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NEPHRITIS AND URIC ACID DIATHESIS IN CAPTIVE WILLOW PTARMIGAN (LAGOPUS L. LAGOPUS)

EFFECT OF FEED PROTEIN CONCENTRATION AND GRASS MEAL ADMIXTURE

By
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HANSSEN, INGOLF: *Nephritis and uric acid diathesis in captive willow ptarmigan (Lagopus l. lagopus). — Effect of feed protein concentration and grass meal admixture.* Acta vet. scand. 1982, 23, 446—455. — Mortality due to nephritis and uric acid diathesis was observed during the fall and winter in captive willow ptarmigan. The present study examined how feed protein concentration influences feed and water consumption, plasma uric acid concentration, and mortality due to nephritis and uric acid diathesis in captive willow ptarmigan. An increase in feed protein concentration from 14 to 24 % resulted in reduced feed consumption and increased plasma uric acid concentration. Mortality due to nephritis and uric acid diathesis was not influenced by variations in feed protein concentration, but an admixture of 14 % grass meal to the diet reduced the incidence markedly.

willow ptarmigan; nephritis; feed protein; grass meal.

Avian nephritis and uric acid diathesis have been observed in a variety of different species. Most commonly the disease has been observed in domestic fowl, turkey, goose, duck, psittacine cage birds and birds of prey (*Gratzl & Köhler* 1968, *Petrak* 1969), but wild tetraonids have shown the disease as well (*Hülphers & Lilleengen* 1948). The disease is usually classified as a metabolic disorder, characterized by uric acid and urates being deposited in various parts of the organism. The etiology of this alignment is obscure, but close confinement and exposure to cold and dampness, excessive protein consumption, vitamin A and B_{12} deficiency, intoxications and certain infectious diseases have been mentioned as predisposing factors (*Patterson* 1928, *Svensson* 1937, *Siller* 1959).

Since 1972 willow ptarmigan (Lagopus l. lagopus) have been reared for experimental use at the University of Tromsø, Norway. In the autumns mortality caused by nephritis and uric acid diathesis was repeatedly observed, mainly among juvenile birds.

Ptarmigan are obviously adapted to live on low protein diet during the winter (*Moss & Hanssen* 1980). This might implicate that they do not tolerate heavy protein loads at this time of the year, but die due to accumulation and precipitation of protein catabolism end product, that is uric acid, in the viscera. Support for this theory is the positive correlations between feed protein concentration, plasma uric acid concentration and mortality due to nephritis and visceral gout in chicken (*Svensson* 1937, *Siller* 1959). The present study was conducted to examine how different feed protein concentrations influence food and water consumption, plasma uric acid concentration, and mortality due to nephritis and uric acid diathesis in captive willow ptarmigan. The effect of grass meal admixture on mortality and plasma uric acid has been tested as well.

MATERIAL AND METHODS

Ptarmigan

The birds were derived from eggs laid by wild or captive ptarmigan hens. All eggs were incubated and hatched in machines, and the chicks were reared on a concentrated feed mixture, supplemented with blueberry plants (Vaccinium myrtillus) (*Moss & Hanssen* 1980). From August on the chicks were kept in cages, singly or in pairs. The cages were cubes of ca. 90 cm^3 with wire floors (3/8 × 1″). Some birds were kept indoors and some outdoors. The indoor birds were kept on a light regime synchronous to the outdoor photoperiod. From the beginning of September the chicks were gradually adapted to the experimental feeds over a two-week period. Both feed and water were provided ad libitum.

Extensive trials

During a 5-year period the willow ptarmigan stock was fed different feed during the September-to-April period (Table 1). These mixtures were supplemented with one of their natural

Table 1. Composition and analysis (dry basis) of diets.

