Alcohol accumulation from ingested berries and alcohol metabolism in passerine birds

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Experiments were conducted to determine whether under natural conditions fermentation takes place in various berries (rowan berries *Sorbus aucuparia,* hawthorn haws *Crataegus monogyna,* rose hips *Rosa rugosa)* and whether harmful concentrations of alcohol from the berries accumulate in Waxwings *Bombycilla garrulus* and Bullfinches *Pyrrhula pyrrhula.* In addition, the alcohol metabolism of Waxwings, Starlings *Sturnus vulgaris* and Greenfinches *Chloris chloris* was examined.

The ethanol concentrations in rowan berries and rose hips increased significantly as a result of fermentation. The highest concentrations were 2.4 g/kg for rowan berries in February and 3.2 g/kg for rose hips in January. The levels of alcohol in Waxwings and Bullfinches consuming rowan berries in protected environments were higher in the winter than in the autumn, but the levels were never high enough to influence behaviour.

Waxwings, which are adapted to eating berries, metabolized a test dose of 1 g/kg ethyl alcohol at a rapid rate, 900 mg/kg/h; Starlings, which consume a mixed diet, had a slower rate of alcohol metabolism, 270 mg/kg/h; and Greenfinches, which have a seed diet, had a very slow rate, only 130 mg/kg/h. The alcohol dehydrogenase (ADH) enzymes differed in these species, both in activity and in isoenzyme composition. The ADH activities of Starlings and Greenfinches are so low (0.75 and 1.68 mμmol/min/mg pr.) that they probably restrict alcohol metabolism. These species of passerines eliminated a larger dose (2 g/kg) of ethanol more rapidly than a smaller dose (1 g/kg). A comparison of the rates of alcohol metabolism in Waxwings, Starlings, Bullfinches, and Greenfinches suggests that the rate varies as a function of the quantity of ethanol that the species happens to ingest with its food.

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Introduction

Ethyl alcohol is widely found in plants in nature (Forsander 1978). The concentrations are generally very low so that wild animals usually do not have to deal with significant amounts. Accumulation of alcohol may nevertheless take place when fruits and berries spoil, especially in warm regions where there are large fruits with a high sugar content. African elephants *Loxodonta africana* (Carrington 1958), and wart hogs *Phacochoerus aethiopicus* (Morehead 1962) are known to have become intoxicated after eating spoiled fruit of the tree *Sclerocarya schweinfurthii.* Similarly, cows have been observed to become intoxicated after having eaten spoiled apples (Frost 1923). Bullfinches *Pyrrhula pyrrhula* and Hawfinches *Coccothraustes coccothraustes* are reported to have become intoxicated after eating willow blossoms (Lack, personal communication; Krebs & Perkins 1970). In Finland as well, reason to suspect intoxication has been given by unpublished observations of birds flying against cars and house windows, especially Waxwings *Bombycilla garrulus* and Pine Grosbeaks *Pinicola enucleator,* which are found here in large numbers in the winter. In March 1977 Pine Grosbeaks killed in such accidents were found to contain small amounts of alcohol (P. Eriksson, unpubl. obs.).

Although the alcohol metabolism of mammals has been studied extensively, there is very little information about the metabolism of alcohol in birds. The rates of alcohol metabolism have been measured only in chickens *Gallus gallus* and Feral Pigeons *Columbia livia* (LeBreton et al. 1935, LeBreton 1936). The ADH activity and isoenzyme composition have been studied only in chicks and Quail *Coturnix coturnix* (Castro Sierra & Ohno 1968, Moser et al. 1968).

The present research had three goals: (1) to determine the extent to which the fruits of rowan trees (European mountain ash) *Sorbus aucuparia,* hawthorns *Crataegus monogyna,* and roses *Rosa rugosa* ferment to produce alcohol in nature; (2) to determine if intoxicating concentrations of alcohol accumulate in Waxwings and Bullfinches consuming rowan berries; and (3) to see whether Waxwings, adapted to a berry diet, Starlings *Sturnus vulgaris,* using a mixed diet, and Greenfinches

Chloris chloris, with a seed diet, differ from each other in their alcohol metabolism.

Diet of the birds studied

Four species of birds were chosen for examination on the basis of their varying food habits.

