LIVER LIPIDS IN HUMMINGBIRDS

By FRANK A. HARTMAN and KATHARINE A. BROWNELL

In order to maintain a body temperature above that of their environment, warm-blooded animals must have a constant supply of fuel which comes from recently digested food or from material previously stored. Such storage is primarily in the form of fat. Therefore the determination of the amount of fat gives the most accurate measure of the ability to withstand periods of excessive environmental demand or inadequate food supply.

Because birds have a higher body temperature and are at times subjected to greater demands than mammals, it is important that they have a readily available supply of fuel. Therefore fat deposits in birds are of great importance and should receive more attention than has been given them. Many casual observations on fat have been made in connection with other studies, but quantitative determinations are relatively few and these are mostly in relation to migration.

Because of the difficulty in determining total body fat, and because such deposits represent less readily available stores, it was thought that determination of liver lipid might indicate the importance of fat in the metabolism at a given time. The fat of the liver is that proportion of the total lipids which is being processed either for storage or immediate use. Moreover, it has been shown by Odum and Perkinson (1951) that the amount of liver fat reflects the amount of total body lipid.

The hummingbirds, one of the most active types of birds, possess such large amounts of lipid in the liver that this organ in them frequently becomes creamy white in color. Because of this observation we became interested in determining the actual amount of lipid present. This has been compared with the values in other birds.

ACKNOWLEDGMENTS

This work was supported by grants from the Comly Fund and the Graduate School of Ohio State University. We are indebted to Dr. Carl Johnson, Director of the Gorgas Memorial Laboratory, Panamá, for the use of the Juan Mina Field Station, to Mr. Pablo Brackney, who furnished living quarters near El Volcán, and to Mr. Toliffe Mônniche for his help on our expedition up Mount Copete near Boquete.

METHODS

Birds shot in the field were enclosed in sealed vials or waterproof bags until they reached the laboratory. They were then weighed on a torsion balance sensitive to 2 mg. The livers were weighed on a Roller-Smith precision balance sensitive to 0.02 mg. For determination of body lipids (lipids include fats and a small quantity of other compounds soluble in the solvents used), the head, feathers, feet, and gastro-intestinal tract were removed; the remainder of each specimen was cut into small pieces for later grinding and extraction. All material for lipid determination was preserved for analysis in redistilled ethyl alcohol. Subsequently, this material, liver or body, was ground to a pulp with sand. The pulp was thoroughly extracted with a boiling 3 to 1 alcohol-ether mixture as suggested by Bloor (1943). After removal of the solvent, the residue was extracted with boiling petroleum ether. The ether was removed and the residue, determined gravimetrically, is reported as total lipid.

Hummingbirds were collected in Panamá in January and February both at sea level and at elevations of 4500 and 7000 feet. Other specimens were collected in Oxford County, Maine, in July, August, and September.

