ORAL CITRULLINE AS ARGININE PRECURSOR MAY BE BENEFICIAL IN SICKLE CELL DISEASE: EARLY PHASE TWO RESULTS

William H. Waugh, MD, Charles W. Daeschner III, MD, Beatrice A. Files, MD, Michael E. McConnell, MD, and Sarah E. Strandjord, MD
Greenville, North Carolina

L-Arginine may be a conditionally essential amino acid in children and adolescents with sickle cell disease, particularly as required substrate in the arginine-nitric oxide pathway for endogenous nitrovasodilation and vasoprotection. Vasoprotection by arginine is mediated partly by nitric oxide-induced inhibition of endothelial damage and inhibition of adhesion and activation of leukocytes. Activated leukocytes may trigger many of the complications, including vasoocclusive events and intimal hyperplasias. High blood leukocyte counts during steady states in the absence of infection are significant laboratory risk factors for adverse complications. L-Citrulline as precursor amino acid was given orally twice daily in daily doses of approximately 0.1 g/kg in a pilot Phase II clinical trial during steady states in four homozygous sickle cell disease subjects and one sickle cell-hemoglobin C disease patient (ages 10–18). There soon resulted dramatic improvements in symptoms of well-being, raised plasma arginine levels, and reductions in high total leukocyte and high segmented neutrophil counts toward or to within normal limits. Continued L-citrulline supplementation in compliant subjects continued to lessen symptomatology, to maintain plasma arginine concentrations greater than control levels, and to maintain nearly normal total leukocyte and neutrophil counts. Side effects or toxicity from citrulline were not experienced. Oral L-citrulline may portend very useful for palliative therapy in sickle cell disease. Placebo-controlled, long-term trials are now indicated. (*J Natl Med Assoc.* 2001;93:363–371.)

Key words: sickle cell disease ◆ citrulline ◆ arginine ◆ leukocytosis ◆ adverse events

Higher than normal total leukocyte counts are commonly present during steady states in sickle cell disease.[1,2] Higher total leukocyte counts in the absence of infection during steady states correlate as a significant laboratory risk factor for adverse compli-

 From the Departments of Physiology and Medicine and the Department of Pediatrics, Division of Hematology/Oncology and Division of Cardiology, Brody School of Medicine, East Carolina University, Greenville, North Carolina.

Requests for reprints should be addressed to Dr. Waugh, Department of Physiology, Brody School of Medicine, Greenville, NC 27858. E-mail: Waughw@mail.ecu.edu

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cations, including painful vasoocclusive crises, strokes, acute chest syndromes, and greater mortality rates.[1,3-6]

The high total leukocyte counts during steady states in sickle cell anemia are primarily attributable to increased segmented neutrophils in the peripheral blood; the nonsegmented (band) neutrophils usually account for less than 4% of the total leukocyte count and usually less than 7% of the total neutrophil count.[2] Increased production and use of neutrophils are involved, in addition to a shift from the marginal pool.[7]

Endothelial cells are normally quiescent, but they can become much activated in sickle cell disease.[8,9] There is endothelial damage and inflammatory vasculopathy.[8,9] Neutrophils interact with sickle erythrocytes and endothelial cells, and the neutrophils are stimulated to release injurious substances. Neutrophils can produce injury to vascular endothelial cells by a variety of mechanisms involving oxygen products, such as superoxide, H_2O_2, and the highly reactive hydroxyl free radical, or involving proteolytic enzymes released from the neutrophils.[10] Neutrophils have been implicated in initiation or promotion of vasoocclusive events and tissue damage in sickle cell disease.[11] Superoxide anion, released from endothelial cells or neutrophils, can be involved in the breakdown of nitric oxide, the endothelium-derived vascular relaxing factor.[12] An adequate supply of L-arginine is required as a substrate in the arginine-nitric oxide pathway by endothelial nitric oxide synthase in its activity for homeostatic nitrovasodilation and vasoprotection.[13] Cells produce more superoxide anion generated by nitric oxide synthase activity when cells are depleted of L-arginine, as with ischemia or by lowered arginine supply levels, and less nitric oxide is formed.[14]

We provided evidence that a relative arginine deficiency commonly exists in sickle cell anemia.[15] Evidence is now supplied that L-citrulline, given orally for novel orthomolecular medical use as precursor agent for arginine biosynthesis,[16,17] results not only in symptomatic improvement in sickle cell disease but also in amelioration of the associated neutrophilia.