Waxwings are primarily fruit eaters, although in the summer both adult Waxwings and nestlings consume insects (Siivonen 1941). After berries have ripened in the autumn, their diet includes hawthorn haws, juniper berries, rose hips, apples, and the berries of several ornamental bushes (Cornwallis 1961, Kolunen & Vikberg 1978), but rowan berries are the main constituent. Berries that persist over the winter, such as crowberries and bearberries, are important for the returning migratory birds because there are still few insects available (see v. Haartman et al. 1963—72).

Waxwings have been found to utilize only the pulp of rowan berries (Pulliainen 1978). According to observations of birds both in nature and in cages, the excrement contains only the seeds and peels of the rowan berries. The seeds travel undigested through the bird's alimentary canal. The outer coating of the seeds found in Waxwing excrement presents nearly the same appearance in scanning electron microscope pictures as those merely separated from the berries (Pulliainen & Erkinaro 1978).

Bullfinches are typical seed and leaf bud eaters. Their diet consists of the seeds of various grasses and the leaves and buds of trees. Bullfinches also eat the berries of rowan, juniper, sea buckthorn, and elder, because of their seeds (Erkamo 1948; Newton 1967). Typically, Bullfinches peck the seeds out of the berries, but some of the pulp is nevertheless ingested along with the seeds.

Starlings have a mixed diet. Over the course of the year they are estimated to eat as much plant as animal material (Havlin & Folk 1965). The intake of animal material is predominant during the nesting season, but berries and fruit are the most important constituents in the autumn. In Finland Starlings seem to prefer rowan berries and elderberries, from which they utilize primarily the pulp (Pulliainen 1978). Grain and seeds enter their diet later in the autumn and in the winter (see v. Haartman et al. 1963—72).

Greenfinches are true seed eaters. Animal material constitutes only 0.5 % of the mature bird's diet (Korpinen 1969). Greenfinches eat the seeds from several wild grasses, trees, and bushes, as well as grain.

Material and methods

Berries. Rowan berries, hawthorn haws, and rose hips were picked for measurements of alcohol and sugar contents approximately once a month between 9 September 1979 and 12 February 1980 in the Helsinki area. The samples were always obtained from the same trees and bushes in order to get reliable information about the changes in alcohol and sugar content in the fruit during the period of the experiment. Measurements were made immediately after picking because enzyme activity metabolizing sugar (Salo 1967) and ethanol continues even after the fruits have been picked.

The ethanol concentrations of the fruits were estimated by gas chromatography (Eriksson 1973). Glucose and fructose concentrations were measured with UV methods using Boehringer kit no. 139106; sucrose levels were measured with kit no. 139041.

The rowan berries fed to the Waxwings and Bullfinches at the end of October and beginning of November were picked a few days before the experiment. For the January experiment, however, berries picked earlier in September had to be used because of an extremely poor rowan berry crop. They were frozen and then later defrosted and kept at 11°C for 10 days in order to allow fermentation before being given to the birds. The alcohol concentrations of the berries fed to the birds in the autumn and winter were estimated from five similarly treated samples.

Birds used in the experiment. The Starlings (N=25; mean weight ♂ 72.4 g, ♀ 62.1 g) were captured at the end of August, the Waxwings (N=32; ♂ 60.6 g, ♀ 59.6 g) and Bullfinches (N=35; ♂ 32.1 g, ♀ 25.7 g) in October, and the Greenfinches (N=32; ♂ 28.0 g, ♀ 26.6 g) in November. The birds were housed at Korkeasaari Zoo. No sex differences were found in the alcohol metabolism of the species studied; therefore, the results for the males and females in the berry feeding and alcohol metabolism experiments have been combined.

Berry feeding experiment. The studies were conducted at two times: (1) at the end of October — beginning of November and (2) in January. The reason for this timing was to determine whether the blood alcohol levels of the birds were elevated by the expected higher ethanol concentration of the berries and by the birds having greater caloric needs as a result of the lower temperatures and shorter daylight in winter.

The birds were transferred to outside cages and switched to a diet of only rowan berries 2—3 days prior to the taking of blood samples. The berries were made available throughout the daylight hours. Blood samples vere obtained from approximately ten individuals of one species at dawn and from another group of the same species at sunset usually of the same day but sometimes of the following day. A few days later samples were again taken, this time at noon and at midnight. The samples were taken from the wing artery by making a small incision in the skin with a scalpel and inserting a thin needle into the artery. Blood was collected into 50 µl capillary pipettes, and the ethanol concentration was measured with the methods of Eriksson (1973). These pipettes were not heparinized because commercial heparin contains trace amounts of alcohol (Krebs & Perkins 1970), which could interfere with the measurement of extremely low alcohol concentrations.

