

## STABLE-ISOTOPE ANALYSIS OF CANVASBACK WINTER DIET IN UPPER CHESAPEAKE BAY

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**ABSTRACT.**—A major decline in submerged aquatic vegetation (SAV) in Chesapeake Bay has altered the diet of wintering Canvasbacks (*Aythya valisineria*) from historically plant to a combination of benthic animal foods, especially the ubiquitous Baltic clam (*Macoma balthica*), supplemented with anthropogenic corn (*Zea mays*). Because the isotopic signature of corn is readily discriminated from bay benthos, but not SAV, we used stable-isotope methodology to investigate the corn–SAV component of the winter diet of Canvasbacks. Feeding trials with penned Canvasbacks were conducted to establish turnover rates and fractionation end-point loci of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures of whole blood for individual ducks fed *ad libitum* diets of (1) Baltic clams, (2) Baltic clams and corn, and (3) tubers of wild celery (*Vallisneria americana*). Turnover time constants averaged 4.5 weeks, indicating that signatures of wild ducks would be representative of bay diets by late February. Isotopic signatures of wild Canvasbacks sampled in February fell on a continuum between end-point loci for the Baltic clam and the combination Baltic clam and corn diet. Although that finding verifies a clear dependence on corn–SAV for wintering Canvasbacks, it also reveals that not enough corn–SAV is available to establish *ad libitum* consumption for the 15,000+ Canvasbacks wintering in the upper bay. On the basis of mean  $\delta^{13}\text{C}$  signature of bay Canvasbacks ( $n = 59$ ) and ingestion rates from feeding trials, we estimated that 258 kg corn per day would account for the observed  $\delta^{13}\text{C}$  enrichment and supply 18% of daily energetic needs for 15,000 Canvasbacks. That level of corn availability is so realistic that we conclude that SAV is likely of little dietary importance to Canvasbacks in that portion of the bay. Received 17 February 2000, accepted 16 April 2001.

CHESAPEAKE BAY has long been recognized as a major wintering area for migratory waterfowl (Anatidae), and an exceptional historic wintering area for Canvasbacks (*Aythya valisineria*; Stewart et al. 1958, Bellrose 1976, Haramis 1991, Perry and Deller 1995). In the mid-1950s, winter waterfowl surveys estimated presence of a quarter of a million Canvasbacks on the bay, or ~50% of the continental wintering population. By the late 1980s, that figure had dropped to average ~50,000 or ~20% of the estimated winter population (G. M. Haramis unpubl. data). That decline in use is likely in direct response to habitat degradation, for example, excessive nutrients, sedimentation, turbidity and other contamination of aquatic environments that has resulted in a major decline in foods available. Most notable has been the loss of submerged aquatic vegetation (SAV), especially the energy-rich tubers of wild celery

(*Vallisneria americana*) and sago pondweed (*Potamogeton pectinatus*) that were the historic and preferred food of fall migrant and wintering Canvasbacks. Formerly, Canvasbacks made heavy use of fresh estuarine bays that contained great quantities of wild celery (Wilson 1814, Bent 1923, Stewart 1962, Perry and Uhler 1988). Wilson (1814) recognized the strong tie between Canvasbacks and wild celery and noted “wherever this plant grows in abundance, the Canvasbacks may be expected either to pay occasional visits, or make it their regular residence in winter. . . while in waters unprovided with this nutritive plant, they are altogether unknown.”

Following hurricane Agnes in June 1972, SAV declined sharply in Chesapeake Bay and fluctuates yearly from about 10 to 15% of estimated historic areal coverage as estimated from early summer aerial surveys (Kemp et al. 1983, Orth and Moore 1983, Davis 1985, Orth et al. 1997). The SAV now present in the upper bay is dom-