METHODS

Selection of Patients and Study Design

The first five African-American patients who complied with ingestion of the prescribed number of L-citrulline capsules twice daily for 4 weeks were studied. One of the first six subjects who had agreed to the trial was dropped from the study because of poor compliance without having side effects from citrulline. Four subjects with HbSS disease were studied: 3 males aged 14, 17, and 18 years and 1 female aged 12 years. One 10-year-old male with HbSC disease was also studied. All patients attended and were examined in the Regional Comprehensive Pediatric Sickle Cell Outpatient Clinic at East Carolina University. U.S. Food and Drug Administration (FDA) and local institutional research board approvals were obtained for this pilot Phase II protocol for L-citrulline capsules twice daily as a nutritional supplement in sickle cell disease. Subjects were enrolled after written informed consent. All subjects had been in a steady state (not painful crisis state) for at least 4 weeks and were not on hydroxyurea therapy. On the first (control) day of each study before citrulline was given, focused histories, physical examinations, two-dimentional echocardiographic examinations, and blood specimens were obtained. Brachial blood pressures were obtained in the sitting position. Mean arterial blood pressures were estimated using the formula: mean pressure equals diastolic pressure plus one-third of pulse pressure. Similar 4-week evaluations (post-citrulline) were performed at the end of the first 28-days on citrulline. The echocardiograms were performed in a resting state in a left lateral decubitus position, using an Acuson Sequoia C256 imaging system (Acuson, Siemens Co., Mountin View, CA). Total peripheral resistances at the start and end of the initial 4-week trials were estimated using the formula: TPR units equal mean arterial pressure, in mm Hg, divided by cardiac index, expressed in liters of estimated left ventricular minute output divided by square meters of estimated body surface area. Left ventricular output in the outflow tract was estimated by echocardiography. The citrulline was given orally in the morning and after the evening meal for 28 days. Subsequent to the initial 28-day trials in these five patients, in three of the four patients who agreed to continue taking citrulline, serial 4-to 6-week test periods were performed, after at least 12 weeks of discontinued citrulline therapy. Echocardiograms were not done during these subsequent trial periods or in the fourth patient who continuously remained on citrulline supplementation. Visual analogue scale lines were marked by each subject during these near-monthly repeat

clinic visits to semiquantitate the general symptomatic feelings of the patients' disease at that time.

Hematologic and Biochemical Measurements

Blood specimens were obtained by arm venipuncture. Heparinized blood samples for L-arginine assays were immediately put into crushed ice. Plasmas were separated by centrifugation at 4°C. Aliquots of plasma were mixed with equal volumes of 10 g/dl trichloroacetic acid solution within 45 minutes of venipuncture sampling. The 1:2 deproteinized spun supernates of plasma were transferred into other microcentrifuge tubes, and 0.8 molar K_2CO_3 was added for neutralization. Samples were stored frozen at −20°C before assay. Plasma L-arginine was assayed by a method with absorbance differences for monosubstituted guanidines without and with- 5 to 6-minute incubations at 37°C with bovine liver L-arginase for specificity.[15] All other blood determinations were done by standard hospital methods.

Statistical Analysis

Paired Student's *t*-test for mean differences between control data (before citrulline intake) and 4-week or near-monthly data were used. p Values equal to or below 0.05 were considered to indicate statistical significance.

RESULTS

All five subjects reported subjective feelings of increased well-being during the first 4 weeks of each initial or renewed 4-week or near-monthly trial period. The senses of well-being began during the first 2 weeks. Improved sense of wellness varied from feeling better in one sedate, studious subject to very much better in the other 4.

As an illustration, one patient is now described briefly. Patient AT, a 14-year-old male with sickle cell anemia, presented initially with his usual flat mood and appearance of lassitude before the start of the trial. Subsequently, his mood was bright and he reacted alertly. He said "I can breathe better" and my pep is "better." By "breathing better," he meant that he did not get short of breath as quickly as he had done in the past with normal physical activity. He also reported "I have more energy and stay awake better." "My grades are better because I'm awake and study more." He said that he can run better and play basketball better without tiring as much compared to before the intake of the capsules (of citrulline). No adverse effects or painful episodes were experienced during the periods of the trial while on citrulline.