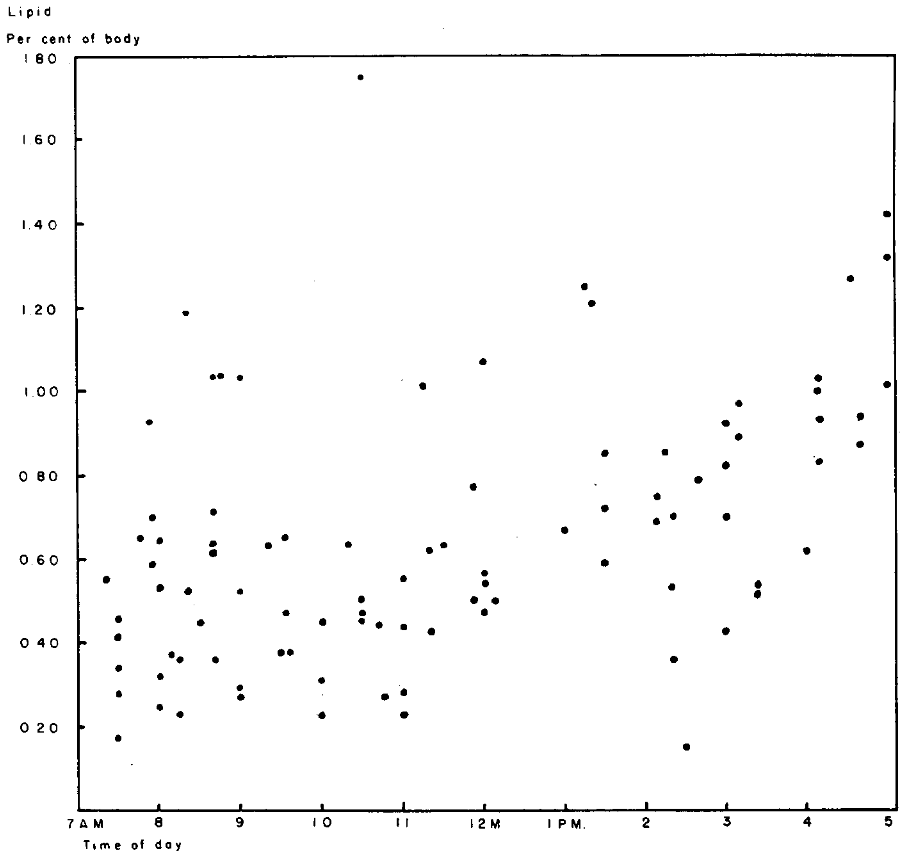

Fig. 1. Liver lipid as per cent of body weight in various species of hummingbirds collected at different times of the day.

RESULTS

The liver and its lipid in hummingbirds.—The two most common species of hummingbirds used in our study were the Rufous-tailed Hummer (*Amazilia tzacatl*) and the Scintillant Hummer (*Selasphorus scintilla*). They were represented by 15 and 12 individuals, respectively. In these species there was no significant difference in liver weights between the sexes or between specimens collected in the morning or afternoon. This was due to the great range in values. On the other hand, if we consider all specimens of hummingbirds collected at sea level, the liver averaged 3.35±0.14 per cent of the body weight before noon and 3.88±0.16 per cent after noon. This difference is significant ($p<0.05$).

The amount of lipid in the liver frequently ranged from 15 to 30 per cent of liver weight. These high values account for the creamy color of the liver. The range of values was determined from more than one hundred individuals distributed among 22 species, collected at different times of the day at elevations ranging from sea level to 7000 feet. Figure 1 shows more than 90 of these determinations. The afternoon values tend to be higher than those in the morning. Except for a few very high morning values the trend does not start upward until after 11 a.m. It then continues upward until 5 p.m. when the last values were obtained. If a median is run, it starts at about 0.50 per cent of body weight in the morning, and is 0.60 per cent at noon, 0.70 to 0.80 per cent at 3 p.m., 0.90 per cent at 4 p.m., and over 1.0 per cent at 5 p.m.

Because of the difference associated with elevation, we recalculated the values for birds from sea level alone. They are 0.37±0.03 per cent (23 specimens) for the forenoon and 0.87±0.05 per cent (22 specimens) for the afternoon.

A breakdown of the species represented by five or more individuals is given in table 1. It can be seen that all show an increase in the afternoon except *Selasphorus,* which was also high in the morning. Species represented by fewer than five individuals conformed more or less to the general pattern. They were *Amazilia amabilis* (3), *Amazilia decora* (2), *Anthracothorax nigricollis* (2), *Chalybura buffonii* (1), *Eugenes spectabilis* (1), *Elvira chionura* (2), *Heliodoxa jacula* (1), *Campylopterus hemileucurus* (3), *Phaethornis longuemareus* (1), *Phaethornis superciliarus* (1), *Phaethornis guy* (3), *Florisuga mellivora* (1), *Colibri thalassinus* (3), and *Eupherusa eximia* (4).