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inated by seed-bearing species such as widgeon grass (*Ruppia maritima*), horned pondweed (*Zannichellia palustris*), and redhead grass (*Potamogeton perfoliatus*), whereas the tuber-forming wild celery and sago pondweed have declined to be virtually unavailable to an estimated 15 to 20 thousand wintering Canvasbacks. Seed-bearing species tend to be of marginal value to Canvasbacks because they are consumed by a diversity of early migrant waterfowl before Canvasbacks arrive and because they die back in winter leaving few perennial stocks for consumption. By comparison, tubers of wild celery and sago pondweed formerly were accessible all winter (barring ice cover) and especially to Canvasbacks that are specially adapted with their long wedge-shaped bills to retrieved subterranean food items. Loss of much of the tuber-forming SAV has left Canvasbacks with no comparable substitute, except anthropogenic corn (*Zea mays*). Corn is fed off docks and along shorelines in the urbanized upper bay. Importance of corn to Canvasbacks is underscored by the observation that diurnal locations of Canvasback flocks coincide closely with feeding stations. That relationship was discovered during a three-year study that tracked ~80 radio transmitter-marked Canvasbacks each year for a 10 week period to estimate winter survival (Haramis et al. 1993). Loss of SAV has increased dependence of wintering Canvasbacks on low-benefit shellfish, especially the ubiquitous Baltic clam (*Macoma balthica*; Perry and Uhler 1988, Lovvorn 1989). The Baltic clam achieves highest densities in mesohaline habitats of the bay where it ranks first in benthic infaunal biomass (Reinharz and O'Connell 1983). Shifts in Canvasback winter distribution generally reflect a declining use of fresh tidal areas, such as Susquehanna Flats where wild celery was formerly abundant, to brackish-water areas where shellfish are more abundant (Perry and Uhler 1988).

We use stable-isotope methodology to determine relative importance of benthic animal food resources versus corn-SAV in the winter diet of Canvasbacks in upper Chesapeake Bay. The efficacy of that approach lies in the predictably different isotopic signature of corn, a C<sub>4</sub> terrestrial monocot that is enriched in  $\delta^{13}\text{C}$  and depleted in  $\delta^{15}\text{N}$ , from bay benthos that is correspondingly depleted in  $\delta^{13}\text{C}$  and enriched in  $\delta^{15}\text{N}$ . Wild celery and other SAV have isoto-

pic signatures similar to corn (e.g. corn values range from  $-11.1$  to  $-13.2$   $\delta^{13}\text{C}$  [Bender 1968], whereas wild celery tubers acquired from Wisconsin measured  $-14.19$  to  $-14.26$   $\delta^{13}\text{C}$ ).

#### METHODS

*Isotopic signature of Chesapeake Bay benthos.*—In 1993 and again in 1998, we sampled bay benthic organisms to establish isotopic signatures of potential Canvasback food resources. Core samples were taken from mud bottoms at three sites in the Chester River and Eastern Bay region of the upper bay frequented by Canvasbacks. Benthic organisms were obtained by flushing core samples through a 1 mm mesh sieve. Samples were sorted by major taxa, oven dried, and pooled for isotopic analysis.

*Feeding trials with penned Canvasbacks.*—To establish turnover rates and metabolic diet-tissue fractionation values for carbon and nitrogen stable-isotope ratios in whole blood, we conducted 12 week winter (January to April 1993) feeding trials with pen-reared Canvasbacks. Ducks were housed in outside pens exposed to natural light and winter weather conditions. Each pen measured  $9.1 \times 4.6$  m and was supplied with a 4 m diameter swim tank that provided *ad libitum* water. For a year prior to being switched to treatment diets, ducks were stabilized on Mazuri waterfowl maintenance diet (14.0% protein, 3% crude fat, 4.5% fiber, 2.5% added minerals) manufactured by PMI Nutrition International Inc. (P.O. Box 19798, Brentwood, Missouri 63144) (use of manufacturer's name does not imply government endorsement). All treatment diets were fed *ad libitum*. We fed an historic diet of wild celery tubers, and present-day diets of either Baltic clams, or a combination of Baltic clams and whole corn, each fed *ad libitum*. We also attempted to feed individual diets of corn and soft-shelled clams (*Mya arenaria*), a common shellfish of Chesapeake Bay also known to be eaten by Canvasbacks (Perry and Uhler 1988), but those diets were discontinued after Canvasbacks developed an unexplained aversion and began losing weight at 28 and 49 days into trials, respectively. Corn is deficient in essential amino acids and certain minerals, especially calcium (Sibbald 1979, Baldassarre et al. 1983, Heitmeyer 1988), and it is possible that the ducks were unable to establish a long-term plane of nutrition on that single dietary source. We did not investigate the nutritional value of soft-shelled clams and rejection of that diet remains unexplained. Wild celery tubers were fall-harvested and purchased commercially from Wisconsin; Baltic clams were obtained fresh about every three days from the upper Chesapeake Bay and fed live. A control group was maintained on a commercial diet. Because of the large quantity of natural foods required to maintain ducks for long periods, we were con-