The average daily caloric intake in the daylight hours of Waxwings in outside cages was measured during 6 to 9 November and on 16, 17 and 19 January. The mean temperature of the three days in November varied from +3° to +5° and in January from —6° to —10°C. The daily rowan consumption of Bullfinches could not be measured because the birds' eating habits made it impossible to collect all of the remaining berries.

Alcohol metabolism measurement. The alcohol metabolism of Waxwings, Starlings and Greenfinches

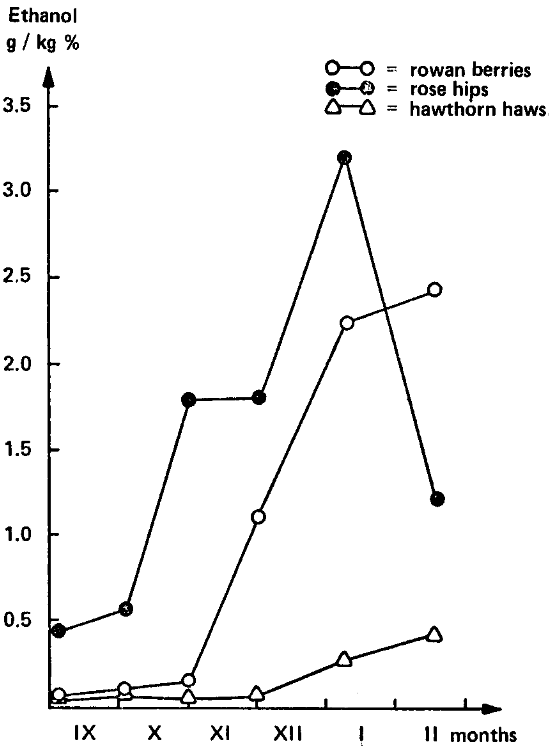

Fig. 1. Alcohol concentrations in rowan berries, rose hips, and hawthorn haws, as g/kg of the fresh weight, from September to February.

was measured at two ethanol doses: ten birds of each species were given 1 g/kg ip ethanol and another group of ten were given 2 g/kg. Alcohol was administered as a 10 % (w/v) solution in normal saline, warmed to 40°C before injection.

The blood samples from the Waxwings receiving the smaller ethanol dose were collected into 50 µl heparinized capillary pipettes at 20, 30, 40, 50, 60, and 70 min. after injection; samples from Waxwings receiving the larger dose were taken at 30, 45, 60, 80, 100, and 120 min. The corresponding samples (50 µl) from Starlings were taken at 30, 60, 90, 120, 150, and 180 min. and at 60, 90, 120, 150, 180, and 210 min. All the samples (25 µl) from Greenfinches were taken at 60, 120, 180, 240, and 300 min. All times were selected on the basis of pilot studies.

Measurements of ADH activities and liver isoenzymes. The livers from the birds that had previously been given 1 g/kg ethanol were used for these analyses. ADH activity was measured with a spectrophotometer (Beckman D8-G) at a wavelength of 340 nm at 40°C with a pH of 7.4, according to the procedure of Bergmeyer (1970). The protein concentration was measured from liver homogenates with the method of Lowry et al. (1951).

The ADH isoenzymes were analysed by horizontal starch gel electrophoresis. A 14 % starch gel with the buffer system described by Shaw and Prasad (1970) was used. Electrophoretic drives were made at 4 ± 2°C with a gradient of 4 V/m. Such a low gradient was used because according to Castro-Sierra & Ohno (1968) the ADH isoenzymes of birds are not visible in the gel following the use of higher gradients. The driving time was 16 h. The gels were dyed according to the method of Poulik (1957).

Results

Alcohol and sugar concentrations in the berries. Alcohol first began accumulating in the rowan berries in November and in rose hips about a month earlier (Fig. 1). The highest alcohol concentration in rowan berries, 2.4 g/kg fresh wt., was found in February; in rose hips, 3.2 g/kg in January. The alcohol levels of hawthorn haws remained low throughout the entire experiment: the highest value, 0.4 g/kg, was found in February.

