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Quantification of Diets

APPROACHES TO AVIAN DIET ANALYSIS

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Abstract. Direct examination of diets is greatly under-represented in studies of avian biology. Much of our knowledge of food habits of North American birds is still based on the early survey work by "economic ornithologists." Here, we review approaches and techniques of sampling and analysis. For species that cannot be captured alive, collection of stomach or esophageal samples is necessary. Potential biases associated with post-mortem digestion, time spent in nets or traps, and differential passage of food through various parts of the gut are discussed. For species that can be captured alive, flushing the digestive tract or forcing regurgitation with warm water is recommended over use of emetics. Fecal samples and pellets, although more difficult to analyze, also provide accurate estimates of diet. Diets of nestling birds may be sampled with neck ligatures, observed or photographed directly at nests, or examined through the transparent skin of the neck. Aids for the identification of fragmented food samples are discussed, including the use of reference collections, collaboration with specialists, and the conversion of arthropod fragment sizes to total prey length, weight, and energy content. Diet data may be presented as percent occurrence, frequency, volume, or weight, each with its own merits and biases. We recommend presenting at least two kinds of results, as well as the raw data, on a per-stomach basis whenever possible. Finally, we describe two under-used sources of diet information: the U.S. Biological Survey stomach analysis card file at Patuxent Wildlife Research Center and the unanalyzed stomach contents collection at Louisiana State University Museum of Natural Science.

Key Words: Diet analysis; emetics; fecal analysis; ligatures; pellets; stomach contents.

Detailed knowledge of diets is critical to many studies of avian biology and ecology. However, direct measures of diets are rarely attempted. The common use of indirect inferences about diets, based on morphology (e.g., bill shape), behavior, or general food availability, has been questioned in several empirical studies (e.g., Rotenberry 1980a, Rosenberg et al. 1982). The extent to which variation in foraging behavior results in differences in diet (cf. MacArthur 1958, Cody 1974) also remains largely untested. Most recent, frequently cited studies of avian foraging guilds or communities (e.g., Rabenold 1978, Eckhardt 1979, Holmes et al. 1979b, Noon 1981a, Sabo and Holmes 1983, Mountainspring and Scott 1985, Remsen 1985) provide no quantitative measure of local diets, although most make conclusions regarding resource partitioning, optimal foraging, or interspecific competition (for exceptions, see Rotenberry 1980a, Rosenberg et al. 1982, Robinson and Holmes 1982, Sherry 1984). Because we lack clear understanding of the connections between foraging site-selection, food availability, and diet, any assumptions made without further empirical study may be unwarranted.

In a recent symposium on neotropical migrants (Keast and Morton 1980), 15 papers specifically discuss foraging ecology; yet, in only three were diets of individual species described to any extent, and only one study (Morton 1980) provided quantitative data on local diets. In this volume, only one paper (Cooper et al.) specifically addresses the determination of avian diets or provides diet data relevant to the study. In a sample of roughly 200 papers on avian food habits compiled from major ornithological journals since 1960, 68 (34%) concern only waterbirds, 70 (35%) are on raptors, and 13 (7%) deal with gamebirds. Finally, of the 50 papers (25% of total) concerning nongame landbirds, 30 were single-species studies, most from single localities, leaving only a handful that may be useful to community ecologists, ecomorphologists, or other comparative biologists. To date, the only source of diet information for most North American bird species remains the survey data of F. E. L. Beal and W. L. McAtee, summarized in Bent's Life history series, and Martin et al. (1951a). Wheelright's (1986) analysis of the American Robin (*Turdus migratorius*), is the only modern study of geographic or seasonal variation in diet in any North American bird.

Why, then, are avian diets so neglected? We think the reasons are more methodological than philosophical: (1) the variety of alternative approaches and options is not generally appreciated; (2) researchers fear the detail and lack the technical expertise required by the fragmented nature of most diet samples; and (3) data on diets are initially collected, but samples are either not analyzed or the results are not subsequently published. We know the latter to be true in several studies cited above. To the extent that reasons

(1) and (2) are true, we offer this review in the hope of alleviating such fears and stimulating further study.

Our goal is not to provide a handbook of techniques, but rather to lead the reader to appropriate references and provide examples in which each technique has either succeeded or failed. Our biases reflect our own work (primarily with stomach analysis) on temperate and neotropical insectivorous birds, although we have attempted to broaden the scope of our review.

Sampling Techniques

The first major review of avian dietary assessment by Hartley (1948) still applies to most modern studies. Hyslop's (1980) review of methods for analyzing stomach contents of fishes discusses many topics relevant to avian studies and may serve as a basic reference in any dietary investigation. Duffy and Jackson (1986) offer the most recent discussion of sampling and analytical considerations for studies of seabird diets, and most of their discussion applies equally well to terrestrial birds. Ford et al. (1982) review modern, nondestructive methods of sampling gut contents. Other useful discussions of general sampling considerations may be found in Newton (1967), Swanson and Bartonek (1970), and Rundle (1982).

Stomach contents

The most common method of avian diet sampling is direct examination of gut contents. Its primary advantages are (1) adequate samples can usually be obtained relatively easily, and (2) the full contents of a bird's gut are obtained. Disadvantages include the need to kill birds, and the many biases associated with stomach fullness, differential digestion rates, identification of fragmented food items, and presentation of results. These biases, however, are common to all techniques involving gut samples, whether or not the bird is sacrificed.