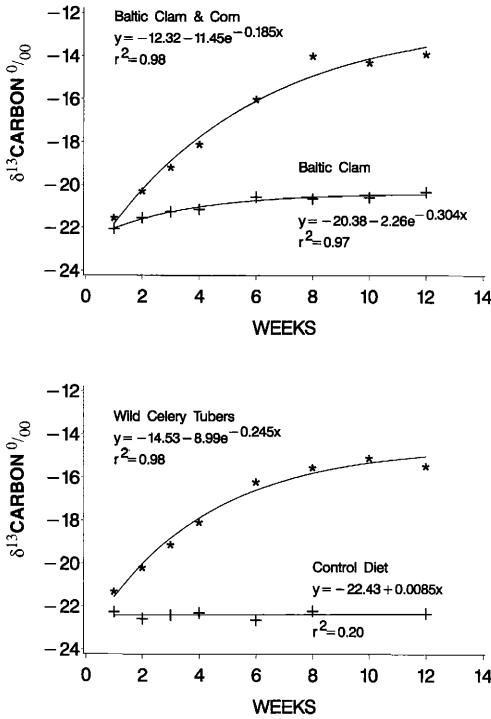


FIG. 1. Changes in whole blood stable-carbon isotope ratios for individual pen-reared Canvasbacks held on simulated winter diets of Baltic clams, Baltic clams and corn, wild celery tubers, and a control diet of commercial feed. Fitted equations result from curvilinear regression for treatment diets and linear regression for the control diet.

strained to feed those diets to single Canvasbacks. All treatment diets were fed *ad libitum*, or virtually so in the case of the Baltic clam diet that was difficult to maintain because of the large quantities of clams consumed.

Samples of whole blood (0.1 ml) were taken weekly for the first four weeks and every other week thereafter from the medial metatarsal vein of ducks under study. Blood samples were taken with a 25 gauge needle and syringe, transferred to clean vials and dried in an oven at 40°C before processing for stable-isotope ratios.

Diet fractionation changes of Canvasback whole blood were exponential in nature and were fitted with regression equations in the form of  $Y = a + be^{ct}$ , where  $Y$  is the isotopic value,  $a$  is the final asymptotic value,  $b$  is magnitude of change in isotopic signature between diets,  $c$  is turnover rate, and  $t$  is time in weeks. The turnover time constant  $T_c$  is calculated as  $1/c$ .

*Sampling wild Canvasbacks.*—To establish isotopic signatures of free-ranging Canvasbacks wintering in upper Chesapeake Bay, ducks were trapped in early

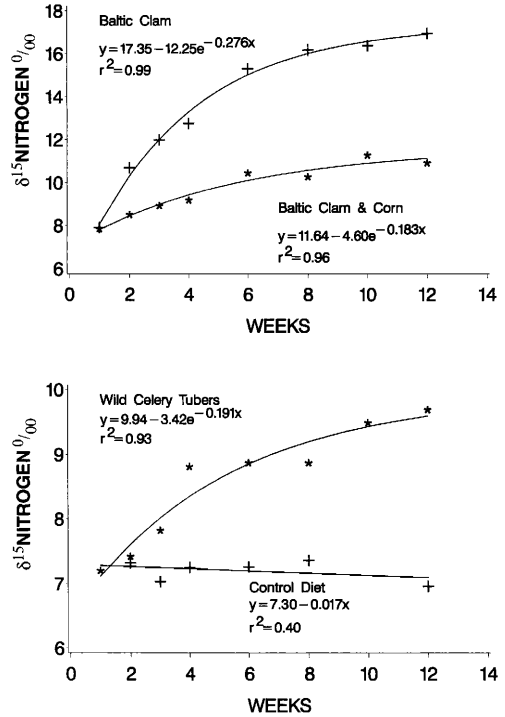


FIG. 2. Changes in whole blood stable-nitrogen isotope ratios for individual pen-reared Canvasbacks held on simulated winter diets of Baltic clams, Baltic clams and corn, wild celery tubers, and a control diet of commercial feed. Fitted equations result from curvilinear regression for treatment diets and linear regression for the control diet.

