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Use of Desferoxamine and S-Adenosylmethionine to Treat Hemochromatosis in a Red Ruffed Lemur (*Varecia variegata ruber*)

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Hemochromatosis was diagnosed in a 14-year-old, male, red ruffed lemur (*Varecia variegata ruber*) on the basis of abnormal results of serum biochemical analysis, including high serum ferritin and transferrin saturation values, and of liver biopsy. Therapy included chelation, using desferoxamine to remove excess iron and S-adenosylmethionine to improve liver function, and monthly peripheral blood removal by phlebotomy to reduce total body iron content. Response to treatment was assessed by changes in the lemur's attitude and appetite, as well as variations in serum biochemical and iron panel values. Initial improvement was associated with the onset of therapy. After 56 days of treatment, results of serum biochemical analysis indicated a decrease in iron panel values Treatment was temporarily discontinued from days 56 to 65, and the lemur's condition worsened, so therapy was re-instituted. However, the lemur died of hepatocellular carcinoma on day 110 of treatment.

Hemochromatosis, the deposition of excess amounts of iron in parenchymal organs, with eventual tissue damage and impaired function of organs, was first recognized more than a century ago in humans (28). It was first described in lemurs in the late 1960s, and is suspected to be, in part, the result of dietary derangements (15, 16, 34). Although adjustments in diets have reduced the prevalence of hemochromatosis in lemurs in the last decade, the disease is still diagnosed in captive animals.

The goal in treating hemochromatosis is to normalize the total body load of iron. In human patients, treatment is best accomplished by reducing blood volume, using weekly phlebotomy to lessen the iron load (1, 12, 29). In cases where hemochromatosis coexists with chronic anemia or other blood disorders that prevent use of phlebotomy, iron chelation may be the best available therapy (18, 29). Although it is less effective than phlebotomy, chelation therapy is less invasive and can be routinely performed in animals that require anesthesia and/or sedation to undergo phlebotomy. Desferoxamine mesylate (DFO; Desferal, Novartis, E. Hanover, N.J.), a naturally occurring trihydroxamic acid produced by *Streptomyces pilosus*, is a chelating agent routinely used to remove excess iron from the body. (3, 10, 13). Administration of S-adenosylmethionine (SAMe; Denosyl, Nutraceutics Corporation, St. Louis, Mo.) has reportedly improved hepatocellular function in various species (2, 4). We describe intramuscular administration of DFO and oral administration of SAMe as viable therapy for hemochromatosis in a red ruffed lemur (*Varecia variegata ruber*).

Case Report

In December of 2001, a 14-year-old, male-red ruffed lemur that was housed with a 13-year-old, healthy, female red ruffed lemur in an indoor/outdoor enclosure and was fed a low-iron, low-vitamin C diet, presented with history of weight loss and decreased appetite. The lemur was anesthetized by intramuscular administration of ketamine (15.3 mg/kg of body weight; Ketaset, Fort Dodge Animal Health, Overland Park, Kans.) and midazolam (0.5 mg/kg; Versed, Baxter Pharmaceutical Products Inc., New Providence, N.J.). Anesthesia was maintained with 2% isofluorane (Forane, Baxter Pharmaceutical Products Inc.). Physical examination revealed a thin animal, but was otherwise unremarkable. There was no evidence of hepatomegaly, splenomegaly, joint disease, or excess skin pigmentation. Complete blood cell count values were within normal range. In contrast, serum biochemical analysis revealed hyperbilirubinemia and high bile acids concentration. Serum iron concentration, total iron-binding capacity (TIBC), and transferrin saturation percentage were markedly high (Table 1). Moderate increase of alkaline phosphatase and creatine kinase activities also was observed (data not shown).