The techniques used to obtain and analyze gut contents today are similar to those first devised by early researchers attempting to determine the economic importance of North American birds (e.g., MacAtee 1912, 1933). The first consideration is the method and design of specimen collecting. Ideally, only actively foraging individuals will be sampled, controlling for habitat heterogeneity, season, time of day, and the like. These factors are most easily controlled by shooting, and many species (e.g., in the forest canopy or very open habitats) can be sampled only in this way. Duffy and Jackson (1986) criticized the random shooting of birds at sea that may be travelling long distances between foraging sites and thus may have empty or mostly digested gut contents. This problem applies to any species that forages only intermittently at specific sites, including some blackbirds (Gartshore et al. 1979) and shorebirds (Rundle 1982), but probably not to most small landbirds that feed more or less continuously.

Mist-netting or trapping may introduce additional biases. For example, birds caught in nets may not be assignable to a specific habitat or foraging zone (i.e., they may be transients in the area of capture), and age and sex classes may not be sampled equivalently. In addition, birds held alive in nets or holding cages for varying periods of time continue to digest their food and may increase the bias associated with differential digestibility of food items (discussed below).

Sample sizes necessary for any particular study may be difficult to determine a priori, because they depend to a large extent on the variability in diet among individuals. Assessing the adequacy of collected samples is discussed by Sherry (1984), based on the methods of Hurtubia (1973). In general, a cumulative plot of diet composition may be constructed by adding the diets of successive individuals until an asymptote is attained. This point represents the number of stomachs beyond which little additional information about diet composition is obtained. In several studies, 10 or fewer stomachs were adequate for assessing species-specific diets at particular sites within a collecting period (Wiens and Rotenberry 1979; Rosenberg et al. 1982; Sherry 1984; Rosenberg and Cooper, unpubl. data). Larger samples may be necessary for studies of individual, temporal, or geographical variation in diet within species. Sample sizes also influence later statistical procedures, as discussed by Duffy and Jackson (1986); for example, parametric tests usually require larger samples than do nonparametric tests.

Differential digestion rates of dietary items impose the largest potential bias in any study of gut contents and may influence every phase of the study. Koersveld (1950) showed that post-mortem digestion may occur in birds. However, the disappearance of food in birds left at 21°C for 3 days hardly approximates potential problems encountered under normal field conditions. Some researchers have injected formalin (usually 1.0 cc at 10% strength) into the stomach immediately upon death to stop digestion. Dillery (1965) compared 80 stomachs of Savannah Sparrows (*Passerculus sandwichensis*) injected with formalin with 47 (presumably) uninjected samples from the U.S. Fish and Wildlife Service files. More individual arthropods were identifiable in the injected stomachs (13.75/bird vs. 5.13). In addition, soft-bodied Homoptera (e.g., aphids) were under-represented in the uninjected samples (9%

vs. 30% of all items), whereas larval Lepidoptera were over-represented (13% vs. 4%).

Sherry (1984) found an average of 15–30 identifiable arthropods/stomach in a variety of neotropical flycatchers (Tyrannidae). Although these stomachs were not injected with formalin, they were removed immediately and placed in 70% ethanol. An average of 10–13 arthropods/stomach was identified in two species of flycatcher (*Empidonax* spp.) in Louisiana (Rosenberg and E. Robinson, unpubl. data). No attempt was made to stop post-mortem digestion; specimens were usually frozen within 1–2 hours after collection and stomachs were removed to 70% ethanol at the time of specimen preparation. Under similar conditions (but without freezing), an average of 11–14 arthropods/stomach was identifiable in two species of antbird (*Myrmotherula* spp.), and 8–14/stomach in two woodcreepers (*Xiphorhynchus* spp.; Rosenberg and A. Chapman, unpubl. data). Clearly, the necessity for and consequences of not injecting bird stomachs with formalin requires further study.

Differential digestion rates can also bias a sample before a bird is collected. Experiments with bird digestion (Stevenson 1933) showed that wild birds varied greatly in the fullness of their stomachs, and that juveniles of several species contained more food than adults. Stevenson (1933) also determined the time of passage through a bird's gut to average 2.5 hr for a variety of foods including insects, seeds, and fruit pulp. Other studies report much shorter digestion times, with an extreme rate of disappearance of 5 min in the Savannah Sparrow (Dillery 1965). Swanson and Bartonek (1970) found that soft-bodied insects may be gone from gizzards within 5 min, whereas hard seeds may persist for several days. These conflicting results are discussed by Custer and Pitelka (1974), who also provide correction factors for differential digestion rates in the Snow Bunting (*Plectrophenax nivalis*). Similar corrections were made by Coleman (1974) after determining what percentage of various foods persisted in European Starling (*Sturnus vulgaris*) gizzards after 2 hr. A method for determining correction factors for insectivorous birds is given by Mook and Marshall (1965). Following those methods, Cooper (unpubl. data) found that second and third instar gypsy moth (*Lymantria dispar*) larvae were completely digested in less than half the time it took birds to digest fourth and fifth instars. In addition, specialized seed dispersers were shown to have higher gut-passage rates than nonfrugivores of equal body weight (Herrera 1984b). In short, the potential biases associated with rates of digestion are poorly understood and point to a continued need for innovative experimentation with live birds (see also Gartshore et al. 1979, Rundle 1982).

