Rheumatology 2003;42:1550–1555
doi:10.1093/rheumatology/keg402, available online at www.rheumatology.oupjournals.org
Advance Access publication 27 June 2003

Preliminary report

Near-iron deficiency-induced remission of gouty arthritis

Francesco S. Facchini

***Objectives*. Previous evidence supports a role for iron in the pathogenesis of gout. For example, iron, when added to media containing urate crystals, stimulated oxidative stress with subsequent complement and neutrophil activation. Conversely, iron removal inhibited these responses as well as urate-crystal-induced foot pad inflammation in rats in-vivo. The objective of the present study was to investigate whether or not iron removal may improve the outcome of gouty arthritis in humans as well.**

***Methods*. Quantitative phlebotomy was used to remove iron in 12 hyperuricaemic patients with gouty arthritis and maintain their body iron at near-iron deficiency (NID) level (i.e. the lowest body iron store compatible with normal erythropoiesis and therefore absence of anaemia).**

***Results*. During maintenance of NID for 28 months, gouty attacks markedly diminished in every patient, from a cumulative amount of 48 and 53 attacks per year before (year –2, –1), to 32, 11 and 7 during induction (year 0) and maintenance (year +1, +2) of NID, respectively. During NID, attacks were also more often of milder severity.**

***Conclusions*. During a 28-month follow-up, maintenance of NID was found to be safe and beneficial in all patients, with effects ranging from a complete remission to a marked reduction of incidence and severity of gouty attacks.**

KEY WORDS: Gout, Arthritis, Hyperuricaemia, Oxidative stress, Near-iron deficiency.

From a pathogenetic standpoint, gout remains a mysterious disease. For example, although hyperuricaemia is very common, a normal serum uric acid concentration (SUAC) does not always rule out the diagnosis of gouty arthritis. Conversely, even though the prevalence of gouty arthritis increases with the severity of hyperuricaemia, some hyperuricaemic patients remain symptom-free for many years [1–3]. Likewise, albeit urate crystalline deposition is the pathological hallmark of gouty arthritis, uric acid crystals can be found in the synovial space between attacks, i.e. in the absence of inflammatory signs and symptoms [3].

Thus, it appears that hyperuricaemia and/or crystal deposition are not sufficient to cause acute gouty attacks and that some other factor(s) are necessary to trigger the inflammation.

Iron could be one candidate factor and such proposition is based upon data showing that human tophi and the synovial membrane contain iron [4], that urate crystals complexed iron cation in redox-active form at physiological pH [5] and that, following this process, there was a dose-dependent stimulation of oxidative stress, with granulocyte and complement activation and subsequent synthesis and release of pro-inflammatory lymphokines [5]. Iron removal, with the iron-specific chelator deferoxamine, fully prevented such responses in-vitro and decreased urate-crystal-induced foot-pad swelling in rats in-vivo [6]. Taken together, these results

Department of Medicine, Division of Nephrology, San Francisco General Hospital and University of California, San Francisco, CA 94143 and Department of Medicine, Kaiser Foundation Hospitals and the Permanente Medical Group Inc., Oakland, CA 94117, USA.

Submitted 30 January 2003; revised version accepted 8 April 2003.

Correspondence to: F. S. Facchini, Box 1341 UCSF, San Francisco, CA 94143-1341, USA. E-mail: fste2000@yahoo.com

support the hypothesis that iron might be a fundamental factor triggering gouty arthritis in humans as well.

Only high doses of deferoxamine suppressed urate-crystal-induced synovitis in rats [6], indicating that a threshold reduction of body iron stores was necessary to induce an anti-inflammatory effect. High-dose deferoxamine, however, carries the risk of significant ocular and cerebral toxicity. Quantitative phlebotomy on the other hand, does not have serious side effects and, when compared to deferoxamine, also has the advantage that the kinetics of iron removal is unaffected by size of body iron stores [7]. In other words, phlebotomy can achieve a more complete depletion of storage iron than deferoxamine, allowing for a better estimation of initial body iron burden [8]. In the present study, quantitative phlebotomy was therefore used to reduce iron stores in patients with gouty arthritis to near-iron deficiency (NID) [9], i.e. to that level where body iron is maximally depleted and yet sufficient to sustain normal erythropoiesis. Subsequently, NID was maintained for an average of 28 months to test whether or not further bouts of gouty arthritis could either be prevented or ameliorated.