(December 1992) and late winter (late February 1993 and 1994) following procedures outlined in Haramis et al. (1982, 1987). Blood samples were collected in the field from the medial metatarsal vein and the ducks were released at the capture site.

*Isotope analysis.*—Canvasback foods and whole blood were converted to  $\text{CO}_2$  and  $\text{N}_2$  using the Dumas sealed-tube method described by Macko et al (1987). Samples were oven dried and powdered with mortar and pestle. Approximately 5 mg of sample powder was mixed with excess copper and precombusted copper oxide in a quartz tube that was evacuated, sealed, and combusted at 900°C for 1 h.  $\text{CO}_2$  and  $\text{N}_2$  were cryogenically separated from other gaseous products and isotopic ratios were measured on a dual inlet, triple collector VG PRISM stable-isotope ratio mass spectrophotometer. Stable-isotope concentrations were expressed as parts per thousand (‰) following the equation  $\delta X = ([R_{\text{sample}}/R_{\text{standard}}] - 1) \times 10^3$ , where  $X$  is  $^{15}\text{N}$  or  $^{13}\text{C}$  and  $R$  is the corresponding ratio  $^{15}\text{N}/^{14}\text{N}$  or  $^{13}\text{C}/^{12}\text{C}$ .  $R_{\text{standard}}$  for nitrogen is atmospheric  $\text{N}_2$ , and the Peedee Belemnite (PDB) from the Peedee Formation of South Carolina

TABLE 1. Asymptotic diet-tissue fractionation end-point values (loci) and turnover time constants of stable-isotope signatures of Canvasback whole blood for various treatment diets as determined from regression. Time constants,  $T_C$ , are reported in weeks.

Diet	$\delta^{13}\text{C}$ Carbon (‰)/ $T_C$	$\delta^{15}\text{N}$ Nitrogen (‰)sh $T_C$
Baltic clams	-20.38/3.29	+17.35/3.62
Baltic clams and corn	-12.32/5.41	+11.64/5.46
Wild celery tubers	-14.53/4.08	+9.94/5.24
Soft-shelled clam	-21.70/—	+19.36/5.06
Commercial diet	-22.39 <sup>a</sup> /—	+7.20 <sup>a</sup> /—

<sup>a</sup> Mean values for sample size of  $n = 7$  for carbon and  $n = 6$  for nitrogen.

is the standard for carbon. Those standards have been assigned a  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  of 0.0‰, respectively. Replicate measurements of internal laboratory standards have indicated a reproducibility of  $\pm 0.1$ ‰ for  $\delta^{13}\text{C}$  and  $\pm 0.2$ ‰ for  $\delta^{15}\text{N}$  (Macko et al. 1987).

## RESULTS

Penned Canvasbacks were predicted to begin the study with similar isotopic signatures after a year on the same commercial diet. Measurement of the commercial food averaged  $-23.99$ ‰  $\delta^{13}\text{C}$  and  $+4.07$ ‰  $\delta^{15}\text{N}$  ( $n = 2$  pooled samples) and Canvasback whole blood signatures just prior to placement on feeding trials averaged  $-22.54 \pm 0.04\text{SE}$ ‰  $\delta^{13}\text{C}$  and  $+7.05 \pm 0.09\text{SE}$ ‰  $\delta^{15}\text{N}$  ( $n = 12$ ). Those isotopic signatures indicate diet-tissue fractionation values for Canvasback whole blood of  $+1.45$ ‰ for car-

bon and  $+2.98$ ‰ for nitrogen. Those fractionation values are consistent with findings from other stable-isotope studies (DeNiro and Epstein 1978, Teiszen et al. 1983, Fry et al. 1984, Mizutani et al. 1991, Hobson and Clark 1992).