The extent to which stomachs from mist-netted birds may differ from those of shot individuals was addressed for two groups of neotropical species (Rosenberg and A. Chapman, unpubl. data). Among two species of antwren (*Myrmotherula* spp.) and two woodcreepers (*Xiphorhynchus* spp.), the number of identifiable arthropods in shot vs. netted samples was similar (12 vs. 9 and 10 vs. 9, respectively), as was the total number of arthropod orders represented. In the antwrens, more beetles and fewer orthopterans were evident in shot samples of *M. leucophthalma*, whereas the opposite was true in *M. haematonota*. In the woodcreepers, beetles and orthopterans were more prevalent in the netted samples of both species, spiders were proportionally more evident in shot *X. spixii* but not in *X. guttatus*, and Lepidopteran larvae were much more common in shot individuals of both. Thus, these preliminary results do not clearly indicate a consistent bias in netted vs. shot samples, and any potential biases can be lessened by minimizing the time that a bird remains alive in the net.

In species with a well-developed crop, the crop contents are thought to be the most unbiased representation of a bird's diet (Hartley 1948). In larger birds, esophageal contents can be compared with stomach contents (e.g., Goss-Custard 1969, Swanson and Bartonek 1970), with the former usually considered preferable. Rundle (1982) argued strongly in favor of esophageal analysis for studies of shorebird diets, providing examples of marked differences from analyses of gizzard contents alone. Although in most small passerines the esophagus is usually empty and cannot be used to calibrate stomach contents (Custer and Pitelka 1974), careful attention to collecting only actively feeding birds may ensure full gullets. For example, Gartshore et al. (1979) found that most foods persisted for up to 20 min in the gullets of Red-winged Blackbirds (*Agelaius phoeniceus*) feeding under natural conditions. In addition, the gullets of many granivorous species often contain large samples of seeds recently eaten (Newton 1967, Payne 1980, Zann et al. 1974).

In most studies, gut contents are preserved in either formalin or alcohol. In general, formalin is better for preserving flesh (including the stomach itself), but may disolve bone or distort insect or vegetation parts (Duffy and Jackson 1986). Ethyl alcohol (70 to 95%) is preferred by entomologists (Borror et al. 1981) and is probably adequate for most studies of insectivorous birds. Well-preserved gut contents may be stored for long periods. Giuntoli and Mewaldt (1978) successfully examined stomachs of Clark's Nut-

crackers (*Nucifraga columbiana*) after storage in formalin for up to 15 years. Thus, samples may be accumulated and stored at central depositories, as discussed below.

Forced regurgitation and flushing

In many cases collecting birds for stomach analysis may be undesirable because of harm to local populations, ethical considerations, or inability to obtain permits. Several approaches allow partial sampling of gut contents via regurgitation or otherwise flushing the digestive tract of live birds. These vary in efficiency and in their effects on individual birds. The various biases associated with differential digestion and sampling concerns, discussed for stomach contents, are equally applicable to any technique involving partially digested or fragmented food samples.

The most common method of forced regurgitation uses chemical emetics. Antimony potassium tartrate (tartar emetic) seems to be the most widely used (Prys-Jones et al. 1974, Zach and Falls 1976a, Robinson and Holmes 1982, Gavett and Wakeley 1986), performing best in comparative trials (Lederer and Crane 1978). Dosages vary but are usually administered orally via a syringe and flexible plastic tubing coated with vaseline. Tomback (1975) found that a 1.5% tartar emetic solution rather than a 1% solution shortened the response period of several species from about 25 min to an average of 10 min, without harming the birds. Other researchers (Prys-Jones et al. 1974, Zach and Falls 1976a, Robinson and Holmes 1982) observed that most insectivores regurgitated samples within 2–3 min using 1% solution. Prys-Jones et al. (1974) found that only 50–64% of the granivores fed tartar emetic regurgitated samples, hypothesizing that more muscular gizzards act as a barrier to regurgitation.

Biases associated with emetics have been examined in several studies. Using captive Ovenbirds (*Seiurus aurocapillus*), Zach and Falls (1976a) found that the action of tartar emetic was independent of the type of prey eaten. Although regurgitation occurred in almost all birds tested, it was not always complete. Thus, no qualitative bias was found, but the material regurgitated did not reflect the quantity of food in the stomach. Gavett and Wakeley (1986) tested the efficiency of emetics in House Sparrows (*Passer domesticus*) by collecting stomachs from a subset of the sampled birds. An average of 58% of the total contents of each stomach was obtained by regurgitation. Although food categories were missing from individual stomachs, the overall emetic sample gave an accurate representation of the total diet in this species.

Mortality caused by emetics can be high and may depend on the species involved, dosage, and stressful effects such as handling. Zach and Falls (1976a) observed 50% mortality in newly caught Ovenbirds fed emetics, and 12.5% mortality in individuals already acclimated to captivity. Successive applications of emetic within a relatively short time invariably resulted in death. Lederer and Crane (1978) observed 20% mortality in wild-caught House Sparrows. Although Prys-Jones et al. (1974) found no difference in survival between treated and control House Sparrows, individuals that regurgitated were more likely to be sighted later than those that did not regurgitate. Emetics also were tried unsuccessfully on Australian honeyeaters (Ford et al. 1982) and various seed-eating species (Zann et al. 1984); in these studies no gut samples were obtained and mortality was often high.

Forced regurgitation also has been used without emetics. Lukewarm water is pumped directly into the stomach using a syringe and thin plastic tube until the stomach and esophageal contents are regurgitated. Brensing (1977) sampled over 2100 migrant passerines of 35 species and reported no loss of weight in birds recaptured after sampling. This technique was also used successfully on 157 Australian passerines (Ford et al. 1982) and on many species of small passerines on migration along the Louisiana Gulf Coast (Franz Bairlein, pers. comm.), with virtually no mortality. Ford et al. (1982) successfully obtained some gut contents from all individuals sampled (13 needed to be flushed twice) and reported equal rates of recapture or resighting of flushed and nonflushed birds.