The glucose concentration of rowan berries in September was 7 % of the dry weight. Because of night frosts it rose to 12 % in the end of October, but by February it had been reduced to only 8 %. The fructose concentration was only 3 % in September, but rose to 8 % in February. In rose hips the sugar content rose slightly from September to October, at which time the concentration of glucose was 11 % and fructose 13 %. By February these levels had fallen to 4 % and 6 %. The sugar content of hawthorn haws was always low, varying between 1.5 % and 5 %. The sucrose levels of all berries were low (<1 %) during the experimental period.

The mean alcohol concentration of the rowan berries fed to the birds in the autumn experiment was 0.69±0.20 g/kg. In the winter experiment, the average concentration in the berries that were artificially fermented was 1.69±0.65 g/kg, which is somewhat lower than the concentration in berries picked at that time (2.2 g/kg).

Blood alcohol levels in Waxwings and Bullfinches on a berry diet. Little or no alcohol was found in either the dawn or midnight blood samples in the autumn experiment and in the dawn samples in the winter experiment (Table 1). At the noon and sunset measurements, however, all the birds had alcohol in their blood. The alcohol concentrations both at noon and at sunset were clearly higher in the winter than in the autumn for the Waxwings, but for the Bullfinches the difference between the autumn and winter values was significant ($P<0.001$) only with the noon measurements.

The mean temperature during the January experiment was approximately 14°C colder than during the October-November experiment (Table 1). The days were about 3 hours shorter during the winter experiment. The mean intake of rowan berries per Waxwing was 66.4±10.5 g/day in November and 84.9±8.3 g/day in January; this difference, however, fails to reach significance ($P<0.10$).

The rates of alcohol metabolism in Waxwings, Starlings, and Greenfinches. Fig. 2 shows the ethanol elimination of the three species of birds, and Table 2 presents the mean rates of alcohol

Table 1. Mean (±SD) blood ethanol concentration as mM in Waxwings and Bullfinches at different times of the day at the end of October — beginning of November and in January (1979—1980). N = number of birds, t = mean daily temperature as °C.

Oct.-Nov. time	Waxwing mM	Bullfinch mM	Jan. time	Waxwing mM	Bullfinch mM
6.35—7.35	0.004±0.006 n=10 t= +2°	—0.007±0.017 n=10 t= +1°	8.20—9.20	0.006±0.006 n=10 t= —6°	0.005±0.006 n=9 t= —9°
11.30—12.30	0.057±0.026 n=10 t= +4°	0.083±0.041 n=10 t= +5°	11.30—12.30	0.148±0.066 n=9 t= —19°	0.453±0.251 n=9 t= —11°
16.40—17.40	0.105±0.048 n=10 t= +2°	0.121±0.084 n=9 t= +1°	15.40—16.40	0.280±0.175 n=10 t= —11°	0.103±0.046 n=10 t= —9°
23.30—00.30	0.006±0.007 n=10 t= +4°	0.008±0.008 n=10 t= +4°	23.30—00.30	0.069±0.083 n=9 t= —11°	0.055±0.039 n=8 t= —11°

metabolism. The Waxwings metabolized alcohol (with the 1 g/kg dose) 3.3 times as fast as the Starlings and 7.2 times as fast as the Greenfinches, while the Starlings metabolized it 2.2 times as fast as the Greenfinches. The rate of alcohol metabolism was approximately 40 mg/kg/h faster after the larger dose than after the smaller dose in all three species. The difference was significant in the Starlings ($P<0.001$) and the Greenfinches ($P<0.01$) but not with the Waxwings because of their larger variation.

ADH activities and isoenzymes. The ADH activity of Waxwing liver, measured as mμmol/min/mg protein was on the average 15 times as high as that of Starlings and 6.2 times as high as that of Greenfinches (Table 3). In Waxwings the liver constitutes 4.9 % of the total body weight; in both Starlings and Greenfinches only 2.8 %. The whole liver ADH activity of Waxwings is 23 times as high as that of Starlings and Greenfinches.

The ADH of Waxwings had two distinctly separate isoenzymes, both of which moved toward the cathode. Greenfinches had a single isoenzyme, which moved toward the cathode. Starlings had two isoenzymes, both appearing only faintly in the gel, one of which migrated toward the cathode and the other of which moved toward the anode. No variations in ADH isoenzymes were found between individuals within a species.