A liver biopsy was performed one month later. To perform the procedure, medetomidine (40 µg/kg; Domitor, Pfizer, Exton, Pa.), butorphanol (0.40 mg/kg; Torbugesic Inj., Fort Dodge Animal Health), and midazolam (0.5 mg/kg) were given intramuscularly to the lemur. Physical examination once again revealed a thin animal, while laboratory results yielded no appreciable change in hematologic and serum biochemical analysis results other than a markedly high serum ferritin concentration (Table 1). Four liver biopsy specimens were obtained using a Wolfe biopsy forceps. Histologic examination of the liver revealed disrupted architecture, with regenerating nodules and thick bands of fibrosis and bile duct hyperplasia in the portal areas (Fig. 1). Using a Prussian blue stain, large amounts of iron were observed in macrophages in the periportal areas, with lesser amounts of iron in hepatocytes (Fig. 2). Clinical and pathologic findings were indicative of hemochromatosis associated with early stages of liver cirrhosis.

A multi-component plan combining serial phlebotomy, chelation therapy, and treatment for hepatic insufficiency was initiated. The iron chelator DFO was administered intramuscularly at a dosage of

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Table 1. Summary of hematologic and iron panel values obtained from a red ruffed lemur (*Varecia variegata ruber*) under treatment for hemochromatosis

Blood parameter	Reference range	12/01	Day 1	Day 33	Day 56	Day 110
Serum ferritin (ng/ml)	8-171[a]	NA	237	92	144	198
Transferrin saturation (%)	29-91[a]	100	NA	83	88.5	91.5
TIBC (µg/dl)	208-379[a]	409	NA	355	341	309
Serum iron (µg/dl)	90-318[a]	409	NA	297	302	283
Bile acids (µmol/L)	NA	40.6	NA	NA	22.7	NA
Total bilirubin (mg/dl)	0.0-3.4[b]	4.3	2.1	2.7	2.3	16.0

[a]Values for Black and White ruffed lemurs (*Varecia variegata variegata*) > 10 years-old, provided by Graham Crawford DVM, San Francisco Zoo.
[b]International Species Information System. Ruffed lemur (*Varecia variegata*) males > 2 year-old.
TIBC = total iron-binding capacity; NA = not available.

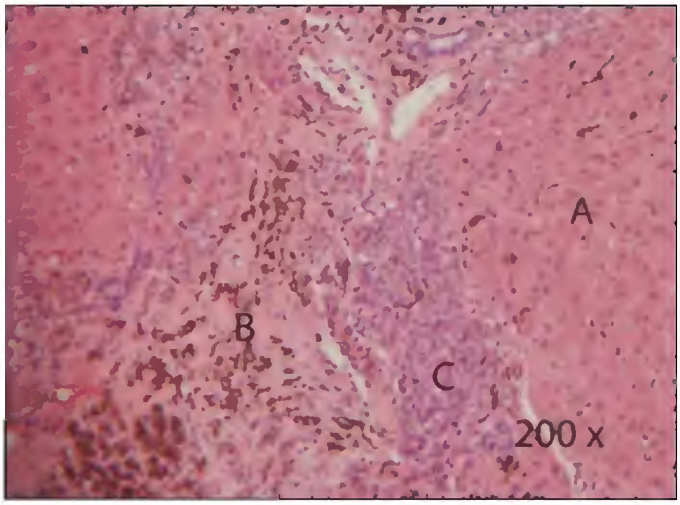

Figure 1. Photomicrograph of a section of liver from a biopsy specimen obtained from a lemur with hemochromatosis. Notice regenerative nodules (A) with portal fibrosis (B) and bile duct hyperplasia (C). H&E stain; magnification, 200×.

