Effect of *Ambrosia maritima* L. on Bovans-Type Chicks

Amel O. Bakhiet
S. E. I. Adam

ABSTRACT. Shoots of *Ambrosia maritima* L. fed to chickens at 2 and 10 percent of the basic diet for 6 weeks were not lethal. Average body weights and efficiency of feed utilization were markedly depressed in the chicks on feed with 10 percent *Ambrosia*, while growth of chicks on 2 percent *Ambrosia* was promoted as compared with chicks on a control diet. Organ lesions correlated with changes in clinical chemistry and hematology. Tissue recovery was incomplete 3 weeks following withdrawal from the test diets containing *Ambrosia*. *[Article copies available for a fee from The Haworth Document Delivery Service: 1-800-342-9678. E-mail address: getinfo@haworth.com]*

KEYWORDS. Chickens, medicinal plants, poisonous plants, toxicology

INTRODUCTION

Ambrosia maritima L. (family Asteraceae), locally known as Damesisa, grows abundantly in Central Sudan. The plant is claimed to have several medicinal properties and dry *Ambrosia maritima* shoots are used in traditional medicine for the treatment of gastrointestinal disturbances. Shoots of *Ambrosia maritima* are also used in some African countries as a molluscicide (10,6) since the sesquiterpene lactones and terpenoid compounds in

Amel O. Bakhiet and S. E. I. Adam are affiliated with the Department of Veterinary Medicine, Pharmacology and Toxicology, University of Khartoum, P. O. Box 32, Khartoum-North, Sudan.

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the plant have molluscicidal activity (1). The plant will produce pathogenic lesions in mice, sheep, goats, and calves (2,3,4,5).

The present study investigated the effect of *Ambrosia maritima* on growth, pathology, and alterations in hematology and serum constituents using 7 day old chickens; information on the pharmacological, chemical, and toxico-nutritional properties of the plant.

MATERIALS AND METHODS

Plant material. Ambrosia maritima L., purchased as dry plant material in Omdurman Central Market, Khartown, Sudan, and identified by Professor E.M. Abdel Bari, Department of Botany, Faculty of Science, University of Khartoum, Sudan, was used in this study. The plant tissue was ground to a fine powder with a mortar and pestle and then mixed in a commercial chick starter mash (Table 1).

Experimental. Bovans cockerels, 1 day old, were purchased from Coral Company, Khartoum. The chicks were placed in a poultry house (artificially illuminated at night and early morning with fluorescent lights for a 16 h day) at the Faculty of Veterinary Science, University of Khartoum and allowed free access to drinking water and feed (starter mash). At the age of 7 days, the chicks were randomly allotted into 3 groups of 12 chicks each. Group 1 chicks continued to be fed the starter mash and served as controls. Group 2 chicks were fed starter mash containing 2 percent of the ground *Ambrosia maritima* and the Group 3 chicks were fed mash containing 10 percent of the ground *Ambrosia maritima* as test diets. Feeding on the control and test diets continued for 6 weeks (treatment period) at which

TABLE 1. Composition of starter ration.

Ingredients	Quantity
	(% of total)
Sorghum	58
Soybean	4
Sesame cake	14
Groundnut cake	12
Wheat bran	5
Marble dust	1
Dicalcium phosphate	1
Superconcentrate	5
Total	100

time the test diets were replaced by the control diet for 3 weeks (recovery period).

Average body weights, weight gains, and feed conversion ratios (kg feed consumed/kg gain) were determined weekly for each group. From each group, 4 randomly selected chicks were slaughtered at 3, 6, and 9 weeks after beginning the test diets for pathological examinations. Blood samples were collected from each chick at slaughter for hematology and serum chemistry analysis. Necropsies were done on all chicks to identify gross lesions. The liver, proventriculus, intestines, spleen, heart, and kidneys were fixed in 10 percent neutral buffered formalin, embedded in paraffin wax, sectioned at 5 µm and stained with hematoxylin and eosin (H&E) to detect any cellular damage.

Chemical and hematological analyses. Serum samples were analyzed for aspartate transaminase (AST), lactic dehydrogenase (LDH), alkaline phosphatase (ALP), γ-glutamyl transferase (GGT), creatine kinase (CK), total protein, uric acid, cholesterol, total lipid, bilirubin, phosphorus, calcium, magnesium, iron and total iron binding capacity (TIBC) using commercial kits (Stanbio Laboratory Inc., San Antonio, Texas, Bio-Analytics, Palm City, Florida, or King Diagnostic Inc., Indianapolis, Indiana). Serum manganese and zinc and hepatic copper, zinc, and manganese were determined using an atomic absorption spectrophotometer (Shimadzh Model AA-670, Germany).