Patients and methods

Twelve patients who satisfied inclusion criteria and agreed to participate in the current trial were studied. The study was approved by the local ethical committees, experimental procedures were in accordance with Institutional guidelines and written informed consent was obtained. All patients were hyperuricaemic, non-smokers and had a history of primary gout for >3 yr. Standard clinical criteria [10] yielding a specificity and sensitivity of ~90% in diagnosing acute gouty arthritis were used. Joint fluid aspiration demonstrating urate crystals had previously been performed in 8/12 patients. Exclusion criteria were a history of anaemia, heart, renal or liver failure, coronary heart disease, chronic infection, cancer of any kind and diseases due to disordered immunity, such as, for example, systemic lupus erythematosus, ulcerative colitis and rheumatoid arthritis. To ascertain health and exclude hyperuricaemia secondary to other pathologies, such as, for example, renal failure, psoriasis, haemolytic anaemia, myeloproliferative disease and certain neoplasms, all patients underwent a baseline physical examination and fasting measurement of uricaemia and routine chemistries. In the patients who qualified and agreed to participate, venesections were started to induce NID: half-litre phlebotomies were performed monthly or bimonthly under topical anaesthesia with 0.5–1.0 ml of 1% lidocaine. Iron indexes and a cell blood count (CBC) were measured before every phlebotomy by routine techniques to prevent anaemia and each patient's haematocrit (Hct) was not allowed to decrease $>5\%$ (from baseline values) by delaying the next phlebotomy as necessary. Phlebotomies were withheld at NID. As previously described [9], NID is the condition where body iron stores are still adequate to avoid anaemia, and yet substantially depleted as indicated by a serum ferritin of $\leq 30\ \mu g/l$, iron-saturation $\leq 15\%$ and a mean corpuscular cell volume (MCV) ≤ 82 fl. In these conditions, generally, there is negligible or absent stainable liver iron and total body iron stores are presumably in the order of 100–200 mg. To estimate baseline body iron stores (mg) the following conventional formula was used: (baseline Hct + NID Hct/2) × blood volume removed × 1 mg/ml [8].

After induction, NID was maintained by periodic venesections performed when monthly serum ferritin and iron saturation indexes increased above NID values.

Gouty attacks were defined as joint symptoms of sufficient entity to require an emergency room or outpatient urgent medical evaluation. They were estimated from medical record review and scored in three different ways.

First, by counting the number of attacks per year, independent of their severity, during the 2 yr prior to phlebotomy (years –2 and –1), during induction of NID (year 0) and in the subsequent 2 yr when NID was maintained (years +1 and +2).

Second, the mean change in the individual frequency of gouty attacks was estimated. For this analysis, attack frequency before (year –2, –1) and after (year +1, +2) was averaged and the mean attack rate per person per year before and after NID calculated.

Third, to establish whether arthritis was mild, moderate or severe, every attack was graded as follows: 1, pain; 2, pain plus erythema; 3, as for 2 plus clinically documented oedema; 4, as for 3 plus complete or nearly complete transient functional impairment; 5, development of deformities.

Data are presented as means ± S.D. Two-tailed paired Student's *t*-test and the Wilcoxon matched pairs rank test were used for difference in parametric and non-parametric variables, respectively. Differences were considered significant at a probability level of $\geq 95\%$.

Results

The patients' clinical and demographic characteristics are shown in Table 1. Body weight and ethanol intake were unchanged. Steroids, either intra-articular or systemic, were never administered. Allopurinol average dose was 180 ± 62 mg/day, not significantly different than 24 months before starting phlebotomies (203 ± 50 mg/day). Body iron status at baseline and follow-up is also illustrated in Table 1. NID was successfully achieved within 8 ± 3 months and thereafter maintained in all patients without significant adverse reactions. The average number of phlebotomies necessary to achieve and maintain NID was 7 ± 2, 3 ± 1, 3 ± 1, respectively, for years 0, 1 and 2. Initial body iron stores were 1.8 ± 0.6 g.