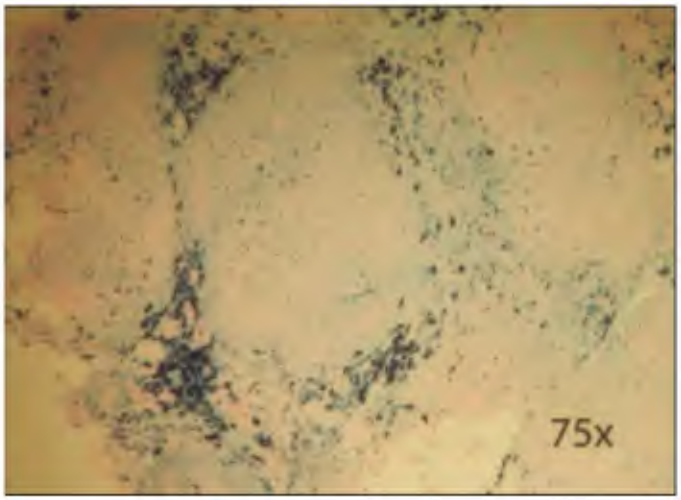

Figure 2. Lower-magnification photomicrograph of the section of liver from Figure 1. Noticed nodular pattern, with densely blue-staining, iron-laden macrophages in the portal areas. Prussian blue stain for iron; magnification, 75×.

10 mg/kg every other day for four weeks. Phlebotomy (10 ml/kg) was scheduled at monthly intervals, and SAMe at a dosage of 30 mg/kg given orally once a day was added to the treatment regimen to improve liver function. Metronidazole (15 mg/kg; Flagyl, G.D. Searle & Co., Chicago, Ill.) was given orally for 14 days as therapy for possible bacterial involvement.

On day 42, DFO administration was decreased to twice a week, and on day 49, it was decreased to once a week. Desferoxamine administration was temporarily suspended between days 56 and 65 due in part to poor patient compliance to injection and concern that the polyuria and polydypsia that had developed may have been caused by the medication. However, DFO administration was re-instituted on the basis of the initial improvement that had been observed in the lemur's condition and serum biochemical values (decreases in: serum ferritin concentration, from 237 ng/ml to 144 ng/ml; transferrin saturation, from 100% to 88.5%; TIBC, from 409 µg/dl to 341 µg/dl; serum iron concentration, from 409 µg/dl to 341 µg/dl; total bilirubin concentration, from 4.3 mg/dl to 2.3 mg/dl; and bile acids concentration, from 40.6 µmol/L to 22.7 µmol/L); polyuria and polydypsia were attributed to being a clinical sign of liver disease. Administration of SAMe was continuous throughout the treatment.

On day 110 of treatment, the lemur was anesthetized for follow-up blood tests and physical examination. The lemur was depressed, and mucous membranes were yellow-green tinged. The serum biochemical profile indicated severe hyperbilirubinemia (Table 1), and the lemur died 12 h later. Pathologic findings were indicative of marked hepatic insufficiency and ascites associated with liver cirrhosis, ischemic necrosis, and hepatocellular carcinoma. The carcinoma was extensive, but did not extend beyond the liver. There were no additional relevant pathologic changes.

Discussion

A number of bird and mammal species are known to accumulate excess amounts of iron under captivity conditions (21, 23, 25, 30). In humans, hemochromatosis is considered the most common type of primary iron overload disease (IOD) (27, 29). Primary IOD, which is hereditary, stems from an inherent defect in iron regulation that results in continuous over-absorption of iron from the gsastrointestinal tract (12, 27). Hemochromatosis has historically been considered a common problem in lemurs in captivity (15, 17, 31, 34). Development of hemochromatosis in lemurs and birds is thought to be multifactorial, with dietary derangements (a combination of excess dietary iron, excess ascorbic acid intake, and lack of tannins in the diet), genetics, and environmental factors as possible contributing causes (9, 17, 31). In this case, the cause of hemochromatosis was unclear as the lemur had been fed a low-iron, low-vitamin C diet, and its long-term cage mate remained unaffected.

Diagnosis of hemochromatosis in human patients is based on physical examination findings, genetic analysis, blood tests, and possible liver biopsy results (27, 29). Although a complete iron panel (serum iron concentration; TIBC; transferrin, serum ferritin, and serum transferrin receptor values; and transferrin iron

saturation percentage) may be necessary to diagnose hemochromatosis, two tests in particular, determination of serum ferritin concentration and transferrin saturation percentage are considered the most reliable methods to detect high tissue iron content. (1, 27, 28). Increase of these values in the absence of other causes of iron overload (transfusion, oral iron supplementation) is strongly suggestive of hemochromatosis.