Discussion

The alcohol concentrations of rowan berries and rose hips clearly increased during the autumn and winter. These results are similar to the finding that strawberries and plums contain more alcohol when they are overly ripe (Forsander 1978). From the decreasing glucose concentrations found in the present study, it can be assumed that at least part of the glucose was fermented to ethanol. Fructose fermentation also appears to have taken place in rose hips because their fructose concentrations decreased similarly to their glucose concentrations.

Ethanol formed as a result of carbohydrate fermentation has previously been found in the crops of chickens (Bolton 1962, 1965) and in the caeca of Willow Ptarmigans *Lagopus lagopus* (McBee & West 1969). In the latter, the ethanol concentration reached 60 mg/100 g of the caecal contents.

Apparently no endogenous alcohol is formed in the crops or intestines of Waxwings and Bullfinches, since the blood ethanol levels were essentially zero at several measurement times, e.g. at the midnight measurement in the autumn experiment. The low alcohol levels observed at midnight in the winter experiment are presumably caused by the continued presence of ethanol in the food that has been stored in the crop and released into the intestines during the evening.

The energy requirements of birds increase prog-

Table 2. Mean (±SD) ethanol metabolic rates of three species of passerines after doses of 1 and 2 g/kg ethanol. (N= 10 for each measurement.)

Alcohol dose	Waxwing mg/kg/h	Starling mg/kg/h	Greenfinch mg/kg/h
1 g/kg	897.7±131.4	270.8±18.6	125.5±17.1
2 g/kg	939.3±150.3	310.6±22.9	169.4±35.0

Table 3. Mean (±SD) ADH activity in the livers of Waxwings, Starlings, and Greenfinches. (N = 10 for each measurement.)

	Waxwing	Starling	Greenfinch
ADH mμmol/ min/mg pr.	11.26±1.88	0.75±0.12	1.68±0.43
ADH μml/min/ whole liver	6.49±1.32	0.27±0.06	0.28±0.08

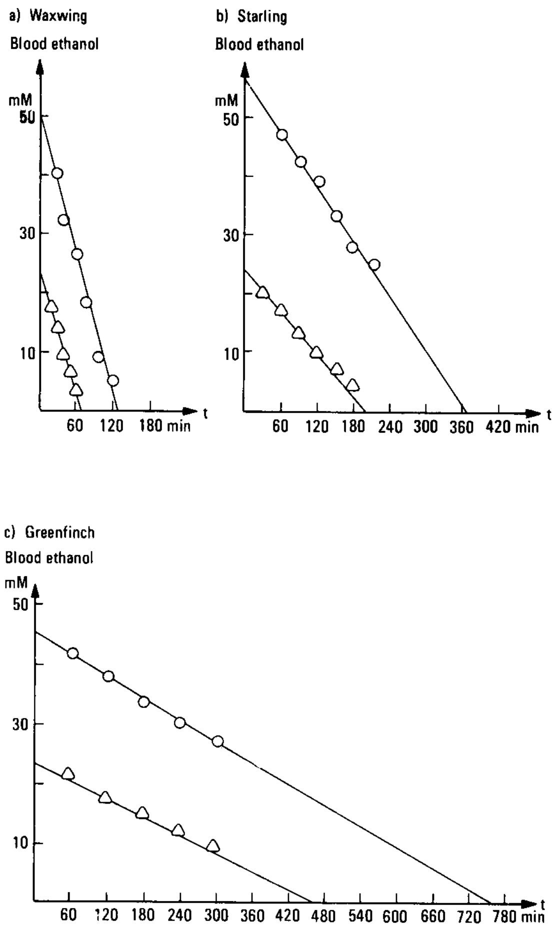

Fig. 2. Alcohol elimination after administration of 1 g/kg ethanol (△) and 2 g/kg (○) in Waxwings (*a*), Starlings (*b*), and Greenfinches (*c*). Each point is the mean value for ten birds.

ressively with decreases in the environmental temperature below the thermoneutral zone (Kendeigh et al. 1977), and cold nights cause birds to increase their daily food intake (Kendeigh et al. 1969). The Waxwings in the present study tended to eat more in January but the difference was not statistically significant ($P<0.10$). The relatively small difference was probably due to the fact that the mean temperature during the January experiment was only 14° colder than during the autumn experiment.