TABLE 1. Demographic, biochemical and clinical data before (Year –2), at start of quantitative phlebotomy (Baseline) and after 2 yr of NID (Year +2)

Variables	Year –2	Baseline	Year +2
Age (years)	50 ± 6	52 ± 6	54 ± 6
BMI (kg/m^2)	28 ± 2	28 ± 2	29 ± 3
Gender male	10/12	10/12	10/12
Ethanol intake (g/day)	12 ± 4	14 ± 5	15 ± 6
Ferritin (μg/l)	287 ± 81	301 ± 98	26 ± 10*
Iron saturation (%)	44 ± 13	45 ± 12	13 ± 2*
MCV (fl)	90 ± 3	89 ± 4	80 ± 2*
Hct (%)	45 ± 2	44 ± 2	43 ± 1**
SUAC (mmol/l)	0.46 ± 0.1	0.50 ± 0.1	0.47 ± 0.1
Allopurinol use	11/12	11/12	11/12
Allopurinol dose (mg/day)	203 ± 50	194 ± 76	180 ± 62
Thiazide diuretic use	4/12	4/12	4/12
Thiazide dose (mg/day)	27 ± 18	30 ± 17	24 ± 15

*$P < 0.001$; **$P < 0.05$.

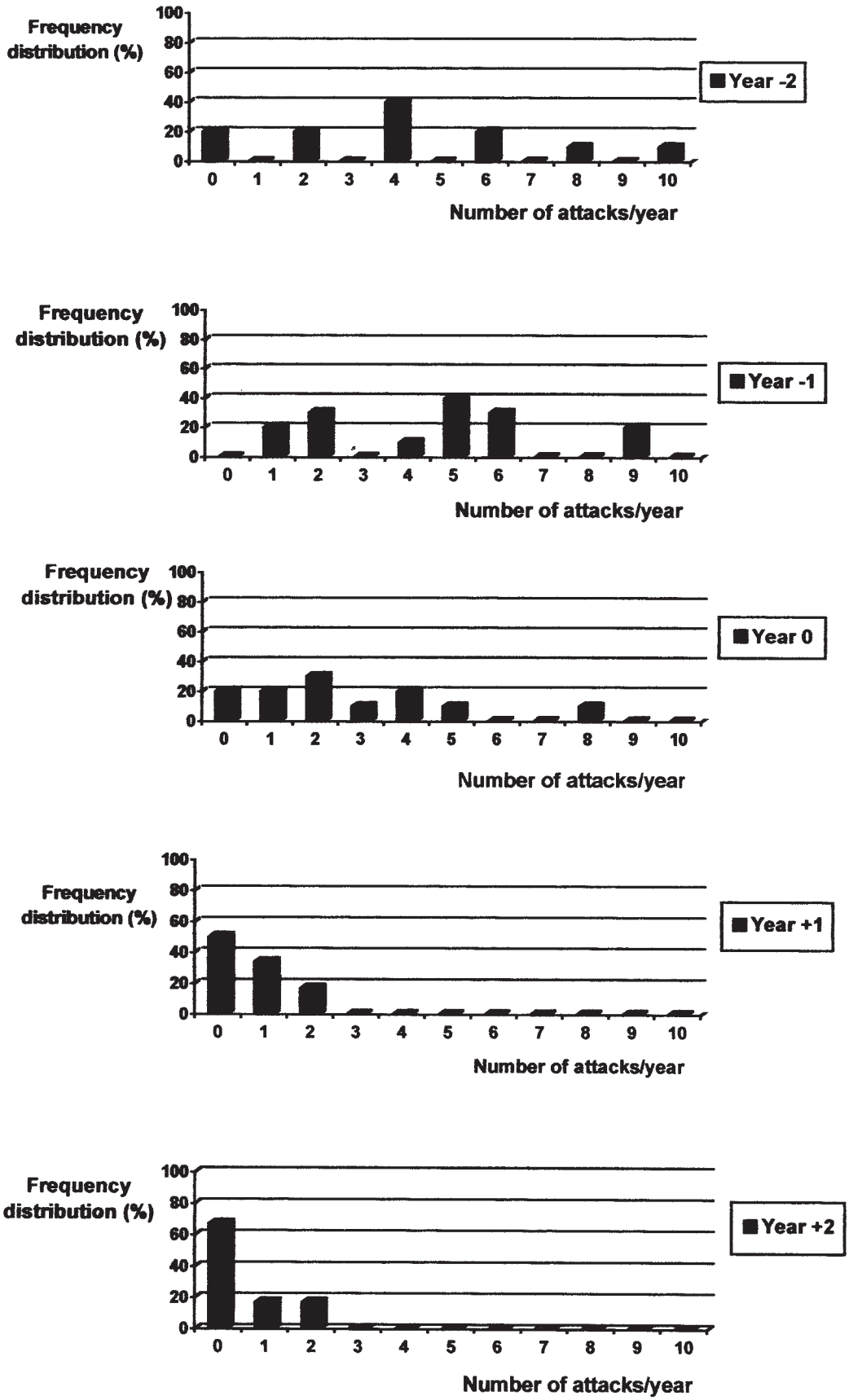

FIG. 1. Frequency distribution of cumulative attack rates during the study period.

There was a cumulative amount of 48 and 53 gouty attacks per year during the 2 yr prior to study entry (years –1 and –2); this diminished to 32, 11 and 7 attacks per year during years 0, 1 and 2, respectively. Percent changes in cumulative attack rate are shown in Fig. 1. In Fig. 2, it can be seen that the intra-individual change in the average number of gouty attack per year decreased from 6.4 ± 3.0 to 2.0 ± 1.0 after achievement of NID ($P < 0.001$). In