	Diets 1972	1973—1975	1976
Ingredients (%)			
Herring meal	8	2	2
Soya meal (extracted)	8	2	—
Maize	25	25	15
Barley	4	5	10
Oats	10	10	12
Wheat	5	5	10
Oat husks	18	30	15
Wheat bran	12.6	12.6	12.1
Brewers yeast	2	1	1
Soya oil	—	—	2
Grass meal	2	2	14
Kelp meal	2	2	2
Limestone	1.5	1.5	1.5
Dicalcium phosphate	1.5	1.5	2.0
Trace mineral premix	0.2	0.2	0.4
Vitamin premix	0.2	0.2	1.0
Analysis (%)			
Crude protein	18.1	13.1	12.6
Oil	4.4	4.0	6.5
Crude fibre	6.8	9.5	11.7
NaCl	0.5	0.4	0.4
Ca	1.2	1.2	1.5
P	0.9	0.8	0.8
Mg	0.2	0.2	0.2
K	0.8	0.6	1.0
Added vitamins (per kg)			
Vit A i.u.	20,000	20,000	7,500
Vit D_3 i.u.	2,800	2,800	1,480
Vit E mg	40	40	250
Vit B_1 mg	—	—	25
Vit B_2 mg	10	10	150
Vit B_6 mg	2	2	45
Nicotinic acid mg	40	40	550
Ca-D-pantothenate mg	12	12	55
Choline chloride mg	—	—	3,525
Folic acid mg	1	1	10
Vit K_3 mg	—	—	10
Vit B_{12} mg	—	—	0.01
Biotin mg	—	—	0.45
Inositol mg	—	—	550
Para-amino-benzoic acid mg	—	—	25
Added trace minerals (mg per kg)			
Fe	110	110	172
Mn	140	140	228
Zn	108	108	200
Cu	32	32	57
Co	2	2	4
I	3	3	8

main winter feed items, willow (Salix spp.), and fresh twigs were offered once or twice a week. All ptarmigan that died during these years were necropsied, and the occurrence of nephritis and uric acid diathesis were recorded.

Specimens from liver and kidney tissues and contents from the small intestine were incubated on 5 % human blood agar and bromthymolblue lactose agar plates (*Nordic Committee on Food Analysis* 1969). The plates were incubated aerobically at 37°C and read after 24 and 48 h. Quantitative bacteriological investigations of small intestine and caecum contents were performed by diluting 1.0 g samples to 10^{-2}, 10^{-4}, 10^{-6} and 10^{-8} by 0.9 % saline. Aliquots of 0.1 ml were transferred onto the surface of blood agar plates and incubated for 24 h aerobically and 5 days anaerobically at 37°C before being read. The bacteria were identified according to Bergey's Manual of Determinative Bacteriology (*Buchanan & Gibbons* 1974).

Intensive trials

Effect of feed protein concentration and length of day on feed and water consumption, plasma uric acid concentration and uric acid excretion. Fifteen birds, of both sexes, were kept indoors and divided into 3 groups. One group (control) got a low protein diet (14 % crude protein) and light adjusted to the outdoor photoperiod (Fig. 1). The second group got a high protein diet (24 % crude protein) and outdoor daylength regime (high protein/normal day group), and the third group got the high protein diet and 18 h daylight (high protein/long day group). The diets did no contain grass meal and the protein concentrations were regulated by varying admixture of herring and soya meal. Added trace minerals and vitamins were as for the 1976 diet (Table 1). The experiment lasted from the beginning of October to the middle of June for the two first mentioned groups, and from October to the middle of January for the last group. Once every 2 weeks feed and water consumption were recorded over a 48 h period. Once in the intermediate weeks the birds were weighed and blood samples (1½ ml) taken from a wing vein. The plasma was stored at —20°C till uric acid was assessed spectrophotometrically, using a method described by *Kageyama* (1971). Every time blood samples were taken, the percentage of pigmented plumage on the head and back was subjectively assessed. Collection of 24 h feces for urate analyses was done at irregular intervals. The feces was dried over night at 105°C, weighed and homogenized in a warring blender. The urate of 1.0 g feces was extracted with 250 ml of a 0.5 % lithium carbonate solution according to *Tinsley & Nowakowski* (1956), and urate in the filtered extract was measured using the same method as for plasma.

Effect of grass meal on plasma uric acid concentration. Fourteen birds were divided into 2 groups. One group received a diet containing 24 % crude protein (control), while the second group was fed a 24 % crude protein diet containing 20 % grass meal (grass meal group). Blood samples were taken every 10 days from a wing vein, and plasma was analysed for uric acid. Students t-test was used to compare mean values during the experiment. Significance was set at the 5 % level.

RESULTS

Extensive trials

Mortality due to nephritis and uric acid diathesis is presented in Table 2. The table shows that all deaths with exception of those taking place in 1975, occurred during the autumn and early winter months, and that mortality was low when ptarmigan were fed a low protein, grass meal containing diet (1976 and later).