Table 1 illustrates the individual general symptoms of illness or sense of well-being, and the individual and average values of cardiovascular parameters found before and during or 1 day after the initial 28-day period of citrulline supplementation. The L-citrulline was prescribed twice daily in daily dosages of 0.09 to 0.13 g/kg during this initial trial period. There were no significant mean changes in measured mean arterial blood pressure, heart rate, left ventricular mass index (LVMI), cardiac output, cardiac index, or total peripheral resistance in the five patients. The initial high cardiac outputs and cardiac indices in the sickle cell anemic subjects remained higher than normal in the 4 HbSS patients. However, in one of these patients, case patient AT, much cardiomegaly was initially evident, with a striking reduction in measured left ventricular end-diastolic dimension from 5.2 to 4.8 cm (not tabulated) and a dramatic reduction in LVMI from 116 to 81 g/m^2 over the 28-day interval. Sizable reductions in his cardiac index of 19% and in his mean arterial blood pressure of 26% were associated, with only modest reduction in total peripheral resistance (see Table 1). This was in one of the four subjects who experienced much less exertional dyspnea and exertional fatigue while on the citrulline supplementation. All four patients with HbSS disease had measured decreases in LVMI in this initial 4-week trial, but the mean paired decrease was not of statistical significance. Cardiac output and cardiac index were within normal limits in the younger patient with HbSC disease (patient SH). No symptoms of light-headedness or episodes of syncope were reported by any of the patients while they were receiving citrulline.

There were significantly mean decreases in total leukocyte counts and segmented neutrophil counts in the five patients during their initial 4-week trials on citrulline. Results are tabulated in Table 2, in which the mean decrease was from 12.46 ± 1.81 to 9.99 ± 1.70 k/μL in total leukocyte count and 6.39 ± 1.44 to 4.18 ± 1.23 k/μL in segmented neutrophil count, with p-values of <0.001 and <0.005, respectively. Table 2 also shows that the citrulline supplementation was not associated with statistical significant mean changes in the degree of

Table 1. Symptoms of Illness and Cardiovascular Measurements Before and During or 1 Day After the Initial 4-Week Trials of L-Citrulline

Variable	Time	Symptoms (U, B, VMB)	MAP (mm Hg)	HR (beats/min)	LVMI† (g/m^2)	CO (L/min)	CI (CO/m^2)	TPR (units)
1AT;	Before	U	97	85	116	6.88	4.41	22.0
HbSS	After	VMB	72	62	81	5.55	3.56	20.2
2JF;	Before	U	91	70	73	5.67	3.34	27.2
HbSS	After	B	74	79	69	6.13	3.57	20.7
3AF;	Before	U	77	83	86	7.57	3.77	20.4
HbSS	After	VMB	96	82	81	10.50	5.36	17.9
4TD;	Before	U	63	96	78	7.98	4.63	13.6
HbSS	After	VMB	90	64	67	6.10	3.47	25.9
5SH;	Before	U	73	73	45	2.43	2.36	30.9
HbSC	After	VMB	74	67	58	2.21	2.16	34.2
Mean ± SEM	Before		80 ± 6.1	81 ± 5	80 ± 11	6.1 ± 1.0	3.7 ± 0.4	22.8 ± 3.0
Mean ± SEM	After		81 ± 4.9	71 ± 4	71 ± 4.4*	6.1 ± 1.3	3.6 ± 0.5	23.8 ± 2.9

*U, B, and VMB represent the general feelings of illness by the patient as usual, better, or very much better. MAP, mean arterial pressure; HR, heart rate; LVMI, left ventricular mass index estimate per body surface area; CO, cardiac output; CI, cardiac index; TPR, total peripheral resistance. SEM represents standard error of the mean.
†*p*-value < 0.10 for mean paired decrease of 13.9 ± 7.2 g/m^2 in LVMI (1-sided *t*-test) in the 4 HbSS subjects.

hemolytic anemias, because mean hemoglobin, reticulocyte, and serum bilirubin values were similar before and at the end of the trial periods. Values for mean red cell volume, mean cell hemoglobin, and mean cell hemoglobin concentration were not apparently influenced by citrulline administration also, as shown in Table 2. However, with citrulline, plasma arginine concentration rose sizably in four of the subjects, but not in the fifth patient with the greatest magnitude of reticulocytosis and hyperbilirubinemia (patient AF). Mean plasma arginine level rose from 77 ± 9.1 to 127 ± 18 μmol/L, with a *p*-value of <0.05. This represented a 65% mean increase in plasma arginine concentration from the initial mean value.