Erythrocyte (RBC) counts, packed cell volume (PCV), hemoglobin concentration (Hb), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV) were estimated by standard methods (12).

Statistical significance was assessed using Student's t-test (11).

RESULTS

Chicks fed a diet containing 10 percent *Ambrosia maritima* had lower body weights and average weight gains than chicks in the control group at 3 and 6 weeks after beginning the test diets (Table 2). Chicks fed 10 percent *Ambrosia* also used feed less frequently than chicks in the control group. The body weights and average weight gain of chicks fed a diet containing 2 percent *Ambrosia maritima*, however, were significantly higher than chicks in the control group at 3 and 6 weeks of feeding. At the end of the recovery period, *Ambrosia* was removed from diets, efficiency of feed utilization in chicks fed the 2 percent *Ambrosia maritima* was significantly less than chicks fed control diets or fed diets containing 10 percent *Ambrosia maritima*.

TABLE 2. Growth changes in chicks fed *Ambrosia maritima*.

Treatment	Feeding period[1]	Body weight	Weight gain	Feed conversion
	(weeks)[2]	(g)	(g/week)	(kg feed/kg gain)
Control	0-3	160 ± 1[3]	52 ± 0	2.4 ± 0.6
	4-6	342 ± 11	182 ± 10	2.0 ± 0.3
	7-9	635 ± 5	293 ± 1	3.5 ± 5.6
Ambrosia @ 2%	0-7	180 ± 2	79 ± 2	2.2 ± 0.2
	4-6	382 ± 0	202 ± 20	7.8 ± 1.1
	7-9	668 ± 4	285 ± 2	5.6 ± 1.6
Ambrosia @ 10%	0-7	154 ± 3	100 ± 6	7.0 ± 5.2
	4-6	308 ± 20	154 ± 37	12.6 ± 7.8
	7-9	600 ± 15	291 ± 4.8	3.7 ± 1.0

[1] All chicks were fed starter mash the first week; at an age of 1 week, groups of chicks were continued on starter mash (controls) or had starter mash mixed with *Ambrosia maritima* at indicated concentrations for 6 weeks; all chicks were returned to starter mash for the last 3 weeks.

[2] Time on test diet; measures were taken at the end of the indicated feeding period.

[3] Means ± S.E.

Pathological changes. At 3 weeks after beginning the test feeding, no significant changes were observed in the intestine or other tissues of the test chicks fed 2 percent *Ambrosia maritima* as compared with chicks on the control diet. Mild degenerative changes were observed, however, in the liver and kidneys of chicks fed diets containing 10 percent *Ambrosia maritima*. At 6 weeks after placing on the test diets, catarrhal enteritis and cytoplasmic fatty vacuolation of the hepatocytes in the inner parts of the liver lobule were more severe in chicks fed a test diet containing 10 percent *Ambrosia maritima* than in chicks fed a test diet containing 2 percent *Ambrosia maritima* or in the chicks on control diets. The kidneys of chicks on a diet containing 10 percent *Ambrosia maritima* had degeneration and/or necrosis of the epithelial cells of the proximal convoluted tubules, scattered lymphoid nodules in the congested cortex and shrinkage of the glomerular tufts (Figure 1). In addition, chicks fed 10 percent *Ambrosia maritima* had aggregates of lymphocytes between the cardiac muscle fibers. At the end of the recovery period, the organ damage was reduced, but neither the hepatocytes nor cells of the renal tubules had completely reverted to normal. No lesions were seen in the control birds.

Serum chemistry. Analysis showed significant differences in LDH, CK, GGT, total lipid, cholesterol, Mn, Fe and TIBC between chicks fed *Ambrosia maritima* and chicks on the control diet (Table 3). ALP activity was higher in chicks on 2 percent *Ambrosia maritima* as compared with chicks

FIGURE 1. Effect of *Ambrosia maritima* on kidney.