Isotopic signature changes for the 12 week duration of the feeding trials produced well-fit curvilinear regressions with  $r^2$  ranging from 0.93 to 0.99 (Figs. 1 and 2). As anticipated, isotopic values of ducks held on the commercial control diet continued unchanged: slopes of linear regressions were not different from zero ( $\delta^{13}\text{C}$ :  $t = -0.46$ ,  $df = 5$ ,  $P = 0.67$ ;  $\delta^{15}\text{N}$ :  $t = 0.43$ ,  $df = 4$ ,  $P = 0.66$ ). End point loci and time constants estimated from regression equations (Table 1) varied from 3.3 to 5.5 weeks, and averaged 4.7 weeks for four individual Canvasbacks on experimental diets.

Blood samples were taken from 20 Canvasbacks that were bait trapped in upper Chesapeake Bay in early winter (22 December) 1992 and from 27 and 32 ducks, respectively, that were trapped in late winter (20 February) 1993 and 1994. Stable-isotope signatures did not vary by sex or age for Canvasbacks trapped on a given date (Table 2), and subsequently samples were pooled. Five of six contrasts between pooled means from the three sampling dates were significantly different (Table 2). Canvasbacks trapped in early winter 1992 displayed isotope values more depleted in  $\delta^{15}\text{N}$  and enriched in  $\delta^{13}\text{C}$  than those trapped in late winter in 1993 and 1994. That difference is clearly depicted by the shift in distribution of the two

TABLE 2. Mean stable isotope values ( $\pm$ SE) for wild Canvasbacks trapped in upper Chesapeake Bay in 1992-94.

Date	$\delta^{13}\text{C}$ Carbon (‰) <sup>a</sup>	$\delta^{15}\text{N}$ Nitrogen (‰) <sup>a</sup>
Early winter: 22 December 1992		
Adult male ( $n = 13$ )	$-16.09 \pm 0.59$	$+10.54 \pm 0.58$
Adult female ( $n = 3$ )	$-18.74 \pm 0.45$	$+12.70 \pm 0.88$
Juveniles-both sexes ( $n = 4$ )	$-18.46 \pm 1.25$	$+11.31 \pm 1.35$
Pooled ( $n = 20$ )	$-16.96 \pm 0.52$ A <sup>b</sup>	$+11.02 \pm 0.49$ A <sup>b</sup>
Late winter: 20 February 1993		
Adult male ( $n = 14$ )	$-17.93 \pm 0.53$	$+13.53 \pm 0.40$
Adult female ( $n = 13$ )	$-18.20 \pm 0.45$	$+13.53 \pm 0.32$
Pooled ( $n = 27$ )	$-18.06 \pm 0.35$ A	$+13.53 \pm 0.25$ B
Late winter: 20 February 1994		
Adult male ( $n = 16$ )	$-19.52 \pm 0.37$	$+15.31 \pm 0.29$
Adult female ( $n = 16$ )	$-19.28 \pm 0.46$	$+15.86 \pm 0.26$
Pooled ( $n = 32$ )	$-19.40 \pm 0.29$ B	$+15.58 \pm 0.19$ C

<sup>a</sup> Results of ANOVA found no difference among gender/age within each sample date or the year  $\times$  gender/age interaction ( $\delta^{13}\text{C}$ : for gender/age,  $F = 2.03$ ,  $df = 2$  and  $72$ ,  $P = 0.13$ , for year  $\times$  gender/age interaction,  $F = 2.35$ ,  $df = 2$  and  $72$ ,  $P = 0.10$ ;  $\delta^{15}\text{N}$ : for gender/age,  $F = 1.33$ ,  $df = 2$  and  $72$ ,  $P = 0.27$ , for year  $\times$  gender/age interaction,  $F = 1.91$ ,  $df = 2$  and  $72$ ,  $P = 0.16$ ).

<sup>b</sup> Pooled category means sharing the same letter between sample dates (A-C), do not differ (Tukey's test,  $P = 0.05$ ).