Severe hemochromatosis was diagnosed in this lemur on the basis of high bile acids concentration, hyperbilirubinemia, and high serum ferritin and serum iron concentrations, and high percentage of transferrin saturation, along with hepatic histologic changes and high iron deposition observed in the liver biopsy specimen. In humans, deposition of iron targets several systems (liver, heart, pancreas, skin, and skeletal) (1, 3, 29). In animals, clinical signs of the disease usually are related to hepatic dysfunction (21). Specifically, lemurs have high prevalence of liver disease characterized most often by hepatocellular necrosis, distorted architecture, and bile duct hyperplasia (15, 17, 21, 31, 34). In this case, hepatic cirrhosis and carcinoma had developed, as noted at necropsy.

Use of phlebotomy for the treatment of iron overload in humans, domestic animals, and birds is well documented in literature (1, 12, 24, 25, 29, 35). However, in the case of this lemur, serial phlebotomy would have required multiple anesthesia events, and was, thus, considered impractical. Instead, therapy was based on iron chelation with DFO, liver support with SAMe, and monthly phlebotomy to reduce total body iron content and to monitor progress. Desferoxamine is the only iron chelator approved in North America and has been successfully used for years in humans, birds, and domestic animals (3, 7, 8, 10, 12, 13, 18, 26).

Although it is not as effective as phlebotomy to achieve rapid removal of excess iron from the body, long-term therapy with DFO slows accumulation of hepatic iron and retards or eliminates progression of fibrosis. In contrast, hepatic cirrhosis due to iron overload rarely resolves in response to therapeutic phlebotomy only (1). The dosage and route of administration of DFO in this lemur were extrapolated from those used to treat chronic iron toxicosis in children under three years of age. Instead of daily administration, DFO was administered every other day because of its potential adverse effects (cardiac, ocular, or auditory abnormalities or cerebral neurotoxicosis) and limited experience with it in non-human primates (21). Adverse effects attributed to DFO administration were not noticed during treatment of this lemur.

S-Adenosylmethionine was administered to help maintain and protect liver function. This medication has been documented to support liver function in a variety of species, including humans, rodents, dogs, and other non-human primates, without inducing cytotoxic effects or relevant adverse effects (2, 4-6, 14, 19, 20, 32, 33). The SAMe contributes to three major biochemical pathways—transmethylation, transsulfuration, and aminopropylation—all of which are essential functions of hepatocytes (11, 19, 22). The SAMe is also a precursor of glutathione, which is a major component of the antioxidant defense system and, thus, may have antioxidant properties (4, 32).

Transferrin saturation percentage, serum iron and serum ferritin concentrations, and TIBC were monitored during treatment. While this lemur was receiving DFO, ferritin, serum iron, and TIBC values initially decreased to near-normal limits after 30 days of treatment. These laboratory findings correlated with improvement in clinical signs of the disease. However, cessation of DFO therapy resulted in the return of an increase in the aforementioned values. On the basis of activity level, appetite, and interaction with keepers, subjectively the lemur's condition improved when SAMe therapy was instituted.

Although this lemur initially improved in response to treatment, during the fifteenth week after treatment, its condition worsened, and on day 110, it died. On postmortem examination, hepatocellular carcinoma was diagnosed as the cause of death.

Hepatocellular carcinoma is reported to be one of the principal causes of death in untreated humans patients with hemochromatosis, and thus, it was not an unexpected finding (1, 12, 29). Although this lemur did not survive, the authors believe that the treatment would have been more successful had it been initiated earlier in the course of disease. Furthermore, the improvement in the serum iron panel and liver function values, and clinical signs of disease warrant further investigation into this protocol as a potential treatment for lemurs with hemochromatosis.