absolute terms, 2/12 and 7/12 patients became attack-free during year 0 and the subsequent 24 months, respectively. These patients, although they continued to take allopurinol, did not require any further anti-inflammatory therapy, with either non-steroidals or colchicine. In Fig. 3 the median frequency distribution of the gouty arthritis severity score is shown before (years –2 and –1), during (year 0) and after achievement of NID in all patients (years 1 and 2).

Discussion

In the present investigation, body iron stores were lowered by means of serial venesections to the lowest values yet compatible with normal erythropoiesis (NID) and NID subsequently maintained for 28 months. NID induced either a full or partial remission of gout in all study patients that persisted for the entire period of observation. Since spontaneous remissions in the absence of obvious confounding factors (e.g. dietary changes, weight loss and medications) are unlikely, it is possible to conclude that NID prevented the relapse of the acute arthritis of primary gout in 58% of patients and markedly reduced its frequency and severity in the remaining 42%.

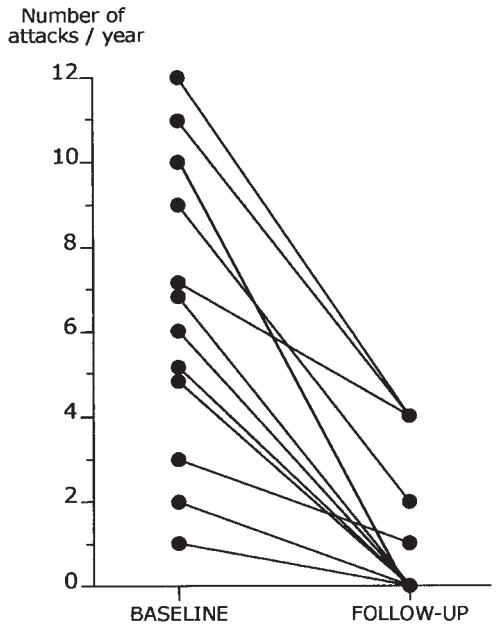

FIG. 2. Mean attack rate per person per year before (baseline) and after (follow-up) NID. See text for details.