A variation is flushing the entire digestive tract with warm saline solution (Moody 1970), which was used by Laursen (1978) to study migrant passerines in Europe. Of 396 birds sampled, 14 (3.6%) died during flushing; comparison of the remaining stomach contents with the flushed samples in these individuals indicated an average efficiency of 52%. Jordano and Herrera (1981) used this technique to document the frugivorous diet of the Blackcap (*Sylvia atricapillus*) in Spain. The use of stomach pumps is recommended by Duffy and Jackson (1986) for studies of seabirds and is discussed in relation to dietary studies of fish by Hyslop (1980). Apparently, the efficiency of this technique decreases with the size of the animal sampled. Overall, stomach pumping and flushing hold great promise for many studies and would seem highly preferable to the use of emetics.

Several similar techniques were developed specifically for use on seed-eating birds. Payne (1980) inserted a plastic tube into the crops of

Red-billed Firefinches (*Lagonosticta senegala*) and sucked small seeds into the tube, rather than flushing them out with water. He also found that the crop contents of nestlings could be observed directly through the translucent skin of the neck. Newton (1967) successfully used this technique for nestlings and some adults of several British finch species and was able to distinguish some invertebrate foods as well as seeds. Alternatively, the crop can be manipulated to facilitate seed removal (Zann et al. 1984). Samples obtained in this way compared well with total crop contents of collected birds and had no noticeable effect on mortality or recapture rates.

Fecal samples

Fecal samples may be collected from any species that can be captured alive, and such sampling can be integrated easily into any study that uses mist nets. Furthermore, droppings may be collected year-round from birds of any age or any reproductive state, and repeated sampling from known individuals is possible (see Ralph et al. 1985, for details). This or similar techniques have been used successfully in studies of flycatchers (Davies 1977b), wagtails (Davies 1976, 1977a), aerial insectivores (Bryant 1973, Waugh 1979, Waugh and Hails 1983), magpies (Tatner 1983), dippers (Ormerod 1985), and Hawaiian passerines (Ralph et al. 1985). Large numbers of samples also can be obtained at communal roosts, at feeding sites, and under nests.

A drawback of fecal analysis is the necessarily fragmented and highly digested state of the samples. For this reason, biases related to differential digestibility and rates of passage may be more serious than for stomach or crop samples. Nevertheless, a close correspondence between fecal and stomach samples has been shown, and the range of food items encountered by Ralph et al. (1985) did not reflect a bias against small or soft-bodied prey items.

Ligatures

Food brought by adults to nestlings can be assessed by placing constrictive ligatures around the nestlings' necks to prevent their swallowing. Ligatures can be made of copper wire (Johnson et al. 1980, Johnson and Best 1982), plastic-coated wire (Owen 1956), metal bands (Kluyver 1961), pipe cleaners (Orians 1966, Willson 1966, Moore 1983), or thread (Pinkowski 1978, Bryant and Westerterp 1983). Detailed diagrams and description of ligature application appear in Johnson et al. (1980).

An advantage of ligatures is that arthropod prey are usually kept intact, so that problems of extreme fragmentation and differential digestibility associated with other methods are minimized. Also, repeated samples can be obtained from individual nestlings.

Ligatures also have problems. Few data are collected per unit time compared with stomach contents analysis. If coupled with direct observation the technique becomes more costly. Although nestlings are disturbed temporarily, feeding behavior and even survival can be affected. Orians (1966) and Willson (1966) found that pipe cleaners caused some mortality of nestling Yellow-headed Blackbirds (*Xanthocephalus xanthocephalus*) less than 3 days old. Johnson et al. (1980), however, successfully used light wire ligatures on nestling Gray Catbirds (*Dumatella carolinensis*) as young as 2 hours old. Young Black-capped Chickadees (*Parus atricapillus*) would not gape for food when wearing metal collars (Kluyver 1961). Handling older nestlings may cause them to leave the nest prematurely (Johnson and Best 1982). If feeding behavior is affected differentially among species, then between-species comparisons may be biased (Orians and Horn 1969). Another problem is that adult birds may remove prey from the nestlings' mouths and eat it (Robertson 1973). Disgorging of food can be a problem as well (Orians 1966, Johnson et al. 1980). Also, the technique can be biased against smaller food items, which may slip past the ligatures (Orians 1966, Walsh 1978).

Many biases associated with ligatures were quantitatively assessed by Johnson et al. (1980), who found that ligatured nestling catbirds gaped less intensely, gasped and disgorged food more often, and were fed less often than nestlings without ligatures. More food was removed by parents of ligatured nestlings than by those of unligatured nestlings. Johnson et al. also observed that, because of disgorging of food and weaker gaping, the average volume of food collected per ligatured nestling was much less than that delivered per unligatured nestling. Larger food items were disgorged more readily, so estimates of prey size eaten were also biased. Only dietary composition (taxonomic) was unaffected, although given the above problems, that should also be examined more thoroughly. They recommended directly observing collared nestlings from a blind and immediately collecting prey after each parental visit.

Moore (1983) examined some of the same sources of error in a study of nestling European Starlings, and concluded that the procedure yielded reliable estimates of diet. Also, Knapton (1980) compared the food removed from ligatured, nestling Brown-headed Cowbirds (*Molothrus ater*) with the food delivered by adult Clay-colored Sparrows (*Spizella pallida*), which were recorded on film, and found little difference. Perhaps these biases are species-specific. Neverthe-

less, we recommend that ligatures not be used alone, and that possible biases be assessed and corrected by simultaneously using direct observation, photography, or video recording.