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References

1. **Barton, J. C., S. M. McDonnell, P. C. Adams, P. Brissot, L. W. Powell, C. Q. Edwards, J. D. Cook, and K. V. Kowdley.** 1998. Management of hemochromatosis. Hemochormatosis management working group. Ann. Intern. Med. **129:**932-939.
2. **Bray, G. P, J. M. Tredger, and R. Williams.** 1992. S-adenosylmethionine protects against acetaminophen toxicity in mice. Hepatology **15(2):**297-301.
3. **Brittenham, G. M., P. M. Griffith, A. Nienhuis, C. E. McLaren, N. S. Young, E. E. Tucker, C. J. Allen, D. E. Farrel, and J. W. Harris.** 1994. Efficacy of desferoxamine in preventing complications of iron overload in patients with thalassemia major. N. Engl. J. Med. **331(9):**567-573.
4. **Center, S. A.** 2000. S-adenosyl-methionine (SAMe) an antioxidant and anti-inflammatory nutraceutical. Proc. Am. Coll. Vet. Intern. Med. **18:**550-552.
5. **Center, S. A., J. F. Randolph, K. Warner, W. E. Hoffman, J. McCabe, and P. Foureman.** 2000. Influence of SAMe on erythrocytes and liver tissue in healthy cats. Proc. Am. Coll. Vet. Intern. Med. **18:**734.
6. **Center, S. A., K. Warner, W. E. Hoffman, P. Foureman, and J. McCabe.** 1999. Influence of S-adenosylmethionine on metabolic and morphologic hepatocellular features induced by chronic glucocorticoid administration in dogs. Proc. Am. Coll. Vet. Intern. Med. **17:**719.
7. **Cork, S. C.** 2000. Iron storage diseases in birds. Avian Pathol. **29:**7-12.
8. **Cornelissen, H., R. Ducatelle, and S. Roels.** 1995. Successful treatment of a channel-billed toucan (*Ramphastos vitellinus*) with iron storage disease by chelation therapy: sequential monitoring of the iron content of the liver during the treatment period by quantitative chemical and image analysis. J. Avian Med. Surg. **9:**131-137.
9. **Cornelius, C. E.** 1988. Animal models, p. 1316-1336. *In* I. M. Arias, W. B. Jakoby, H. Popper, D. Schachter, and D. A. Shafritz (ed.), The liver. Biology and pathobiology, 2nd ed. Raven Press, New York.
10. **Cumming, R. L. C., J. A. Millar, J. A. Smith, and A. Goldberg.** 1969. Clinical and laboratory studies on the action of desferoxamine. Br. J. Haematol. **17:**257-263.