Table 1

Average Values for Liver Lipid in Hummingbirds
(numbers of individuals in parenthesis)

Species	Body weight in grams	Liver lipid as per cent of body weight a.m.	p.m.
Selasphorus scintilla	(9) 2.27±0.07*	(9) 0.71±0.06	(3) 0.75±0.02
Chlorostilbon canivetii	(5) 3.07±0.02	(2) 0.31	(5) 0.71±0.08
Damophila julie	(5) 3.23±0.07	(2) 0.35	(3) 0.75±0.17
Amazilia edward	(8) 4.56±0.04	(6) 0.43±0.01	(2) 0.81
Amazilia tzacatl	(16) 4.94±0.04	(4) 0.41±0.11	(12) 0.93±0.09
Lampornis castaneoventris	(8) 5.31±0.04	(5) 0.44±0.08	(3) 0.58±0.02
Glaucis hirsuta	(5) 6.26±0.12	(4) 0.29±0.06	(1) 0.92
Phaeochroa cuvierii	(6) 8.93±0.06	(3) 0.36±0.11	(3) 0.57±0.17

*Standard error.

Since few birds were collected in the afternoon at high altitudes, only those collected before noon are used in comparing the values at different elevations. The liver lipid of 39 specimens obtained at 4500 feet above sea level was 0.66±0.05 per cent of the body weight. Compared with the birds from sea level already mentioned, this is almost double their value. It might be pointed out that *Selasphorus* was collected at 7000 feet above sea level.

It is interesting to note that a hummingbird (*Chlorostilbon*) weighing 3.08 grams, wounded at 3:25 p.m. and kept in the dark for 22 hours, contained liver lipid after that time which amounted to 0.68 per cent of the body weight.

Lipids of both the liver and body were determined in 21 hummingbirds of species already mentioned, all of which were collected before 1:00 p.m. The values varied considerably. The total lipid in the body was 6.12±0.47 per cent (range, 3.33 to 11.39 per cent). Of this, 0.70±0.07 per cent, or a little more than one-tenth, was present in the liver, which averaged 3.68±0.25 per cent of the body weight. The liver lipid bore no relationship to the total body lipid. A low body lipid might be accompanied by a high liver lipid and *vice versa*: to illustrate, in two *Amazilia* one had 3.97 per cent body lipid and 7.62 per cent liver lipid while the other had 4.03 per cent body lipid and only 3.17 per cent liver lipid. Two hummingbirds with liver lipids of 9.23 and 8.72 per cent, respectively, contained 4.80 and 9.82 per cent body lipids.

Liver lipid in warblers.—The appearance of the livers of birds in families other than the Trochilidae indicated lower lipid content. We therefore chose to examine the livers of warblers because these birds are active and are largely insectivorous.

The liver lipid in 43 warblers distributed among eleven species is shown in figure 2. The livers of birds collected in the morning weighed less (26 individuals, 3.63±0.095 per cent of the body weight) than those of birds collected in the afternoon (16 individuals, 4.35±0.098 per cent). The difference was highly significant ($p < .01$). The lipid content of the liver of those obtained in the morning was 0.28±0.013 per cent and of those obtained in the afternoon 0.54±0.029 per cent of the body weight, a highly significant difference ($p < .01$). The body weight of the warblers ranged from 7.3 to 19.2 grams, most of them being 9 grams or more. These values for liver fat are significantly lower than those in hummingbirds and the afternoon increase was less.

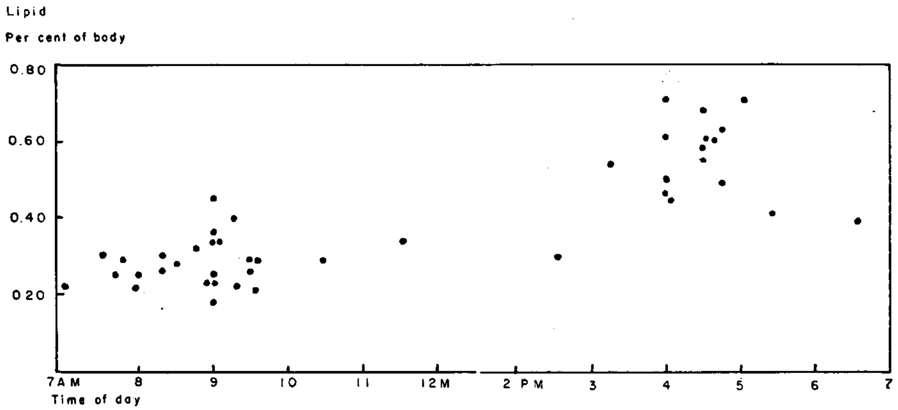

Fig. 2. Liver lipid as per cent of body weight in several species of warblers.