In former studies of iron depletion in animals, high-dose deferoxamine could mitigate, but not prevent, urate-crystal-induced foot-pad swelling, but only at high doses. Lower doses of deferoxamine had no anti-inflammatory effect [6]. Deferoxamine-mediated iron excretion decreases as body iron stores shrink [7], while during quantitative phlebotomy the kinetics of iron removal is unaffected by size of body iron stores. Therefore, when compared to those results obtained in rats treated with high-dose deferoxamine, the better outcome shown in the present study is presumably related to a greater degree of depletion of body iron stores.

The present findings were not unexpected and are in agreement with results from in-vitro studies showing the pro-inflammatory role urate crystals have when they complex iron. While soluble urate has antioxidant properties by coordinating iron in a redox-inactive form, crystalline urate binds iron loosely, permitting the crystal-bound metal to become a Fenton's catalyst and catalyse the production of reactive oxygen species [5]. Thus, prior exposure of urate crystals to ferric iron salts markedly enhanced deoxyribose oxidation while either hydroxyl radical scavengers or deferoxamine suppressed it [5]. After iron complexation, urate crystal-mediated oxidant generation stimulated neutrophils and production of Leukotriene B4 (LTB4), a potent cytotaxin and an important chemical mediator in acute gouty attacks [11]. Metal-catalysed oxidant generation also activated complement, a further chemotactic and activating stimulus for neutrophils [12]. Ferric iron loading (of the crystals) enhanced, in a dose-dependent fashion, neutrophil chemotaxis and all related inflammatory responses while deferoxamine or hydroxyl radical scavengers suppressed them [5].

Interestingly, hyperinsulinaemia and insulin resistance also induce oxidative stress and recent data showed carbohydrate restriction, an intervention known to reduce insulin resistance and hyperinsulinaemia, was indeed quite effective in curbing the frequency of gouty attacks [13]. Iron lowering to NID markedly improves both oxidative stress [14] and insulin resistance [9] and this dual action might explain the greater effect of NID, as compared with that of carbohydrate restriction, on the incidence and severity of gouty attacks. However, neither insulin resistance nor inflammatory pathway activation were evaluated, and therefore whether similar mechanisms influenced the outcome of the present study or not is pure speculation. Nonetheless, although preliminary, the effect of NID shown is congruous with the notion that oxidative stress and gout can be promoted by increasing amounts of iron in-vitro, and prevented or blunted by use of iron chelators in-vitro, as well as in animals in-vivo [6].

There is the possibility that arthritic symptoms were not always related to re-exacerbation of gout. One limitation of the present study concerns the issue that severity assessment of gouty flares is typically based on far-from-perfect criteria. Inter-individual variability in pain threshold, in doctors' documentation of patients'