Pellets

Raptors periodically regurgitate pellets of nondigestible matter (hair, bone, feathers, sclerotized insect parts), which can be collected at nest or roost sites and frozen for analysis at a later date (Errington 1930, 1932). After drying, pellets are picked apart and sorted by hand, until all identifiable prey parts are accounted for. Pellets may be soaked in water (Short and Drew 1962, but see Holt et al. 1987) or boiled in NaOH solution (Schueler 1972, Longland 1985) to facilitate separation of bone from other matter.

Major advantages of this method are simplicity and accuracy without handling or otherwise disturbing birds. Pellet analysis has been facilitated by published dichotomous keys to skulls, dentition (e.g., Driver 1949), and hair (e.g., Mathiak 1938, Williams 1938) of mammalian prey commonly found in pellets. Techniques for differentiating pellets of some species have also been developed (Holt et al. 1987), although more work is needed in this area. Some potential biases are associated with this method, however. First, different raptors eat and digest bone to different degrees. Owls swallow entire many small- and medium-sized prey. Larger prey are torn apart and consumed. Hawks often pluck feathers and fur away before tearing off and swallowing small parts of their prey. They also digest bone more readily than owls do (Craighead and Craighead 1956). Thus, a greater amount of bone is found in owl than in hawk pellets, making dietary comparisons between these groups difficult. For example, Craighead and Craighead (1956) found that approximately 69% of the rodents fed to a captive Northern Harrier (*Circus cyaneus*) were evident in pellets, whereas nearly 100% were found in Short-eared Owl (*Asio flammeus*) pellets.

Second, a single prey item, especially if large, may be represented in more than one pellet and may be egested at more than one location (Craighead and Craighead 1956, Smith and Richmond 1972, Lowe 1980). Short and Drew (1962) found that 100 g or more of mice consumed overnight often produced two or more pellets. Leg bands and stained bones of rodents fed to captive Tawny Owls (*Strix aluco*) were retained for up to two days (Lowe 1980). Smith and Richmond (1972) induced pellet egestion by allowing a captive Common Barn Owl (*Tyto alba*) to see a live rodent. They determined that pellets are not egested at a fixed interval after taking a meal; rather, the interval depends in part upon the quantity of food consumed, time of feeding, and availability of a subsequent meal. Individual pellets should, therefore, not be treated as the sampling unit. Instead, all pellets collected in a particular location or several locations for a particular bird during a specified time interval should be the sampling unit (e.g., Marti 1974, Lowe 1980).

Third, remains of some animals survive the pellet-forming process better than others. In a study of captive Short-eared Owls, Short and Drew (1962) found that *Microtus* formed pellets that held together better than *Peromyscus*; only 25% of *Peromyscus* found in pellets had the proper proportions of skulls, innominates, and mandibles present. Lowe (1980) was unable to account for 21% of the rodents fed to Tawny Owls. The percentage varied with season and age of prey. The problem is likely to be more acute for falcons and accipiters, which often eat large percentages of arthropods (e.g., Smith et al. 1972) and birds (e.g., Cavé 1968).

Little research has been done comparing pellet analysis and other methods of diet analysis of non-captive raptors. Smith et al. (1972) found that not all prey fed to nestling kestrels was represented in pellets. Collopy (1983), who compared pellet analysis and remains in nests with direct observation of prey brought to Golden Eagle (*Aquila chrysaetos*) nestlings, found little difference between the two methods in estimated prey species composition, either by percent frequency or percent biomass. However, collections of pellets and remains consistently underestimated daily capture rates. He suggested, and we agree, that direct observation, which is costly and potentially disruptive to nesting birds, be used periodically to correct for prey biomass unaccounted for in collections.

The fact that pellets of different raptor species are not equally satisfactory for analysis does not alter the fact that they provide useful data. Analysis of a sufficient number of pellets will probably show feeding trends with less expense and disturbance to birds than any other method.

Direct observation

We can study the diets of many species by direct observation to obtain information on consumption rates, food handling, and diet selectivity not detectable from gut contents alone. These studies are easiest for frugivores and nectarivores, for which we can identify the species of food plant, or at least describe the size, shape, and color of the fruit or flowers (e.g., Leck 1971, Snow 1981, Moermond and Denslow 1985, Stiles 1985c). Direct observations may not elucidate the proportions of animal foods in the diets of these species, however. For example, the high frequency of arthropods evident in the stomachs

of most hummingbirds (Remsen et al. 1986) was not apparent from observations of visitation to various flowers.

Price (1987) observed the seeds eaten by Darwin's Finches (*Geospiza* spp.) and successfully related diet selection to individual morphology and varying ecological conditions. Newton (1967) reported that the foods of cardueline finches that fed above the ground on the seedheads of various plants could be easily quantified, whereas direct observation of the seeds eaten by ground-foragers was not possible.

For insectivorous birds, identification of prey items in the field is much more difficult. Whereas large or common prey may be easy to distinguish, many inconspicuous foods will be overlooked, and such observations by themselves may be highly biased (e.g., Rundle 1982). For example, using direct observation, Cooper (unpubl.) concluded that Scarlet Tanagers (*Piranga olivacea*) preyed almost exclusively on larval and adult Lepidoptera, but stomach contents showed that Lepidoptera comprised only 20–40% of the diet of adult birds. Robinson and Holmes (1982) supplemented gut samples (using emetics) with direct observations of prey captures for 11 species of forest insectivores. Prey were identifiable in from 1.1% (Least Flycatcher, *Empidonax minimus*) to 37.9% (Solitary Vireo, *Vireo solitarius*) of the observed foraging maneuvers. Prey size often may be estimated, even when prey type is unknown, by comparison with bill or head length, although this method has several biases (Bryan 1985, Goss-Custard et al. 1987).