A chick was fed mash contalning ground shoot of *Ambrosia maritima* for 6 weeks; tubular cell degeneration is evident, 90×.

fed the control. Zn concentration was only increased in chicks on 2 percent *Ambrosia maritima* diet as compared with chicks fed the control diet. No differences in serum total protein, bilirubin, calcium, phosphorus, magnesium and AST were observed. At 3 weeks after withdrawal from the experimental diets cholesterol, Mn and Fe were lower in chicks on the test diet than in chicks in the control group. During the same period, LDH and GGT in chicks fed 10 percent *Ambrosia maritima* and uric acid and TIBC in chicks fed 2 percent *Ambrosia maritima* were lower than the chicks fed the control diet. CK and TIBC were higher in chicks fed *Ambrosia maritima* than the chicks fed the control diet. No significant differences between the chicks on the test diets and chicks on the control diet in serum ALP, total lipid and zinc during the recovery period were noted.

TABLE 3. Changes in serum constituents of chicks fed *Ambrosia maritima* L.

Treatment	Schedule[1]	LDH[2]	ALP	GGT	CK	Lipid	Cholesterol	Uric acid	Mn	Zn	Fe	TIBC
		(iu)	(iu)	(iu)	(iu)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)	(µg/dl)	(µg/dl)
Control	Feeding	65.2 ± 2.2	111.8 ± 0.7	41.8 ± 1.4	129.6 ± 4.1	5.0 ± 1.7	203.1 ± 4.1	5.2 ± 1.7	1.4 ± 0.0	0.35 ± 0.03	111.1 ± 4.3	134.9 ± 37.3
	Recovery	93.3 ± 4.2	114.4 ± 5.2	45.3 ± 1.3	135.8 ± 28.1	4.2 ± 0.9	218.0 ± 18.3	5.0 ± 1.7	1.6 ± 0.1	0.40 ± 0.0	234.2 ± 0.4	142.2 ± 0.4
Ambrosia @ 2%	Feeding	186.8 ± 4.2	112.9 ± 1.2	59.2 ± 2.1	168.8 ± 3.8	3.6 ± 0.2	176.6 ± 7.9	2.6 ± 0.4	1.1 ± 0.1	0.43 ± 0.0	232.5 ± 3.4	101.6 ± 6.1
	Recovery	94.9 ± 4.3	112.3 ± 0.0	44.2 ± 0.4	159.0 ± 7.7	5.0 ± 0.0	167.0 ± 36.4	2.6 ± 0.1	1.2 ± 0.0	0.41 ± 0.01	184.0 ± 1.0	94.2 ± 1.2
Ambrosia @ 10%	Feeding	152.2 ± 3.0	115.5 ± 0.7	74.7 ± 2.2	164.3 ± 6.2	4.1 ± 0.2	237.3 ± 38.4	5.2 ± 1.7	1.1 ± 0.1	0.33 ± 1.4	298.8 ± 2.3	149.9 ± 36.3
	Recovery	81.8 ± 5.3	112.9 ± 2.1	41.8 ± 0.5	140.7 ± 69.2	4.5 ± 0.6	108.3 ± 0.2	4.7 ± 1.5	1.2 ± 0.0	0.43 ± 0.0	234.2 ± 67.0	188.8 ± 1.2

[1] Feeding period lasted 6 weeks beginning with 1 week old chicks; recovery period was the 3 weeks following the feeding period; control chicks were maintained on starter ration throughout the experiment.

[2] LDH = lactic dehydrogenase, ALP = alkaline phosphatase, GGT = Gamma glutamyl transferase, CK = creatine kinase, and TIBC = total iron binding capacity.

Hematological changes. There were significant decreases ($P < 0.05$) in RBC and PCV or increases ($P < 0.02$) in MCH between the groups fed *Ambrosia maritima* and the controls (Table 4). No significant differences in the values of Hb, MCHC or MCV were observed during the 6 week feeding. At the end of the recovery period RBC values were still lower ($P < 0.02$) in the test birds than in the controls.

Hepatic zinc, copper and manganese levels. Hepatic zinc and manganese concentrations did not change during feeding of *Ambrosia maritima* L. or at the end of the recovery period. Hepatic copper concentration was significantly lower ($P < 0.05$) in the chicks fed 2 percent *Ambrosia maritima* during the feeding period and in the chicks fed 2 and 10 percent *Ambrosia maritima* during the recovery period ($P < 0.001$) as compared with the controls (Table 5).