With continuous L-citrulline supplementation in two of the HbSS patients over 5 near-monthly intervals and in one of the patients with HbSS disease over 9 near-monthly periods, Table 3 documents that mean total leukocyte counts and segmented neutrophil counts continued lower than found in these three individuals before their initial control time states. In the patient with HbSC disease, these white cell parameters remained lower than in the three HbSS subjects. Mean visual analog scale line values for well-being or sense of illness at the end of the near-monthly periods remained very much lower than the control before values for each of the four patients (see Table 3). Mean plasma arginine levels were elevated sizably and up to 2.9 times higher than the control before levels in three of the patients. Mean arginine level was found to be barely raised in patient AF with the greatest magnitude of hemolytic anemia, as was similarly found in the initial near-monthly (28-day) period shown in Table 2 for the same patient, AF.

Only one patient experienced a severe painful vasoocclusive crisis during the above near-monthly periods. This patient, TD, was hospitalized for acute, multiple somatic pains and acute chest syndrome during 1 of her 5 near-monthly continuous trial periods. Upon hospitalization for this acute crisis, her total leukocyte count was very elevated to 24.9 k/μL, her segmented neutrophil count was very high at 20.42 k/μL, and her plasma arginine level was found much lower than before, at 48 μmol/L. Citrulline supplementation was given under supervision during her hospitalization at her usually prescribed, prehospital daily doses of 3 g in the morning and 4 g at night without untoward results. Near the end of her hospital stay (for 9 days) with conventional concurrent therapy, her total leukocyte count had declined to 12,500 k/μL and her plasma arginine concentration was much higher at 201 μmol/L.

Table 2. Leukocyte Counts, Hemoglobin Values, Red Cell Parameters, Serum Bilirubin, and Plasma Arginine Levels Before and After the Initial 4-Week Trials of L-Citrulline

Variable	Time	WBC* (k/μL)	NEUTR (k/μL)	HB (g/dL)	RETIC (k/μL)	MCV (f/L)	MCH (pg)	MCHC (g/dL)	BILIR (mg/dL)	ARG (μMol/L)
1AT;	Before	9.66	4.63	9.2	296	68	22.5	33.2	0.8	56
HbSS	After	7.77	2.95	9.4	252	69	23.0	33.5	0.7	127
2JF;	Before	12.20	4.51	9.0	289	80	28.7	35.8	3.3	109
HbSS	After	9.91	2.28	9.0	285	81	28.0	34.6	2.8	120
3AF;	Before	18.00	12.06	7.8	493	93	35.4	38.1	11.1	71
HbSS	After	15.60	9.05	7.5	629	96	34.7	36.2	15.7	79
4TD;	Before	14.70	5.88	7.8	332	85	28.9	33.9	4.7	83
HbSS	After	11.20	3.14	8.0	469	89	31.8	36.1	6.9	189
5SH;	Before	7.76	4.86	11.2	89	71	25.4	35.7	1.2	66
HbSC	After	5.49	3.46	9.8	218	72	24.6	34.0	1.1	109
Mean ± SEM	Before	12.46 ± 1.81†	6.39 ± 1.44‡	9.0 ± 0.6	300 ± 64	80 ± 4.6	28.2 ± 2.2	35.3 ± 0.8	4.2 ± 1.9	77 ± 9.1§
Mean ± SEM	After	9.99 ± 1.70†	4.18 ± 1.23‡	8.7 ± 0.4	371 ± 78	81 ± 5.0	28.4 ± 2.2	34.9 ± 0.6	5.4 ± 2.8	127 ± 18.0§

*WBC, total leukocyte count; NEUTR, segmented neutrophil count; HB, hemoglobin; RETIC, reticulocyte count; MCV, mean red cell volume; MCH, mean cell hemoglobin; MCHC, mean cell hemoglobin concentration; BILIR, total bilirubin; ARG, arginine values.
†P-value < 0.001 for mean paired decrease of 2.47 ± 0.27 k/μL.
‡P-value < 0.005 for mean paired decrease of 2.25 ± 0.29 k/μL;
§P-value <0.05 for mean paired increase of 48 ± 19 μMol/L.