Although measurable amounts of alcohol were found in the blood of the Waxwings and Bullfinches, the levels are probably not high enough to be of any behavioural importance. Even the highest individual levels observed, 0.52 mM (0.02 ‰) and 1.03 mM (0.05 ‰), are so low that they cannot be assumed to have any influence upon the bird's flying ability and behaviour.

The Waxwings ingested an average of 0.7 g/kg of alcohol per day during the autumn experiment and 23 g/kg during the winter experiments. Despite this high intake in winter the blood alcohol levels cannot have risen very high because Waxwings possess such an extremely fast rate of alcohol metabolism: approximately 1 g/kg is eliminated in one hour.

The alcohol values observed in caged birds cannot be compared directly to the values in birds in their natural environment. Keeping the cages outside duplicated the natural temperature and lighting conditions. Birds in cages require, however, less nutrition than they do in nature, where they expend 30—50 % more energy in finding food and in their other activities (e.g. Uramoto 1961; this estimate may be too high because some of the birds studied by Uramoto were nesting at the time). The alcohol concentrations in rowan berries may be higher in nature than they were in the artificially fermented berries. The rate of eating also varies during the day, mostly according to the weather conditions and the amount of available nutrition.

We were not able to measure alcohol concentrations in birds that were in their natural environment, and, therefore, do not know the true values. It is possible, however, to make a rough estimate of how high the alcohol levels in birds in nature might go. If a Waxwing in nature ate 100—120 g of rowan berries per day that averaged 2.4 g/kg of ethanol (the highest value measured in rowan berries), 4—5 g/kg of ethanol daily would enter the bird's intestines. If the Waxwing ate 30 g of these berries in one hour, the alcohol concentration in the water phase of the bird would be 10.9 mM, in spite of its alcohol elimination. This high ethanol concentration might be expected to affect the bird's ability to fly. Unfortunately no behavioural tests were included in the alcohol metabolism experiment here. Consequently, although intoxication was clearly evident after injection of the larger dose and some indications were seen after the smaller dose, it is not possible to specify the blood alcohol level necessary to impair flying. In humans, however, alcohol levels of 10.9 mM, i.e. 0.5 ‰, have been found to cause impairment of motor coordination (Mattila & Ylikahri 1977), and it seems likely that this same alcohol level might influence flying, which requires very precise coordination.

Bullfinches probably consume less food daily than do Waxwings. The food stays in a Bullfinch's alimentary canal longer, because seeds are more difficult to digest than the berry pulp and because the Bullfinch has an intestine that is 4.6 times as long as that of the Waxwing (Pulliainen et al. 1981). While Bullfinches are eating rowan berries some of the berry pulp gets into their alimentary canal along with the seeds, and therefore the blood alcohol levels may rise. The amount of alcohol ingested is however far less than in Waxwings, which eat the whole berries. Consequently, although a preliminary experiment showed that Bullfinches (N=3) eliminated alcohol at about 200 mg/kg/h, i.e. 4.5 times as slowly as Waxwings, it is unlikely that the blood alcohol becomes high enough in Bullfinches to produce intoxication.

The rate of alcohol metabolism has been found to correlate positively to the basal metabolic rate (BMR) of many animals (Lester & Keokosky 1967, Wallgren & Barry 1970, Videla et al. 1975). The rapid alcohol metabolism of Waxwings can be related largely to their high BMR, 272 kcal/kg/day (Pinowski & Kendeigh 1977). The BMR of Bullfinches, however, is even higher: 375 kcal/kg/day. Although ADH activity is not generally considered to be a limiting factor in ethanol metabolism (Wallgren & Barry 1970, Higgins 1979), both Greenfinches and Starlings have such low liver ADH activities that they probably do limit the alcohol metabolism. Pyrazole, which inhibits ADH activity in a concentration-dependent manner, is known to lower the rate of alcohol metabolism *in vivo* (Kalant et al. 1975), thus demonstrating that reductions in ADH activity can affect ethanol elimination. The ADH enzymes of Greenfinches and Starlings also differ in their isoenzyme composition from that of Waxwings. The enzyme kinetics for the ADH enzymes of passerines has not been studied in detail, however, and it is possible that the K_m values of the enzymes were high for ethanol, in which case only higher amounts of ethanol can saturate the enzyme.

Both the BMR of Starlings — 247 kcal/kg/day (Pinowski & Kendeigh 1977) — and their ADH activity are lower than in Greenfinches. Alcohol is metabolized, however, twice as rapidly by Starlings as by Greenfinches. This could indicate that Starlings possess a route other than ADH, for example, MEOS (microsomal ethanol-oxidizing system), which is largely responsible for their alcohol metabolism (cf. Burnett & Felder 1980).

