reciprocally regulates nitric oxide synthase activity and contributes to endothelial dysfunction in aging blood vessels. *Circulation.* 2003;108:2000-2006.

Blum A, Hathaway L, Mincemoyer R, et al. Oral L-arginine in patients with coronary disease on medical management. *Circulation.* 2000;101:2160-2164.

Loscalzo J. Adverse effects of supplemental L-arginine in atherosclerosis. *Arterioscler Thromb Vasc Biol.* 2003;23:3-5.

Takimoto E, Champion HC, Li M, et al. Oxidant stress from nitric oxide synthase-3 uncoupling stimulates cardiac pathologic remodeling from chronic pressure overload. *J Clin Invest.* 2005;115:1221-1231.

Chen J, Kuhlencordt P, Urano F, Ichinose H, Astern J, Huang PL. Effects of chronic treatment with L-arginine on atherosclerosis in apoE knockout and apoE/inducible NO synthase double-knockout mice. *Arterioscler Thromb Vasc Biol.* 2003;23:97-103.

Hochman JS. Cardiogenic shock complicating acute myocardial infarction: expanding the paradigm. *Circulation.* 2003;107:2998-3002.

Mancini GB, Henry GC, Macaya C, et al. Angiotensin-converting enzyme inhibition with quinapril improves endothelial vasomotor dysfunction in patients with coronary artery disease: the TREND (Trial on Reversing Endothelial Dysfunction) Study. *Circulation.* 1996;94:258-265.