Post mortem examinations. Some birds were in good condition, while most of them were dehydrated and showed breast muscle atrophy of varying degree. Catarrhal inflammation

Table 2. Mortality in juvenile willow ptarmigan caused by nephritis and urid acid diathesis during a 5 year period.

Year	Diet	Number of juveniles 1st Sept.	Monthly mortality												Mortality %
			S	O	N	D	J	F	M	A	M	J	J	A	
1972	18 % crude protein	114	2	6	1	2									9.6
1973	13 % crude protein	205	5	22	1										13.6
1974	13 % crude protein	250		7	18	21									7.6
1975	13 % crude protein	360		7	13	11			1	3		1	1		10.2
1976	13 % crude protein with 14 % grass meal	415					3								0.7

in the small intestine was regularly observed, and dilatation of the caeca was occasionally seen. The kidneys were swollen and showed urate precipitations. Ureteres were dilated and packed with urates. Urate precipitations were occasionally seen on the serous membranes of the heart, liver and small intestine. Histological examinations of the kidneys never showed more than a slight cellular inflammatory reaction. Pathogens could not be found in the gut, and bacteria were never isolated from other organs. However, quantitative bacteriological examinations of the gut contents from 3 birds showed high numbers of Clostridium perfringens type A (10^5—10^8/g contents) both in the small intestine and the caeca.

Intensive trials

Both the control and the high protein/normal day groups showed only small variations in body weight during the experiment. All birds increased their weight with about 10 % from the beginning to the end of the experiment, and an additional 10 % increase was recorded for both sexes prior to egg laying.

The feed consumption in the same 2 groups is shown in Fig. 1. During the first 3 weeks of the experiment both groups ate the same amount of feed. After the birds had moulted, and during the entire 5 h light period, feed consumption was significantly lower in the high protein group than in the control group. At the time when the length of the day started to increase, feed consumption increased over a 2 weeks period in both groups, but abruptly fell back to the previous level. Then a gradual increase, which was steepest for the high protein group, occurred towards the egg laying period.

From the feed consumption curve, nitrogen consumption for both groups has been determined (Fig. 1). While the nitrogen consumption curve of the control group was fairly flat during the entire experiment, the nitrogen consumption curve of the high protein group decreased during the autumn moult, and showed peaks, the first in connection with increasing day length, and the second prior to egg laying. During the whole experiment, nitrogen consumption was significantly higher in the high protein group than in the control group.

The water consumption was closely related to food consumption in both groups.