DISCUSSION

Although *Ambrosia maritima* L. shoot is widely used in Sudan and other parts of the world for the treatment of various ailments, any toxicological information on chicks or other species of livestock is unavailable. In our studies, dry *Ambrosia maritima* L. shoots were not lethal to Bovans-type chicks when fed at 2 or 10 percent of diet for 6 weeks or during the 3 weeks following withdrawal from the experimental diets. Feeding *Ambrosia maritima* L. at 10 percent of the diet, however, had a profound effect on growth of the chicks. The pathological and biochemical data were indicative of the plant constituent(s) affecting the intestine, liver and kidneys and damage to these organs could explain the loss of body weight. The mechanism whereby the plant constituents injured body tissues cannot be stated from the present study, but the damage to these organs probably contributed to the raised serum LDH, CK and GGT activities and to the decreased serum total lipid and cholesterol concentrations. The concentrations of many serum constituents returned to normal by the end of a 3 week recovery period, but some of them such as CK and cholesterol were still abnormal, indicating incomplete repair of tissue damage by this time. The absence of hypoproteinemia suggests mild dehydration.

Interestingly, the mean body weight of chicks fed 2 percent *Ambrosia maritima* was significantly elevated. This increase in mean body weight may be due to the presence of relatively mild tissue damage and high protein content in *Ambrosia maritima* shoot, but neither growth promoter effects nor an exact proximate analysis of the plant tissue is available. The decrease in RBC levels and increase in MCH in chicks fed *Ambrosia maritima* suggest anemia. Similar results, except for increased MCV, occur with malabsorption of folic acid and vitamin B_{12} (8,9).

TABLE 4. Hematological characteristics of chicks fed *Ambrosia maritima* L.

Treatment	Schedule[1]	RBC[2]	PCV	Hb	MCHC	MCH	MCV
		(No./mm^3 × 10^6)[2]	(%)	(g/100ml)	(%)	(ng)	(mm^3)
Control	Feeding	2.75 ± 4.00	38.0 ± 2.8	9.8 ± 2.8	25.7 ± 1.5	35.5 ± 0.9	132.2 ± 11.1
	Recovery	2.58 ± 0.36	36.8 ± 3.3	9.5 ± 0.5	26.6 ± 1.8	37.3 ± 4.3	144.1 ± 2.2
Ambrosia @ 2%	Feeding	2.16 ± 17.90	30.0 ± 0.7	9.0 ± 0.6	30.2 ± 1.4	42.1 ± 3.0	139.0 ± 3.5
	Recovery	2.26 ± 17.9	31.2 ± 1.1	9.0 ± 0.6	28.7 ± 1.9	37.7 ± 3.5	138.4 ± 1.8
Ambrosia @ 10%	Feeding	2.29 ± 14.45	33.5 ± 2.5	8.9 ± 3.1	26.6 ± 1.2	40.0 ± 0.8	151.5 ± 1.9
	Recovery	2.43 ± 35.30	33.8 ± 1.1	9.0 ± 0.6	26.9 ± 1.9	38.0 ± 1.9	140.1 ± 5.1

[1] Feeding period lasted 6 weeks beginning with 1 week old chicks; recovery period was the 3 weeks following the feeding period; control chicks were maintained on starter ration throughout the experiment.

[2] RBC = number of erythrocytes, PCV = packed cell volume, Hb = hemoglobin, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, and MCV = mean corpuscular volume.

TABLE 5. Hepatic mineral level in chicks fed *Ambrosia maritima* L.

Treatment	Schedule[1]	Copper	Zinc	Manganese
		(mg/dl)	(mg/dl)	(mg/dl)
Control	Feeding	1.04 ± 0.04	0.20 ± 0.03	0.21 ± 0.09
	Recovery	1.2 ± 0.02	0.25 ± 0.01	0.25 ± 0.05
Ambrosia @ 2%	Feeding	0.60 ± 0.07	0.16 ± 0.0	0.32 ± 0.00
	Recovery	0.16 ± 0.00	0.25 ± 0.02	0.20 ± 0.00
Ambrosia @ 10%	Feeding	1.04 ± 0.01	0.19 ± 0.00	0.21 ± 0.01
	Recovery	0.11 ± 0.02	0.26 ± 0.01	0.20 ± 0.04

[1] Feeding period lasted 6 weeks beginning with 1 week old chicks; recovery period was the 3 weeks following the feeding period; control chicks were maintained on starter ration throughout the experiment.