Waxwings, Starlings, and Greenfinches all eliminated the larger dose of alcohol (2 g/kg) about 40 mg/kg/h faster than the smaller dose (1 g/kg). The difference was significant only in Starlings and Greenfinches, but the similar magnitude of the differences suggests that the same factor is responsible for the increased rate of elimination in all three species. This factor may be ADH enzymes with high K_m values for ethanol, because concentration differences in this low range have generally not been found to influence the metabolic rate in rats (Higgins 1979), whose ADH has a K_m value of only 2 mM (Theorell & McKinley-McKee 1961). There are, howerer, factors other than high K_m ADH that can cause higher ethanol concentrations to be eliminated more rapidly. In the rat, for example, a very high dose of ethanol, 5 g/kg, increases alcohol elimination from the blood (Wendell & Thurman 1979), an effect that cannot be attributed to high K_m ADH.

A comparison of the rates of alcohol metabolism in Waxwings, Starlings, Bullfinches, and Greenfinches seems to suggest that in each species the rate is related to the relative quantity of ethanol-containing food in its diet. The Waxwing is, according to Mosen (1975), the only avian species in the temperate zone whose primary source of nutrition is berries. Furthermore, the Waxwing consumes a large quantitity of berries daily, which it is able to do because its intestines are short and the easily digested food normally travels rapidly through its alimentary canal (Pulliainen et al. 1981). Borowski (1966) found that half of the berries he fed to a Waxwing were excreted within 27 minutes and 90 % within 33 minutes. Restricted to food sources that frequently contain alcohol and consuming large amounts daily, the Waxwing would often face the danger of having ethanol accumulate in its system if the bird were not capable of metabolizing it rapidly. The Waxwing's liver relative to its body weight is considerable larger than that of other birds that have been studied. Voronov & Voronov (1978) suggest that the large liver is an adaptation for producing and storing glycogen from the carbohydrate-rich diet. A large liver could as well be an adaptation for removing toxins, such as alcohol, that are found in the berries.

Starlings, like Waxwings, have short intestines (Pulliainen et al. 1981) and also eat berries when they are available. Consequently, alcohol may also accumulate in Starlings and the species requires a relatively rapid rate of ethanol metabolism. Starlings, however, are not restricted to berries, but rather eat a mixed diet. A very high rate of ethanol metabolism would, therefore, not be as important for Starlings as it is for Waxwings.

The diet of Bullfinches only rarely includes alcohol and the species probably also consumes a smaller total quantity of food per day compared with Waxwings. Bullfinches metabolize alcohol slowly. Greenfinches are even less efficient than Bullfinches at metabolizing alcohol, but this species, being strictly a seed eater, never ingests alcohol in its diet.

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Selostus: Marjaravinnon sisältämän alkoholin kertymisestä lintuihin sekä varpuslintujen alkoholiaineenvaihdunnasta

Tutkimuksessa selvitettiin, missä määrin pihlajan- ja orapihlajanmarjoissa sekä ruusunkiulukoissa tapahtuu alkoholikäymistä luonnossa, sekä kertyykö marjaravinnosta tilhiin ja punatulkkuihin haitallisia määriä alkoholia. Lisäksi tutkittiin, eroavatko marjaravintoon sopeutunut tilhi, sekaravintoa käyttävä kottarainen ja pääasiassa siemenravintoa käyttävä viherpeippo alkoholiaineenvaihdunnaltaan toisistaan.

Luonnonmarjojen etyyli-alkoholipitoisuudet määritettiin kaasukromatografisesti noin kuukauden välein syys-helmikuussa. Pihlajanmarjojen ja ruusunkiulukoiden etanolipitoisuudet kohosivat selvästi syksyn ja talven kuluessa (kuva 1). Orapihlajanmarjojen alkoholipitoisuudet pysyivät alhaisina seurannan ajan.