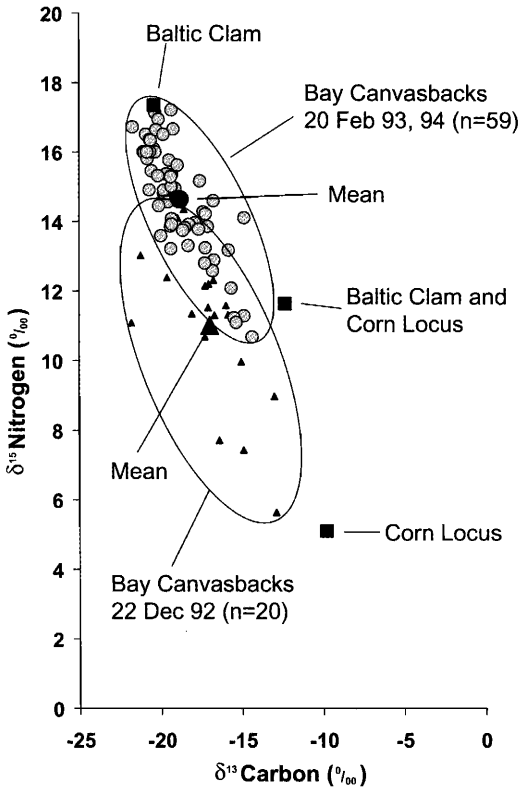


FIG. 3. Distribution and means of whole blood carbon-nitrogen isotopic signatures for early (22 December 1992) and late-winter (20 February 1993 and 1994) Chesapeake Bay Canvasbacks (separate ellipses). End point loci for Baltic clam and Baltic clam and corn treatment diets were determined from regression asymptotes (Figs. 1 and 2). The corn locus was estimated assuming a fractionation of +1.5‰ for  $\delta^{13}\text{C}$  and +3‰ for  $\delta^{15}\text{N}$ .

data sets circled by separate ellipses in Figure 3. In contrast, late-winter Canvasbacks had isotopic signatures that fell closely on a continuum between the Baltic clam locus and the Baltic clam and corn locus as determined from penned feeding trials (Fig. 3). In comparison to the February 1993 samples, February 1994 samples were on average depleted 1.34‰ in  $\delta^{13}\text{C}$  and enriched 2.05‰ in  $\delta^{15}\text{N}$  (Table 2). The mean isotopic signature of Canvasbacks sampled in late winter 1993 and 1994 ( $n = 59$ ) was  $-18.78\text{‰}$   $\delta^{13}\text{C}$  and  $+14.64\text{‰}$   $\delta^{15}\text{N}$ .

The common benthos from mud bottoms of the upper bay were grouped taxonomically as amphipods, polychaetes, and isopods, along with four common bivalves, Baltic clam, nar-

rowed macoma (*Macoma mitchelli*), soft-shelled clam, and brackish water clam (*Rangia cuneata*). We also included three species of submerged aquatic vegetation: wild celery obtained from Wisconsin, and widgeon grass and horned pondweed collected from the upper bay (Table 3).

## DISCUSSION

The generally good fit observed between the late-winter Canvasback data points and the diet end-point loci depicted in Figure 3 supports our fundamental premise that Canvasbacks are dependent on benthic animal resources supplemented with corn-SAV in the upper Chesapeake Bay. A closer look at that fit (Fig. 4) reveals the line connecting the two diet loci (points A and B, line  $y'$ , respectively, Fig. 4), and the linear regression of 59 stable-isotope signatures of free-ranging, late-winter Canvasbacks (line  $y$ , Fig. 4), to be virtually equal in slope with an offset 'D' of 0.5‰  $\delta^{13}\text{C}$  and 1.1‰  $\delta^{15}\text{N}$ . That offset cannot be statistically confirmed given the lack of information about variation, that is, the diet-loci line is the result of only two data points. However, the small offset, if real, seems acceptable, even remarkable, given resulting variation in isotopic values associated with slightly variable diets of wild Canvasbacks, apparent variation of isotopic signature of food items between locations in the upper bay (Table 3), and any lack in measure of the variation in diet-tissue fractionation among Canvasbacks. Given those sources of variation, we conclude that the pen and field data are in reasonably close agreement and demonstrate precision of the fractionation process and accuracy of our estimated end-point locus values.