Further studies are necessary to determine whether *Ambrosia maritima* shoot at the dietary doses used in this study will also exert an effect on the metabolism of the trace elements zinc, manganese, copper or iron in chicks and other animals. Zinc is known to competitively inhibit copper absorption and copper plays an integral role in hematopoiesis (7,13). Whether the interaction between zinc and copper plays an important role in anemia in the Bovanstype chicks fed *Ambrosia maritima* is unknown.

REFERENCES

1. Abdalla, O.M., A.A. Ali, and H. Itokawa. 1991. Cytotoxic activity of sesquiterpene lactones isolated from *Ambrosia maritima*. *Pharm.* 46:472.
2. Ahmed, O.M.M. and S.E.I. Adam. 1979. Toxicity of *Jatropha curcas* in sheep and goats. *Res. Vet. Sci.* 27:89-96.
3. Ali, B. and S.E.I. Adam. 1978. Effects of *Acanthospermum hispidum* on goats. *Journ. Comp. Pathol.* 88:533-544.
4. Barri, M.E.S. and S.E.I. Adam. 1980. The toxicity of *Crotalaria saltiana* to calves. *Journ. Comp. Pathol.* 91:621-627.
5. Barri, M.E.S., S.E.I. Adam, and O.H. Omer. 1988. Toxic effects of *Crotalaria saltiana* in mice. *Vet. Hum. Toxicol.* 30:429-431.
6. Belot, J., P. Bornarel, M. Sidhom, and M. Diouf. 1989. *Ambrosia maritima*. Etude des proprietes molluscicides de plusieurs generations d'origine Egyptienne et cultivees au Senegal. *Ann. Pharm. Fr.* 47:62-67.
7. Dunlap, W.M., W.G. James, and D.M. Hume. 1974. Anemia and neutropenia caused by copper deficiency. *Ann. Int. Med.* 80:470-476.
8. Eichner, E.R. 1973. Hematological disorders of alcoholism. *Am. Journ. Med.* 54:621-630.

9. Eichner, E.R. 1984. Macrocytic anemia. *In* J.L. Spivak, ed. Fundamentals of *Clinical Hematology.* Harper and Row, Philidelphia.

10. El Sawy, M.F., J. Duncan, S. Amir, H. El-Ruweini, and N. Brown. 1989. The molluscicidal properties of *Ambrosia maritima* L. (Compositae). Temporal and spatial distribution of Biomphalaria alexandrina in Egyptian village irrigation systems with reference to schistosomiasis transmission control. *Trop. Med. Parasitol.* 40:103-106.

11. Mendenhall, W.S. 1971. *Introduction to Probability and Statistics,* 3rd ed. Wadsworth Publishing Co. Inc., Belmont, California.

12. Schalm, O.W. 1965. *Veterinary Hematology.* Bailliere, Tindall and Cassell Ltd., London.

13. Underwood, E.J. 1977. *Trace Elements in Human and Animal Nutrition,* 4th ed. Academic Press, New York.

Dynamics of Tropane Alkaloids in *Duboisia myoporoides* Leaf During Development

Sanjay Mishra
Rajender Singh Sangwan

ABSTRACT. Physiological and metabolic status and tropane alkaloid content and composition during the time course of *Duboisia myoporoides* R. Br. leaf development were studied. The leaf life span of 110 days could be differentiated into distinct physiological phases of development: expansion, maturation and aging, and senescence and abscission. The expansion phase was accompanied by rapid accumulation of photosynthetic pigments and soluble proteins and translational activity. While this metabolic status was essentially maintained during maturation and aging, the senescence and abscission phase was characterized by accumulation of carotenoids, loss of dry matter, and a diminution of protein synthesis. Hyoscyamine and scopolamine (the major tropane alkaloids) accumulated only during the rapid leaf expansion. The senescence and abscission phase was accompanied by tremendous loss of these alkaloids. *[Article copies available for a fee from The Haworth Document Delivery Service: 1-800-342-9678. E-mail address: getinfo@haworth.com]*

Sanjay Mishra and Rajender Singh Sangwan are affiliated with the Division of Biochemistry and Molecular Biology, Central Institute of Medicinal and Aromatic Plants, P. O. CIMAP, Lucknow-226 015 (UP), India.

Address correspondence to Rajender Singh Sangwan.

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