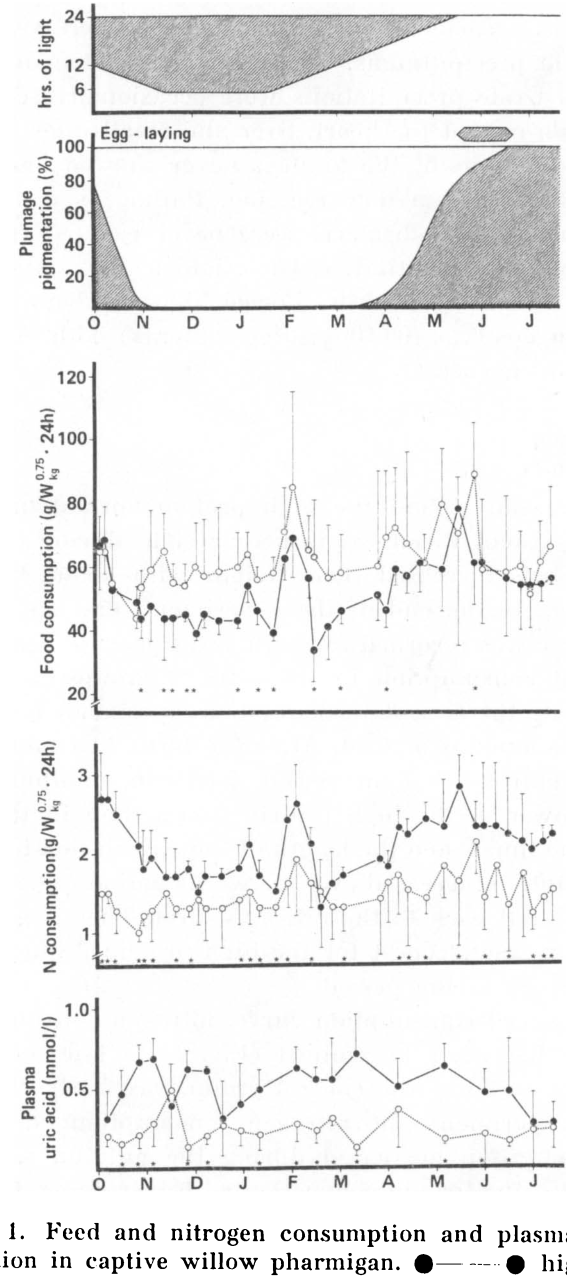

Figure 1. Feed and nitrogen consumption and plasma uric acid concentration in captive willow pharmigan. ●——● high protein/normal day group (n = 3 ♂, 1 ♀), and ○——○ low protein/normal day group (n = 3 ♂, 2 ♀). Vertical bars indicate standard deviations. The seasonal changes in normal daylength and plumage are indicated. An asterisk indicates significantly different means ($P < 0.05$).

The plasma uric acid consumption was higher in the high protein group compared to the control (Fig. 1).

The 24 h excretion of uric acid was similar in the 2 groups except in late November and during egg laying when the high protein group excreted significantly more (1200—1300 mg) uric acid than the controls (650—750 mg). The amount of uric acid excreted per g N consumed was fairly constant during the whole period, though a slight tendency toward less excretion was observed for both groups at the beginning of June. There was, however, no significant difference between the groups.

The high protein/18 h daylength group showed continuing moult during the whole experimental period. Neither food consumption nor plasma uric acid concentration was significantly different from the values recorded for the high protein/normal daylength group.

Admixture of 20 % grass meal to the diet did not make any significant reduction in plasma uric acid concentration.

DISCUSSION

The extensive trials showed that the mortality due to nephritis and uric acid diathesis occurred in the fall and early winter months, which is in accordance with data for chicken (*Svensson* 1937, *Siller* 1959) and free-ranging tetraonides (*Hülphers & Lilleengen* 1948).

In 1976 the ptarmigan received a diet containing 14 % grass meal, and both in this and subsequent years, when the same diet was used, mortality due to nephritis and visceral gout was low. Without pointing to any factors or mechanisms *Patterson* (1928) stated that green feed prevents nephritis in domestic fowl. Green feed is considered to be an important source of carotenes, carotenoids and consequently vitamin A, and deficiency of the latter substance is known to be associated with increase in plasma uric acid of chicken, probably by a rise in liver xanthine dehydrogenase and kidney arginase activity (*Bruckental & Ascarelli* 1975). In the present series of experiments, however, the admixture of vitamin A was considerably higher in those years when nephritis occurred than in recent years when the incidence was low. The admixture of the vitamin B complex was, however, more complete since 1976 than in earlier years.

The intensive trials showed that enhancing the dietary crude protein concentrations from 14 to 24 % resulted in a slight but significant increase in plasma uric acid concentration in willow ptarmigan. It was also shown that food and water consumption were slightly influenced by daylength, moult and reproduction, while plasma uric acid concentration showed no tendency toward seasonal variations.

The birds fed a high protein diet remained healthy during the whole experiment and the plasma uric acid concentrations of these birds were constantly low compared with values obtained from chicken with nephritis and uric acid diathesis (*Siller* 1959). It is further evident from the extensive trials that a reduction in feed protein concentration to 13 % did not reduce the mortality (Table 2). This indicates that the disease mechanism in ptarmigan nephritis and uric acid diathesis is more complicated than a simple feed protein overloading.

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SAMMENDRAG

Nyrebetennelse og urinsur diathese hos liryper i fangenskap. Effekt av grasmel og varierende proteinkonsentrasjon i fóret.

Nyrebetennelse og urinsur diathese forekom om høsten og vinteren hos liryper i fangenskap. I dette arbeidet er det testet hvordan forskjellige proteinkonsentrasjoner i fóret influerer på fór- og vannopptak, urinsyrekonsentrasjon i plasma og mortalitet forårsaket av nyrebetennelse og urinsur diathese hos liryper i fangenskap. En økning i proteinkonsentrasjon fra 14—24 prosent resulterte i nedsatt fóropptak og økt urinsyrekonsentrasjon i plasma, mens mortaliteten pga nyrebetennelse og urinsur diathese var upåvirket. En tilblanding av 14 prosent grasmel til fóret reduserte forekomsten av nyrebetennelse og urinsur diathese betraktelig. Årsaken til dette er foreløpig uklar.

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