11. **Davidson, G.** 2002. S-Adenosylmethionine. Compend. Contin. Educ. Pract. Vet. **24 (8):**600-603.
12. **Felitti, V. J., and E. Beutler.** 1999. New developments in hereditary hemochromatosis. Am. J. Med. Sci. **318(4):**257-268.
13. **Franchini, M., G. Gandini, M. de Gironcoli, A. Vassanelli, C. Borgna-Pignatti, and G. Aprili.** 2000. Safety and efficacy of subcutaneous bolus injection of desferoxamine in adult patients with iron overload. Blood. **95(9):**2776-2779.
14. **Frezza, M. and M. Terpin.** 1992. The use of S-adenosyl-L-methionine in the treatment of cholestatic disorders. Drug Invest. **4(Suppl 4):**101-108.
15. **Gonzalez, J., K. B. Benirschke, P. Saltman, J. Roberts, and P. T. Robinson.** 1984. Hemosiderosis in lemurs. Zoo Biol. **3:**255-265.
16. **Junge, R. E.** 1997. Diseases of lemurs in captivity. Proc. Annu. Conf. Am. Assoc. Zoo Vet. Houston **1:**235-239.
17. **Junge, R. E.** 1999. Diseases of prosimians, p. 365-368. *In* M. E. Fowler and R. E. Miller (ed.), Zoo and wild animal medicine, current therapy 4. W. B. Saunders Co., Philadelphia.
18. **Kontoghiorghes, G. J., K. Pattichi, M. Hadjigavriel and A. Kolnagou.** 2000. Transfusional iron overload and chelation therapy with desferoxamine and deferiprone (L1). Transfusion Sci. **23 (3):**211-23.
19. **Lieber, C.** 2002. S-Adenosyl-L-methionine and alcoholic liver disease in animal models: implications for early intervention in human beings. Alcohol. **27:**173-177.
20. **Lieber, C. S., A. Casini, L. DeCarli, C. Kim, N. Lowe, R. Sasaki, and M. A. Leo.** 1990. S-Adenosyl-L-methionine attenuates alcohol-induced liver injury in the baboon. Hepatology. **11(20):**165-172.
21. **Lowenstine, L. J., and L. Munson.** 1999. Iron overload in the animal kingdom, p. 260-268. *In* M. E. Fowler, and R. E. Miller (ed.), Zoo and wild animal medicine, current therapy 4. W. B. Saunders Co., Philadelphia.
22. **McClain, C. J., D. B. Hill, Z. Song, R. Chawla, W. H. Watson, T. Chen, and S. Barve.** 2002. S-Adenosylmethionine, cytokines, and alcoholic liver disease. Alcohol **27:**185-192.
23. **Miller, G. F., D. E. Barnard, R. A. Woodward, B. M. Flynn, and J. W. M. Bulte.** 1997. Hepatic hemosiderosis in common marmosets *Callithrix jacchus*: effect of diet on incidence and severity. Lab. Anim. Sci. **47:**138-142.
24. **Morris, P. J., S. E. Avergis, and R. E. Baumgartner.** 1989. Hemochromatosis in a greater Indian hill mynah (*Gracula religiosa*): case report and review of the literature. J. Assoc. Avian Vet. **2:**87-92.
25. **Pearson, E. G., O. R. Hedstrom, and R. H. Poppega.** 1994. Hepatic cirrhosis and hemochromatosis in three horses. J. Am. Vet. Med. Assoc. **204(7):**1053-1056.
26. **Porter, J. B.** 2001. Desferoxamine pharmacokinetics. Semin. Hematol. **38(1):** 63-68.
27. **Powell, L. W., D. K. George, S. M. McDonell, and K. V. Kowdley.** 1998. Diagnosis of hemochromatosis. Ann. Intern. Med. **129(11):**925-931.
28. **Powell, L. W., E. C. Jazwinska, and J. W. Halliday.** 1994. Primary iron overload, p. 227-270. *In* J. H. Brock, J. W. Halliday, M. J. Pippard, and L. W. Powell (ed.), Iron metabolism in health and diseases. W. B. Saunders Co., Philadelphia.
29. **Powell, L. W. and K. J. Isselbacher.** 1998. Hemochromatosis, p. 2149-2152. *In* A. S. Fauci, E. Braunwald, K. J. Isselbacher, J. D. Wilson, J. B. Martin, D. L. Kasper, S. L. Hauser, and D. L. Longo (ed.), Principles of internal medicine, 14th ed. McGraw-Hill, New York.
30. **Sheppard, C. and E. Dierenfeld.** 2002. Iron storage disease in birds: speculation on etiology and implications for captive husbandry. J. Avian Med. Surg. **16(3):**192-197.
31. **Spelman, L. H., K. G. Osborn, and M. P Anderson.** 1989. Pathogenesis of hemosiderosis in lemurs: role of dietary iron, tannin, and ascorbic acid. Zoo Biol. **8:**239-251.
32. **Stramentinoli, G.** 1987. Pharmacologic aspects of S-adenosylmethionine. Am. J. Med. **83(suppl. 5A):**35-42.
33. **Tsai, S., K. Lee, and L. Tsai.** 2001. Effects of S-Adenosyl-L-methionine on liver damage in experimental obstructive jaundice. Kaohsiung J. Med. Sci. **17:**455-460.
34. **Wood, C.** 2002. Hemosiderosis and lemurs. Incidence, diagnosis, genetics and dietary prevention. Proc. XIX Congress Int. Primat. Soc. Beijing, China.**19: (1)**
35. **Worell, A. B.** 1991. Phlebotomy for the treatment of hemochromatosis in two Sulfur-breasted toucans. Proc. Annu. Conf. Assoc. Avian Vet. **(2):**9-14.s