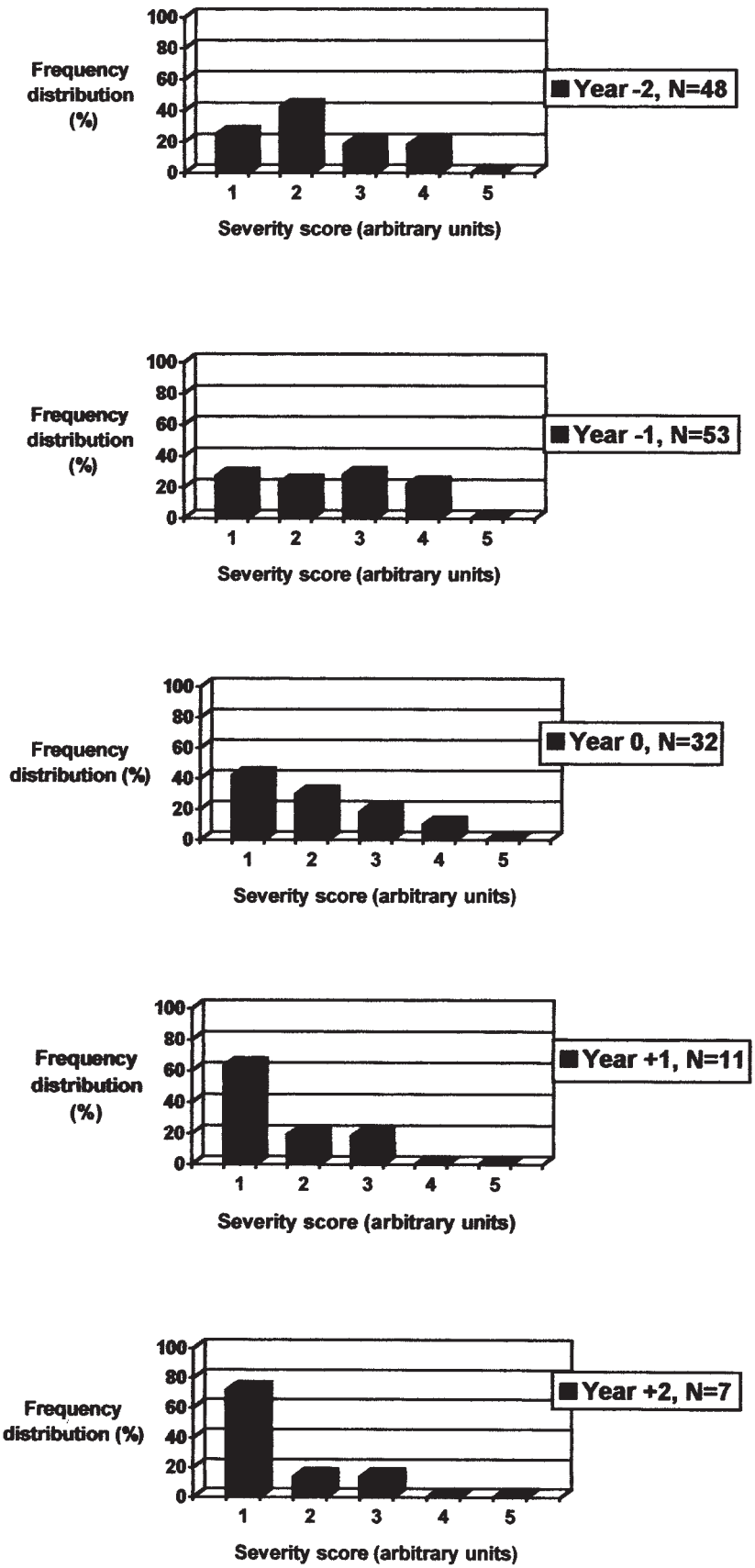

FIG. 3. Frequency distribution of joint inflammation score (for details see methods section) during the study period. N = total number of attacks.

symptoms and signs and the possibility of misdiagnosis all exist. However, the frequency of flares unequivocally decreased as documented by the overall reduction of cumulative (Fig. 1) and individual (Fig. 3) clinic appointments and such findings are hard to dispute. The issue of diagnostic accuracy is also important. Stringent diagnostic criteria, based on synovial fluid analysis, were in fact used only in 8/12 patients. However, urate crystals are not always present in acute gout. Furthermore, it was shown that iron removal was also effective in other types of experimental arthritis such as, for example, in rheumatoid [15] and adjuvant-induced arthritis, even when iron depletion was achieved by means of iron-poor diets [16]. Therefore, the fact that all patients improved, including the 1/10 possibly affected by a condition other than gout [10], indicates the likelihood that NID may lessen other types of joint inflammation as well.

At this time, it seems premature to suggest widespread use of venesections to NID as a treatment modality for gout. However, considering the growing pathogenetic importance of iron sufficiency in a variety of chronic age-related disease, from type 2 diabetes and its complications [9, 17–19], to atherosclerosis [20], chronic hepatitis C [21] and colon cancer [22] it seems obvious that, at the very least, there is an urgent need to overcome the dogmatic credence that appreciable amounts of iron should always be maintained in storage at any age and by all means.

Rheumatology	Key Messages
	1. Depletion of body iron stores to a level of NID by means of quantitative phlebotomy had a sustained beneficial effect on clinical gouty arthritis. 2. During a 28-month follow-up period NID was tolerated without adverse effects. If confirmed, these results should strongly encourage NID as a cost-effective and safe therapeutic option for gouty arthritis.

Acknowledgement

The author wishes to thank Elizabeth Vidal for the skilled graphical assistance.