Table 3. Visual Analog Scale Values of Illness, Leukocyte Counts, Hemoglobin Values, and Plasma Arginine Levels Before Initiation and at the End of Continued Near-Monthly Periods While on Continuous L-Citrulline Supplementation

Variable	No. of periods	VAS (0 to 5)	WBC* (k/μL)	NEUTR (k/μL)	HB (g/dL)	ARG (μMol/L)
AT;	Before 5	2.5	9.10	5.46	9.1	69
HbSS	Mean ± SEM	0.21 ± 0.09	8.27 ± 0.80	3.10 ± 0.68	9.0 ± 0.1	201 ± 26
AF;	Before 9	2.5	18.00	12.06	7.8	71
HbSS	Mean ± SEM	0.6 ± 0.6	15.31 ± 0.76	8.55 ± 0.51	7.1 ± 0.1	81 ± 7.9
TD;	Before 5	2.5	24.70	11.85	7.5	81
HbSS	Mean ± SEM	1.0 ± 1.0†	13.94 ± 1.38	7.04 ± 1.13	7.8 ± 0.2	175 ± 36
SH;	Before 5	2.5	5.31	3.08	10.2	118
HbSC	Mean ± SEM	0.1 ± 0.1	6.83 ± 0.94‡	3.97 ± 0.80	10.8 ± 0.2	162 ± 18

*VAS represents visual analog scale line values of general feelings of illness by each patient in each period of 4 to 6 weeks, based on O for very good, 2.5 for prior usual degree of illness, and 5 for very bad. WBC, total leukocyte count; NEUTR, segmented neutrophil count; HB, hemoglobin; ARG, arginine.
†Hospitalized for several days with one acute chest syndrome crisis during one period when WBC rose to 24.9 k/μL and plasma arginine level was found to be low at 48 μmol/L.
‡This patient had one episode of acute upper respiratory tract infection with WBC elevation to 10.3 k/μL at the end of one of the 5 period intervals.

DISCUSSION

It is problematic as to the mechanisms by which the ingestion of L-citrulline twice daily resulted in remarkable symptomatic improvements in these five patients with steady-state sickle cell disease. Although we cannot rule out the placebo effect, the considerable change in each patient's symptoms suggests more than a simple placebo effect. None of the five patients had contact with another patient receiving citrulline during the initial five sets of 28-day trial periods. In addition, in the subsequent four patient sets of continuous trial periods with supplementation, visual linear analog scale values, self-administered by the four patients at the end of each near-monthly period, showed that they maintained much improvement in feelings of well-being. This appears to confirm the presence of more than a simple placebo effect with citrulline intake. Visual linear analog scale values have been shown to be reliable in subject self-assessment for evaluating well-being and pain under various conditions.[18] Patient self-assessment analog scale scores have been used successfully before for well-being and pain in patients with sickle cell disease.[19,20]

The remarkable lessening of exertional fatigue and lessening of exertional dyspnea associated with sizeable improvement in left ventricular end-diastolic dimension from 5.2 to 4.8 cm (data not tabulated) and in a large reduction in left ventricular mass index (LVMI) of 25 g/m^2 in the patient, AT, over the initial 28-day interval with citrulline, strongly suggests that compensated cardiac functional impairment was present initially in this patient, which lessened with citrulline intake. In all four patients with HbSS disease, LVMI decreased somewhat with the initial 28-day course of citrulline (p-value < 0.10) (see Table 1).

In patient AT, there occurred measurable reductions in the high mean arterial blood pressure and resting high cardiac index towards a normal and more nearly normal value, respectively, with lowered peripheral vascular resistance (Table 1) at the end of the 28-day interval. These associations apparently indicate that a systemic vasodilator, antihypertensive effect was operational with the citrulline supplementation. Similarly, the other subjects may have had improved cardiac performance and systemic capillary nutrient perfusion performance with exercise. This may have been responsible for their lessening of exertional fatigue and improved sense of well-being and sense of more energy while on citrulline intake. Cardiomegaly and ventricular diastolic abnormalities are commonly early features in sickle cell anemia even without overt symptoms of decompensated heart failure or hypertension.[21-25]

Contributions from blood flow shunting with arteriovenous bypassing of certain systemic capillary vascular beds is present in the steady state of sickle

cell disease.[26,27] Such shunting with reduced cardiac afterloads are said to account for the increased resting cardiac outputs and cardiac indices found typically in sickle cell anemia disproportionate to the degree of anemia.[26-28] Unusually high indices also were found in our four subjects with sickle cell anemia (see Table 1).