Observations also may be made at nests to determine nestling diets, feeding rates, and other aspects of parental behavior. This technique is most often used for large species such as raptors (e.g., Collopy 1983) but has also been used successfully for passerines (Tinbergen 1960, Sealy 1980, Biermann and Sealy 1982). These observations are often greatly facilitated by the use of blinds, high-powered telescopes, or photography.

Photography

Various photographic devices have been used to record prey brought to nests. A major advantage is that film can be reviewed later, often allowing identification of prey type and size. Probably the most popular apparatus is the 8- or 16-mm movie camera fitted to a nestbox (Royama 1959). Upon entering the nestbox, adult birds trip a switch and a single-frame picture is taken of the bird's head and bill contents. Often a watch and metric ruler are fastened next to the entrance hole, so that the time of feeding and prey size can be determined (Royama 1970, Dahlsten and Copper 1979, Minot 1981). A major advantage is that an observer need not be present, because movie cameras may be operated automatically by a car battery. Because cameras are expensive, the number of nests to be photographed will usually exceed the number of cameras available. This problem can be circumvented by designing nestboxes so that the camera can be fitted to each one (Royama 1970), or by moving nests to a special box fitted with a camera (Dahlsten and Copper 1979).

Video recorders and 35-mm cameras fitted with telephoto lenses have been used to record prey brought to nestlings of open-nesting species. Knapton (1980) and Meunier and Bedard (1984) placed a stick next to the nest where the adults perched to feed young and were easily photographed. A disadvantage of hand-operated cameras is that an observer must be present, usually in a nearby blind, and must also be a skilled photographer. Video recorders will probably be used with increasing frequency in diet studies, because they record continuously.

Diet Analysis

Diet analysis generally consists of (1) sorting and identifying food items and (2) presenting the results in terms of occurrence, frequency, volumetric, or gravimetric measures (reviewed by Hartley 1948, Hyslop 1980, and Duffy and Jackson 1986). Most researchers recognize the need for presenting diet data in more than one form to minimize biased interpretations (e.g., Otvos and Stark 1985).

Sorting and identification

Little literature exists for sorting and identifying fragmented gut contents, and methods are rarely published in enough detail to be useful to others (but see Calver and Wooller 1982). In general, contents are examined under a dissecting microscope, preferably one with variable power (up to 30×) and fitted with an ocular micrometer. The procedure is more or less a game of matching similar parts and determining the minimum number of prey ingested by counting heads, mandibles, wings, legs, or other parts of known number in the intact state. Seeds are often encountered whole; however, other vegetative matter (e.g., fruit pulp) usually occurs in a form that prevents the enumeration of individual foods. The amount of grit present in a sample may be determined by "ashing" the contents at extremely high temperature (540°F), after identification and weighing (Shoemaker and Rogers 1980).

The ability to detect the full range of dietary items present rests on learning the specific parts, however tiny, that survive digestion. We believe such clues exist for virtually every type of solid

food a bird may eat. Ralph et al. (1985) and Tatner (1983) listed commonly encountered fragments representing a variety of arthropod taxa, accompanied by photographs or sketches of the most distinctive parts. Diagnostic structures, at least to the familial level, appear to be remarkably invariate across diverse geographic regions. Accordingly, we found these lists from Hawaiian Islands and Great Britain valuable in identifying stomach samples from the southeastern United States and Amazonian rainforests.

The identification of arthropods and seeds is greatly facilitated by a reference collection of intact food items and of fragmented parts (e.g., mandibles, spider fangs) taken from known, intact specimens and mounted on clear microscope slides for easy comparison. Such reference collections permit determination of original size or weight of ingested foods from identified fragments. Calver and Wooller (1982) provided detailed equations for estimating total length from the size of diagnostic parts (e.g., head width, elytra length) for various families of Diptera, Coleoptera, and Hymenoptera.

Collaboration with entomologists or botanists is also recommended, although even experts may not be familiar with fragmented specimens. In addition, a technician without formal entomological or botanical background may be easily trained to recognize and sort diagnostic parts in fragmented samples (Ralph et al. 1985, Rosenberg pers. obs.). A primer on entomological terms commonly encountered in analysis of bird diets is offered by Calver and Wooller (1982).

In most studies, arthropod remains are identified only to family (sometimes only to order). Levels of prey identification affect the subsequent categories used in dietary comparisons, as discussed by Greene and Jaksic (1983) and Cooper et al. (this volume). In general, more inclusive categories tend to overemphasize the similarities among samples and underestimate diet diversities. Rotenberry (1980a) used the criterion that any taxonomic category represented in at least 5% of his samples would be included in further analyses, with rare taxa not meeting this criterion lumped into the next-most-inclusive category. Prey categories may be combined on the basis of ecological characteristics (e.g., phytophagous or predaceous; Robinson and Holmes 1982), or according to their modes of escape (e.g., flying, jumping, hiding), activity patterns and typical locations (Cooper et al., this volume), or, in the terminology of early diet researchers (e.g., MacAtee 1912), "harmful" vs. "beneficial." Sherry (1984) combined all morphologically identical specimens in his diet samples into "morphospecies" that were assumed to be encountered in patches by the foraging birds. Knowledge of the natural history of the arthropod (and plant) foods, as well as of the birds, will aid in the meaningful assignment of diet categories.