The liver and its lipid in other families.—The observations on warblers led us to examine the liver lipid content in birds of other families. The values are shown in table 2. The liver in per cent of body weight ranges from 1.23 to 5.10 but in many of them it ranges from 3 to a little more than 4 per cent. Few birds were collected in the afternoon but all that were, except the Catbirds (*Dumetella carolinensis*), showed an increase in liver lipid. The liver lipid for birds collected in the morning ranged from 0.12 to 0.35 per cent of the body weight, while the range in the afternoon was from 0.16 to 0.47 per cent.

The parakeets should be considered apart, since they were laboratory reared, indoors, from the time of hatching. They were also of the same strain from two nests and were four and six months old. Their food consisted of seeds with occasional leafy vegetables. It will be noted that the livers were small and that the liver lipid was very low.

Among these families we have chosen five which are largely insectivorous, namely, tyrant flycatchers, titmice, nuthatches, kinglets, and vireos, for a comparison of morning and afternoon liver lipids. In the morning, liver lipids formed 0.25±0.03 per cent and in the afternoon 0.38±0.045 per cent of the body weight. This difference, which is significant ($p < 0.05$), indicates a 50 per cent increase for the afternoon, but this is a smaller increase than was present in the warblers and it is a very much smaller increase than that found in hummingbirds. Comparison of the three groups, hummingbirds, wood warblers, and other insectivorous birds, is shown in table 3. It will be noted that the relative liver weights do not differ greatly while the liver lipid gain in the afternoon is much the greatest in the hummingbirds.

Thus, the lipid content of the liver of hummingbirds is higher at all times than that of other birds studied and it increases more in the afternoon than does that of other birds.

Table 2

Average Values for Liver Lipids in Other Families

Group	Body weight in grams	Liver weight as per cent of body weight	Liver lipid as per cent of body weight a.m.	p.m.
Woodpeckers	(2) 46; 66	(2) 1.78; 4.57	(2) 0.22; 0.13	
Tyrant flycatchers	(4) 9; 10; 21.4; 21.5	(4) 3.22±0.18	(3) 0.24±0.018	(1) 0.39
Titmice	(8) 11.1±0.21	(8) 3.13±0.19	(7) 0.25±0.013	(1) 0.37
Nuthatches	(3) 10.2±0.56 (5) 21.7±0.36	(8) 3.11±0.37	(5) 0.15±0.022	(3) 0.28±0.032
Kinglets	(3) 5.92±0.053	(3) 4.12±0.27	(3) 0.32±0.012	
Vireos	(3) 16.1±0.8	(3) 3.45±0.18	(2) 0.25; 0.26	(1) 0.47
Jays	(2) 89.8; 93.6	(2) 3.17; 1.89	(1) 0.12	(1) 0.16
Thrushes	(4) 34.4±1.40	(4) 3.77±0.11	(4) 0.26±0.032	
Catbirds[1]	(2) 36.8; 38.3	(2) 5.10; 4.40	(1) 0.35	(1) 0.33
Sparrows[2]	(3) 18.4±0.37	(3) 3.58±0.19	(3) 0.23±0	
Parrots[3]	(8) 33.7±0.53	(8) 1.23±1.01	(8) 0.063±0.003	

[1] *Dumetella carolinensis*; [2] *Melospiza georgiana*; [3] *Melopsittacus undulatus*.

Table 3

Comparison of Values for Liver and Liver Lipids

	Range in body weight in grams	Liver as per cent of body weight (average) a.m.	p.m.	Liver lipid as per cent of body weight (average) a.m.	p.m.	Per cent increase in p.m.
Hummingbirds	3–10	(23) 3.35±0.14	(25) 3.88±0.16	0.37±0.030	0.87±0.050	135
Wood warblers	7–10	(26) 3.63±0.095	(16) 4.35±0.098	0.28±0.13	0.54±0.029	93
Other insectivorous birds	6–16	(20) 3.28±0.25	(6) 3.23±0.22	0.25±0.03	0.38±0.045	52

DISCUSSION

In our observations, differences in sex or time of collection did not affect liver weights. Riddle (1928), who worked with specimens of the same strain of Ring Dove (*Streptopelia risoria*) under controlled conditions, found that liver weights were greater in the female than in the male. We used a small number of specimens, many of which are undoubtedly of different genetic constitution. If we compare all species of hummingbirds, the difference between the morning and afternoon values is significant. A greater number of determinations might have increased the significance. Fisher and Bartlett (1957) found a decrease in liver weight overnight in both Redwinged Blackbirds (*Agelaius phoeniceus*) and Starlings (*Sturnus vulgaris*).