Lonsdorfer et al.[28] showed that sickle cell anemia subjects in steady states are chronically maintained at the upper limit of physiological compensatory mechanisms with high systemic vasodilator activity when they are not in painful crises. Nitrate formation as an index of nitric oxide production is also increased in steady-state sickle cell disease.[29] It had been speculated by Waugh et al.[15] that the effective compensatory reduced after-load mechanisms include overactivity of the endogenous endothelial arginine-nitric oxide system.[13] This process may need to be maintained by a continuous supply of arginine as available plasma substrate for nutrient capillary flow and to avoid or lessen vasoocclusive crises.[14,15] With sufficient available plasma arginine for sufficient activity of endothelial nitric oxide synthase in producing vasoprotective nitric oxide as product, proinflammatory cytokines and adhesion molecules by the endothelium and leukocytes will be suppressed, as suggested by previous studies.[30] Exogenous sources for formation of nitric oxide can limit the degree of endothelial activation and leukocyte adhesion.[30] Cytokines such as tumor necrosis factor α and endothelin-1 are sometimes high in peripheral plasma in steady-state sickle cell disease.[31-33] Increased expression of proinflammatory cytokines may be manifested by significant fatigue.[34] Increasing evidence indicates that neutrophils are activated and play an important role in the initiation and propagation of vasoocclusive processes in sickle cell disease.[11,35]

There is much increased active bone marrow expansion with increased hematopoietic activity in sickle cell anemia, with increased vascularity and arteriovenous shunting of blood.[21,36] There is much increased bone marrow blood flow in sickle cell anemia.[37] The expanded bone marrow vascularity with increased hematopoiesis, as judged by increased reticulocytosis (see Table 2), may have accounted partly for the increased cardiac outputs and cardiac indices that were maintained during supplementation in the four patients studied with sickle cell anemia (Table 1). However, increased nutrient capillary perfusion and lessening of arteriovenous shunting in various parts of the body may have resulted because of improvement in the endothelial arginine-nitric oxide pathway.

The elevated cardiac outputs and cardiac indices typical of sickle cell anemia in the steady state may be considered analogous to the increased cardiac outputs and cardiac indices found in advanced Paget's disease of bone. When the hyperemic bone disease is generalized, Paget's bone disease is associated with blood being shunted through the affected bones.[38] Cardiac hyperkinesis analogous to that found with open arteriovenous communications results.[38] In sickle cell disease, sufficient activity of the constitutive endothelial L-arginine-nitric oxide pathway in bone marrow may be necessary to maintain bone marrow nutrient capillary blood flow without much stasis and vasoocclusive events. This trial with citrulline as arginine precursor suggests that citrulline supplementation may tend to prevent painful bone crises in sickle cell disease.

Increased plasma hemoglobin levels, which tend to vary with the degree of hemolysis in sickle cell disease, tend to correlate inversely with plasma arginine levels in sickle cell anemia, because of arginase being liberated from red cells in vivo when they are hemolyzed.[15] This may underlie the failure to elevate plasma arginine levels to much higher levels than control values with the amounts of citrulline used in our 1 patient with the greatest degree of reticulocytosis and hyperbilirubinemia, patient AF (see Table 2). Free hemoglobin in circulating plasma is also a very avid scavenger of formed nitric oxide as endothelium-derived vascular relaxing factor.[39] It is indeed relevant that inhibition of the usual activity of endothelial nitric oxide synthase enzyme leads to cerebrovascular adherence of human sickle red cells and vasoocclusive strokes when sickle red cells are injected in a rat model.[40]

This study showed that L-citrulline supplementation in patients with sickle cell disease reduced high total leukocyte counts and segmented neutrophil counts towards or to normal counts. Associated was symptomatic improvement in the patients. Administered orally, L-citrulline is an efficient precursor for the endogenous biosynthesis of L-arginine.[16,17,41] L-Arginine and formed nitric oxide participates importantly in the homeostatic control of the vasculature, is vasoprotective normally, inhibits cytokine formation and myointimal hyperplasias, and is involved in immunity.[13,42]

Therefore, further investigation of the likely beneficial effects of L-citrulline supplementation in sickle cell disease appears warranted using placebo-controlled studies. Citrulline may be synergistic even to the use of hydroxyurea in sickle cell disease. This drug appears to have part of its beneficial therapeutic effect due to myelosuppressive effects on peripheral total leukocyte and neutrophil counts.[6,43]

Citrulline therapy has been nontoxic, even in quite large doses of 5.7 g daily given chronically to a 4-year-old child with a dibasic amino acid defect.[44] It may be safe to administer citrulline even in the presence of infection or sepsis in sickle cell disease.[45,46] With infection or sepsis, there is lack of significant expression of activity of inducible nitric oxide synthase away from the nidus of infection.[46] It is believed that citrulline supplementation for palliation may eventually be shown to improve the quality of life in many thousands of African Americans suffering from serious sickle cell disease.

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