Percent occurrence

Occurrence usually refers to the number of samples in which a particular food type appears, although it is sometimes used as a synonym of frequency. Percent occurrence is the simplest and crudest measure of diet. Its primary advantage is that virtually all food types can be counted, even if individual items ingested cannot be quantified. For example, the presence of certain fruits or wing scales of adult lepidopterans may be detected by a distinctive color, and their occurrence is therefore easily determined. Hyslop (1980) discussed the application of subjective dominance rankings that allow the addition of relative importance values to occurrence measures. In general, species-level comparisons using percent occurrence tend to emphasize similarities among samples, whereas frequency and volume estimates tend to emphasize differences (Hartley 1948).

Frequency

Frequency is usually applied to the enumeration of individual food items. Ideally, the original diet can be "reconstructed" from these identified parts; however, some food types, such as fruit or green vegetation, do not occur in a form that can be counted. Individual samples are often pooled to create a single sample for a particular species, season, or geographic region. In these, the differences between frequency and occurrence measures depend on the patchiness of the foods encountered in nature and, therefore, in the individual diets (Hartley 1948). If the individual samples are considered separately, however, the average frequency per sample with its associated variance would reflect this patchiness. Sherry (1984) discussed the determination of patchiness of food items and its use as an independent characteristic of a species' diet and contrasted the use of pooled vs. individual samples in dietary analyses. In general, we recommend the use of per-sample measures to express frequencies or volume estimates.

The biases associated with differential digestion or passage through the gut are reflected in the differences between frequency and bulk (e.g., volumetric) estimates of diet (Hartley 1948, Hyslop 1980). In general, frequency measures tend to exaggerate the importance of small items and those whose parts persist longest in the digestive tract (MacAtee 1912). For example, a stomach that contains 20 small ants and one large cicada

would indicate a diet of mostly ants in a frequency analysis but mostly cicadas in a volumetric analysis. The ants may better reflect the foraging effort and time of the bird, but the cicada may represent the bulk of the energy gained from that collection of food. Correction factors have been applied by Custer and Pitelka (1974) and others to account for these different rates of passage.

Percent volume and weight

The volume of a food type may be estimated as it appears in the sample and then expressed as a percentage of the total volume of the contents or the capacity of the stomach. This procedure allows almost all food types to be considered, including those that cannot be enumerated individually. Therefore, this may be the only way to describe diets of largely vegetarian species. In contrast with frequency measures, volumetric estimates tend to give greater importance to large, mostly undigested food items (Hartley 1948, Duffy and Jackson 1986). MacAtee (1912) considered this the best method to represent the "economic importance" of a bird species (i.e., its potential impact on the range of prey it consumed). He also noted that large samples minimized the potential bias of essentially ignoring the tiny, long-persisting fragments that would be counted in a frequency analysis.

Volumes also can be estimated by reconstructing the original diet based on the frequency and size of various food types (Martin et al. 1946, Hartley 1948). In this way, all food items are counted, but the largest items (at the time of ingestion) are given greatest importance. The determination of original volumes depends on the use of a reference collection of whole specimens or on various correction factors, as discussed by Hyslop (1980) and Calver and Wooller (1982).

Estimates of weight or biomass may be derived in the same ways as for volumes; however, these are often more tedious and time-consuming (Hartley 1948, Duffy and Jackson 1986). The use of wet- vs. dry-weight measures is discussed by Hyslop (1980). Dry weights of arthropods may be estimated from specimens of known or estimated length, using regression equations in Rogers et al. (1976, 1977) and Beaver and Baldwin (1975). Knowledge of original weights is necessary for calorimetric determinations. Estimates of the energy content of various foods are found in Golley (1961), Thompson and Grant (1968), Bryant (1973), Ricklefs (1974a), Norberg (1978), and Bell (this volume). Using these estimates, Calver and Wooller (1982) derived a general equation for determining energy content directly from prey length. Rosenberg et al. (1982) used a similar procedure to estimate the dietary requirements of a bird assemblage preying on cicadas. These measures should be used with caution, however, because of the potential to overestimate the nutritional value of large or long-persisting food types (Hyslop 1980).

Diet Information Sources

Here, we describe two important sources of raw data on the diets of North American and many Neotropical species. The first is the large collection of stomach samples compiled by the U.S. Biological Survey, representing over 250,000 individual birds of over 400 species (see MacAtee 1933). Stomach contents were meticulously identified by expert entomologists and botanists (often to species level). These data appear in various forms in numerous publications by W. L. MacAtee, F. E. Beal, and others and were summarized for most species by Martin et al. (1951a). The raw data are stored on cards filed at the Patuxent Wildlife Research Center of the U.S. Fish and Wildlife Service in Laurel, Maryland. Each card represents a single stomach sample and contains information on the bird's sex, location, habitat, time of day, and date of collection. Contents are listed individually, along with the relative volumes of each food type in relation to the total volume of the contents, and the relative volumes of total plant and animal matter.

This tremendous source of information has barely been exploited by modern ornithologists. Wheelright (1986b) used these data to describe seasonal and geographic variation in the American Robin and urged their wider application. Although the samples for most species are from wide geographic regions and dispersed over many years of collection, precluding many community-level analyses, their potential for studies of ecomorphology, predator–prey relationships, plant–animal interactions, and seasonal variation is great. For example, Hespenheide (1971) reanalyzed the published data for several flycatcher species to test the theoretical relationships between predator and prey sizes.