In regard to the liver lipid in hummingbirds a few aberrant values make the range great, but despite this there is no general change during the morning until 11 a.m. After this, there is a very definite increase continuing through 5 p.m. when the last values were obtained. This holds as well for individual species except in *Selasphorus scintilla* where the value was as high in the morning as in the afternoon. We are unable to suggest any reason for this sustained high value in this species unless it is a result of altitude.

Liver lipid, essentially fat, represents material mobilized from the storage depots or synthesized from food. In the morning much sugar is being ingested and by afternoon carbohydrate which has not been utilized to sustain the immediate activities of the bird is being processed as fat. During the night the greatly reduced activity of the hummingbird in sleep or torpidity no doubt is accompanied by reduced processing and might account for the lipid fall. Pearson (1950) has shown that the metabolism of the torpid hummingbird may drop to as low as $\frac{1}{28}$ of the level of the daylight hours.

The amount of lipid in the liver accounts for a very small portion of the energy used in 24 hours. Pearson (1954) calculated that the energy exchange of a male Anna Hummingbird (*Calypte anna*) weighing 4 grams was 7.55 calories (assuming torpidity at night) or 10.32 calories (assuming sleep at night) for 24 hours. The average weight of the hummingbirds which were used in our work to calculate the difference in liver lipid content between the morning and afternoon was 5.29 grams. The difference between the liver lipid taken in the morning and afternoon was 0.50 per cent of the body weight (0.87–0.37); this would yield 0.25 cal. [0.50 per cent of 5.29 (average body weight) $\times$ 9.4 cal. (accepted value for 1 gram of fat)]. This could be used at night or shifted to the body stores. However, it is but a small portion of that needed since either 0.89 or 4.53 calories would be used at night if the situation were similar to that in Pearson's bird.

Fisher and Bartlett (1957) reported a decrease in both fat and glycogen stores in the liver overnight. However, the time permitted to elapse between the killing and freezing of the liver for glycogen determinations (one-half hour to one and one-half hours) would make the results questionable, as glycogen is broken down rapidly after death. The great range for glycogen values bears this out.

The greater amount of liver lipid at higher altitudes is noteworthy. It might be a reflection of the increased metabolism required because of the lower temperatures at high elevations.

All the hummingbirds in our study were tropical forms which apparently do not store as large a fat reserve as do species which migrate. The average total lipid in the body for 21 birds collected before noon at sea level was fairly low (6.12$\pm$0.47 per cent) while Norris, Connell, and Johnston (1957) found a range of 11 to 15 per cent in specimens of the Ruby-throated Hummingbird (*Archilochus colubris*) collected in June and a range of 41 to 46 per cent in pre-migratory individuals. Our highest value was 11.39 per cent. Odum and Connell (1956) reported that the total lipid in the body of the Ruby-throated Hummingbird averaged 13.4 per cent during the non-migratory period and 43 per cent just prior to migration. Musacchia (1953) obtained high lipid in the liver in Anseriformes and Charadriiformes in late summer during molting and the pre-migratory period.

Why are liver lipids of hummingbirds so high? Trochilids are not only the most active of birds but they are the smallest. Smaller birds present a greater surface exposure, and they require a higher rate of metabolism to maintain body temperature than do larger birds. Since diet may be a factor in the production of high liver lipid, we have also studied other small and fairly active birds, the wood warblers, which have a more uniform diet of insects. In this group the increase in liver weight in the afternoon over that in the morning was highly significant ($p < 0.01$), this increase being of greater significance than that in hummingbirds ($p < 0.05$). However, the liver lipid was lower and showed a relatively smaller increase in the afternoon in the warblers. Thus, the additional carbohydrate in the diet of hummingbirds may be a factor influencing the high lipid content of the liver.