Tilhien ja punatulkkujen marjaruokintakokeet tehtiin ulkohäkeissä loka-marraskuun vaihteessa ja tammikuussa. Linnut saivat ravinnokseen pihlajanmarjoja, joita pidettiin tarjolla vuorokauden valoisan ajan. Verinäytteet otettiin auringonnousun aikaan, keskipäivällä, auringonlaskun aikaan ja keskiyöllä (ns. 0-arvot). Talvikokeessa tilhien ja punatulkkujen veren alkoholipitoisuudet keski- ja iltapäivällä olivat selvästi korkeampia kuin syyskokeessa (taul. 1); marjoissa oli keskitalvella enemmän alkoholia ja linnut kuluttivat kylmällä säällä runsaammin ravintoa. Korkeimmatkin yksilölliset veren etanolipitoisuudet, 0,52 mM (0,02 ‰) ja 1,03 mM (0,05 ‰), olivat kuitenkin niin pieniä, ettei niiden voida olettaa vaikuttavan humalluttavasti ja siten lintujen lentokykyä ja käyttäytymistä haittaavasti. Verestä mitattujen ns. 0-arvojen perusteella kuvussa ja suolistossa ei tapahtunut endogeenistä alkoholin muodostumista.

Teoreettisten laskelmien perusteella tilhien veren alkoholipitoisuus saattaa luonnossa kohota humalluttavalle tasolle (0.5 ‰) lintujen syödessä paljon käyneitä marjoja. Punatulkut, jotka syövät siementen ja silmujen ohella vain vähän marjaravintoa eivät luonnossa koskaan humaltune.

Alkoholin poistumisnopeudet määritettiin tilheltä, kottaraiselta ja viherpeipolta kahdella annosalueella (1 g/kg ja 2 g/kg; kuva 2). Maksojen alkoholidehydrogenaasieli ADH-aktiivisuudet määritettiin spektrofotometrisesti ja ADH- isoentsyymit geelielektroforeettisesti. Alkoholi (1 g/kg) poistui tilhen verestä 3.3 kertaa nopeammin kuin kottaraisen ja 7.2 kertaa nopeammin kuin viherpeipon verestä (taul. 2). Kottarainen hapetti alkoholia 2.2 kertaa nopeammin kuin viherpeippo. Tilhen vilkas perusaineenvaihdunta, 272 kcal/kg/vrk, nopeuttaa alkoholin poistumista. Viherpeiposta alkoholi poistui yllättävän hitaasti, vaikka sen aineenvaihdunta, 397 kcal/kg/vrk, on vilkkaampi kuin tilhellä. Vaikka alkoholidehydrogenaasiaktiivisuutta ei yleensä pidetä palamisnopeutta rajoittavana tekijänä, viherpeipon ja kottaraisen ADH-aktiivisuus on niin alhainen (taul. 3), että se rajoittanee alkoholin palamista. Näiden lajien ADH-entsyymit erosivat myös isoentsyymirakenteeltaan tilhen entsyymeistä. Kaikki tutkitut varpuslinnut poistivat suuremman alkoholiannoksen suhteellisesti nopeammin kuin pienemmän (taul. 2), mikä viittaa siihen, että näiden lajien ADH:n K_m-arvot etanolille olisivat korkeita.

Alkoholin eliminoituminen näyttää olevan sitä nopeampaa, mitä enemmän marjaravintoa laji käyttää. Tilhi on lauhkean vyöhykkeen ainoa pääravintonaan marjojen maltoa käyttävä laji. Sen suolisto on lyhyt ja helposti sulavan ravinnon kulkeutuminen ruoansulatuskanavassa on nopeaa, mikä mahdollistaa runsaan päivittäisen marjojen kulutuksen. Linnun on kyettävä poistamaan tehokkaasti elimistöstään marjaravinnon mahdollisesti sisältämää alkoholia. Tilhen maksa suhteessa ruumiin painoon on huomattavasti suurempi kuin muiden tutkittujen lintujen, mikä saattaa olla sopeutuma myrkkyjen kuten alkoholin poistamiseen elimistöstä. Kottaraisen suolisto on lähes yhtä lyhyt kuin tilhen. Laji syö muun ravinnon ohella myös marjoja, ja voi saada elimistöönsä alkoholia, joka poistuu melko tehokkaasti. Esikokeessa todettiin punatulkun polttavan alkoholia hitaasti (200 mg/kg/t), mutta siementen ja silmujen syöjänä sen ravinto vain harvoin sisältää alkoholia. Viherpeipon alkoholin poistamismekanismi oli punatulkunkin mekanismia tehottomampi; laji onkin täydellinen siemensyöjä eikä sen ravinto sisällä alkoholia.

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