The second source is the collection of unanalyzed stomach contents at the Louisiana State University Museum of Natural Science (LSUMNS). In most cases these are whole stomachs, taken from birds during routine specimen preparation, and preserved in 70% ethanol. All samples are labeled to correspond with skin or skeleton specimens deposited at LSUMZ and accompanied by complete data on location, habitat, age, sex, reproductive condition, fat, and molt. The ability to measure the morphological features of birds from which diet samples were taken should aid in studies of ecomorphology and individual variation (e.g., Herrera 1978b).

This collection has a strong Neotropical representation, including over 2500 samples from ca. 700 species, mostly from the Andes and low-

TABLE 1. COMPARISON OF COMMON METHODS USED TO OBTAIN AVIAN DIET SAMPLES

Method	Advantages	Disadvantages	Example of use
Direct examination of collected birds	Whole stomachs collected; if shot, then exact bird desired can be obtained.	Birds are killed; multiple samples from one bird impossible.	Rotenberry (1980a), Sherry (1984)
Chemical emetics	Birds not killed directly.	Mortality may still be substantial; multiple samples from one bird often results in mortality; birds must be captured; partial samples obtained; unsuitable for some species.	Zach and Falls (1976a), Robinson and Holmes (1982), Gavett and Wakely (1986)
Stomach pumping	Birds not killed.	Birds must be captured; partial samples obtained.	Moody (1970), Brensing (1977)
Fecal samples	Birds disturbed minimally; samples easily obtained.	Birds usually must be captured; samples highly fragmented; samples must be treated before analysis.	Ralph et al. (1985)
Ligatures	Arthropod prey usually intact; can be effective when combined with direct observation.	Restricted to nestlings; feeding behavior and survival can be affected; estimates of prey size can be biased.	Johnson et al. (1980)
Pellets	Birds not disturbed; samples easily obtained; keys to mammal skulls and hair available.	Restricted to pellet-forming species; may be biased by prey type, size.	Errington (1930)
Direct observation (adult birds)	Birds not disturbed; foraging behaviors that resulted in prey capture are observed.	Difficult for insectivorous birds; observations biased towards large, conspicuous prey.	Robinson and Holmes (1982), Price (1987)
Direct observation (nestlings)	Birds not disturbed; can be effective when used in conjunction with ligatures.	Time consuming, labor intensive; biased as above.	Tinbergen (1960), Johnson et al. (1980)
Photography	Birds not disturbed; automatic movie cameras provide many samples for little effort.	Restricted to nestlings; Equipment relatively expensive; hand operated cameras time consuming, labor intensive.	Royama (1959, 1970), Dahlston and Copper (1979)

land rainforests of Peru and Bolivia. These include many poorly known species for which little basic natural history information exists. Sample sizes for some species are large enough to permit geographic and guild-level analyses. The LSUMNS collection also contains about 1500 stomach samples from common birds in Louisiana, as well as smaller collections from other regions. Research use of any materials is welcomed; inquiries should be directed to: Curator of Birds, Museum of Natural Science, Louisiana State University.

RECOMMENDATIONS AND CONCLUSIONS

With the broad range of techniques now available (Table 1), direct examination of avian diets is possible in nearly any study. For many species that cannot be captured alive, collection of stomach or esophageal contents remains the only means of diet assessment. When collection is necessary, care is needed to maximize sampling efficiency, taking only actively foraging individuals from known habitats or foraging sites, and ensuring adequate sample sizes. When capture is possible, we recommend the use of flushing techniques to force regurgitation of gut contents, avoiding emetics. Fecal samples are probably the easiest to obtain but present added difficulties in analysis and interpretation. When other techniques are unavailable, routine collection of fecal samples will give an adequate representation of many species' diets. For any species that regularly regurgitates pellets, large samples of prey remains can be collected and may give an accurate estimate of diet.

For studies of the diet of nestling birds, several additional techniques are available, including ligatures, photography, and direct observation of the nest. Direct observation of foraging birds may be a sufficient means of assessing diet in some species, particularly in specialized nectarivores or frugivores. Observations of foods eaten can supplement any of the techniques discussed and may aid in the minimization of certain biases associated with highly digested gut contents.

Biases caused by differential rates of passage and digestibility remain poorly documented and understood. Continued experimentation with live

birds is needed to determine the advisability or consequences of various collecting, preserving, and analytical procedures. We also urge the publication of additional lists, descriptions, sketches, or photographs that can aid in the identification of fragmented diet samples. Expanded use of reference collections with additional calculations of prey length and weight from fragment size is also recommended.

Each of the several methods of presenting diet data has its advantages and drawbacks. Therefore, more than one method should be presented whenever possible, including at least one that represents occurrence and one that represents frequency or relative volume. Although pooling results may be desirable in cases with small sample sizes or when only population averages are needed, we recommend the use of per-sample measures with their associated variances to characterize species' diets.

We urge that gut contents be routinely preserved from specimens collected for any reason; with limitations placed on present and future collection of birds, the maximization of information from each specimen is highly desirable. We also urge the expanded application of diet analysis techniques to a wide range of ecological pursuits. Our knowledge of avian food habits lags far behind our knowledge of habitat use, foraging behavior, and morphology. In most cases, gathering diet data by any means available is preferable to ignorance. We think that many of the biases and difficulties will be alleviated when more careful attention is paid to sampling design, prey identification, and overall foraging ecology.

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