It should be emphasized that other birds used in our study were collected during the pre-migratory period and are therefore perhaps not comparable with our hummingbirds which were all collected in the tropics and were non-migratory. Despite this, the difference in the liver lipid of the hummingbirds is striking.

If we compare the wood warblers with somewhat larger insectivorous birds we find that the liver lipids of the former were higher in the afternoon ($p < 0.01$ than those of the larger birds, although in the morning, values of both groups were nearly the same.

The difference between the two groups of seedeaters (fringillids and psittacids) can be explained. The sparrows were living in a wild state and were pre-migratory while the parakeets were domesticated and non-migratory. This suggests that the increased processing of the food in the liver is accompanied by higher lipid content.

As we noted earlier, Odum and Perkinson (1951) found that the liver lipid bore a direct relationship to the total body lipid, that is, when the latter was low, as in the post-migration period, the liver lipid was also low, whereas during the pre-migration period, when body lipid was high, the liver lipid was also high. Their observations were on the White-crowned Sparrow (*Zonotrichia leucophrys*), a seedeater, whereas the hummingbird takes much sugar. The latter is processed more rapidly than are starches, fats, and proteins. This might account for the greater fluctuation in liver lipid. Thus the liver lipid might follow more closely the sugar intake.

We may conclude, tentatively, that the great difference in lipid content of the liver in hummingbirds as compared with other birds may be accounted for by the large amount of carbohydrate in their diet. Other birds studied were mostly insect eaters, a few being seedeaters. Small size is no doubt a factor, in causing high lipid values, but in addition muscular activity is important, the hummingbirds being one of the most active of all birds during their waking hours.

SUMMARY

Liver lipids were determined in 22 species of hummingbirds collected in Panamá. They were compared with those of members of other families collected in Maine. The livers of hummingbirds collected in the afternoon were possibly heavier than those obtained in the morning. The lipid values in these livers tended to be higher in the afternoon than in the morning. In sea level birds, the morning values were lower than the afternoon values. The morning values of the liver lipid of birds collected at 4500 feet above sea level were higher than morning values at sea level. The total lipid in the body was 6.12 ± 0.47 per cent whereas the liver lipid in these specimens was 0.70 ± 0.07 per cent.

In warblers the livers were heavier in the afternoon and contained more lipid than in the morning. The lipid content of the liver was lower than that in hummingbirds and the afternoon increase was less.

In other families the liver lipid was still lower. The relative liver weights of the different families of birds did not differ greatly except in domesticated parakeets in which both the liver and liver lipid were much the smallest.

The great difference in lipid content of the liver in hummingbirds as compared with other birds may be partly accounted for by the large amount of carbohydrate in the diet. The small size of the bird is no doubt a factor but in addition muscular activity is important.

LITERATURE CITED

Bloor, W. R.
1943. Biochemistry of the fatty acids and their compounds, the lipids (Reinhold, New York).

Fisher, H. I., and Bartlett, L. M.
1957. Diurnal cycles in liver weights in birds. Condor, 59:364–372.

Musacchia, X. J.
1953. A study of lipids in arctic migratory birds. Condor, 55:305–312.

Norris, R. A., Connell, C. E., and Johnston, D. W.
1957. Notes on fall plumages, weights and fat condition in the ruby-throated hummingbird. Wilson Bull., 69:155–163.

Odum, E. P., and Connell, C. E.
1956. Lipid levels in migrating birds. Science, 123:892–894.
Odum, E. P., and Perkinson, J. D., Jr.
1951. Relation of lipid metabolism to migration in birds: seasonal variation in body lipids of the migratory white-throated sparrow. Physiol. Zool., 24:216–230.
Pearson, O. P.
1950. The metabolism of hummingbirds. Condor, 52:145–152.
1954. The daily energy requirements of a wild Anna hummingbird. Condor, 56:317–322.
Riddle, O.
1928. Sex and seasonal differences in weight of liver and spleen. Proc. Soc. Exp. Biol. (New York), 25:474–476.

Department of Physiology, The Ohio State University, Columbus, Ohio, October 1, 1958.