References

1. Hall AP, Barry PE, Dawber TR, McNamara PM. Epidemiology of gout and hyperuricemia. Am J Med 1967;42:27–37.
2. Scott JT. Gout. Bailliere's Clin Rheumatol 1987;3: 525–46.
3. Agudelo CA, Weinberger A, Schumacher HR, Turner R, Molina J. Definitive diagnosis of gout by identification of urate crystals in asymptomatic metatarsophalangeal joints. Arthritis Rheum 1979;22:559–60.
4. Muirden KD, Senator GB. Iron in the synovial membrane in rheumatoid arthritis and other joint diseases. Ann Rheum Dis 1968;27:38–47.
5. Ghio AJ, Kennedy TP, Rao G, Cooke CL, Miller MJ, Hoidal JR. Complexation of iron cation by sodium urate crystals and gouty inflammation. Arch Bioch Biophys 1994;313:215–21.
6. Blake DR, Hall ND, Bacon PA, Dieppe PA, Halliwell B, Gutteridge JMC. Effect of a specific iron-chelating agent on animal models of inflammation. Ann Rheum Dis 1983;42:89–93.
7. Hallberg L, Hedenberg L. Deferoxamine-induced iron excretion in normal and iron deficient subjects. Scand J Hematol 1967;4:11–20.
8. Haskins D, Stevens AR, Finch S. Iron stores in man as measured by phlebotomy. J Clin Invest 1952;31:543–7.
9. Facchini FS, Hua NW, Stoohs RA. Effect of iron depletion in carbohydrate-intolerant patients with clinical evidence of nonalcoholic fatty liver disease. Gastroenterology 2002; 122:931–9.
10. Wallace SL, Robinson H, Masi AT, Decker JL, McCarty DJ, Yu T-F. Preliminary criteria for the classification of the acute arthritis of primary gout. Arthritis Rheum 1977;20:895–900.
11. Rae SA, Davidson EM, Smith MJ. Leukotriene B4, an inflammatory mediator in gout. Lancet 1982;2(8308):1122–4.
12. Vogt W, Damerau B, von Zabern I, Nolte R, Brunahl D. Non-enzymic activation of the fifth component of human complement, by oxygen radicals. Some properties of the activation product, C5b-like C5. Mol Immunol 1989;26: 1133–42.
13. Dessein PH, Shipton EA, Stanwix AE, Joffe BI, Ramkgadi J. Beneficial effects of weight loss associated with carbohydrate restriction and increased proportional intake of protein and unsaturated fat on serum urate and lipoprotein levels in gout: a pilot study. Ann Rheum Dis 2000;59:539–43.
14. Sullivan JL, Till G, Ward PA, Newton RB. Nutritional iron restriction diminishes acute complement-dependent acute lung injury. Nutr Res 1989;9:625–34.
15. Dabbagh AJ, Trenam CW, Morris CJ, Blake DR. Iron in joint inflammation. Ann Rheum Dis 1993;52:67–73.
16. Andrews FJ, Morris CJ, Lewis EJ, Blake DR. Effect of nutritional iron deficiency on acute and chronic inflammation. Ann Rheum Dis 1987;46:859–65.
17. Fernandez-Real JM, Penarroja G, Castro A, Garcia-Bragado F, Hernandez-Aguado I, Ricart W. Blood letting in high-ferritin type 2 diabetes. Diabetes 2002;51:1000–4.
18. Facchini FS. The iron factor of aging. Tucson, AZ: Wheatmark Inc., 2002.
19. Facchini FS, Saylor KL. A low-iron available, polyphenol-enriched, carbohydrate-restricted diet to slow progression of diabetic nephropathy. Diabetes 2003; 352:1204–9.
20. Sullivan JL. Are menstruating women protected from heart disease because of, or in spite of, estrogen? Am Heart J 2003;145:199–203.
21. Yano M, Hayashi H, Wakusawa S *et al.* Long-term effects of phlebotomy on biochemical and histological parameters of chronic hepatitis C. Am J Gastroenterol. 2002;97:133–7.
22. Sullivan JL. Re: Association between hemochromatosis (HFE) gene mutation carrier status and the risk of colon cancer. J Natl Cancer Inst 2003; In press.