The mean isotopic signature for the 59 wild Canvasbacks clearly indicates that an estimated 15,000–20,000 Canvasbacks wintering in upper Chesapeake Bay do not have enough corn-SAV available to achieve an *ad libitum* consumption. Importantly, signatures of other common bay benthos potentially consumed by wintering Canvasbacks (Table 3) are, as predicted, similar in nature to Baltic clams in being more depleted in  $\delta^{13}\text{C}$  and enriched in  $\delta^{15}\text{N}$  than corn-SAV. We note that is not a complete inventory of all possible foods eaten by wintering Canvasbacks, but represents the most common items

TABLE 3. Stable isotope signatures from pooled samples of common benthic invertebrates taken from 3 sites in the Chester River–Eastern Bay region of upper Chesapeake Bay in 1993 and 1998, and from submerged aquatic vegetation taken in 1998.

Taxon	$\delta^{13}\text{C}$ Carbon (‰)		$\delta^{15}\text{N}$ Nitrogen (‰)		n
	Range	Mean	Range	Mean	
Benthic infauna					
Amphipods	-18.47, -22.48	-20.57	11.13, 14.13	12.70	3
Polychaetes	-13.75, -21.00	-17.05	11.54, 14.77	13.80	10
Isopods	-13.81, -19.32	-16.02	13.39, 16.27	15.15	7
Bivalves					
<i>Macoma balthica</i>	-17.92, -20.92	-19.47	12.14, 14.51	13.28	9
<i>Macoma mitchelli</i>	-14.54, -19.04	-16.95	11.61, 13.33	12.57	5
<i>Mya arenaria</i>	-20.32, -20.86	-20.63	13.00, 13.63	13.37	5
<i>Rangia cuneata</i>	-20.26, -22.18	-20.99	13.44, 14.10	13.74	3
Submerged aquatic vegetation					
<i>Vallisneria americana</i> <sup>a</sup>	-14.26, -14.19	-14.23	5.84, 5.76	5.80	2
<i>Ruppia maritima</i>	-12.58, -15.23	-14.05	7.61, 12.21	9.24	4
<i>Zannichellia palustris</i>	-12.27, -13.54	-12.91	7.13, 11.56	9.35	2

<sup>a</sup> Tubers obtained from Wisconsin.

from our best knowledge of their food preferences. Those findings substantiate the unique isotopic signature of corn–SAV in a winter bay environment where benthic infauna are the foods most available to Canvasbacks.

Turnover time constants were more variable than anticipated although with the restricted sample size of one Canvasback per diet, a true measure of variation was unobtainable. Whether the variation observed (Table 1) is simply

representative of natural differences in metabolism in individual ducks housed in outside pens in winter, or whether it represents differences in accommodation to the abrupt diet change from commercial to natural foods, is not known. However, all Canvasbacks seemed to accommodate well to the diet change except for the corn-only and soft-shelled-clam-only diets to which ducks developed an aversion (see above). Feeding trials did demonstrate ability of individual Canvasbacks to establish a long-term plane of nutrition on single dietary sources of wild celery tubers and Baltic clams, two traditional fall–winter foods of the species.

We estimate that most wintering Canvasbacks are resident on the bay by 15 December and this provides a 10 week residency until the sampling date of 20 February. On the basis of our measured worst- and best-case time constants (Table 1: 5.46 vs. 3.29 weeks), Canvasback isotopic signatures would transition from 84 to 95% of end-point values in that 10 week period. That estimate is conservative because many Canvasbacks arrive on the bay before 15 December and also because free-ranging ducks typically have higher metabolism and thus higher turnover rates than their penned counterparts (Nagy 1987). We therefore conclude that our late-winter stable-isotope measurements of Canvasback whole blood do provide a valid representation of bay diet near asymptotic end-point values. We note that since our work was initiated, Hobson and Clark (1993) reported isotopic turnover in blood plasma to

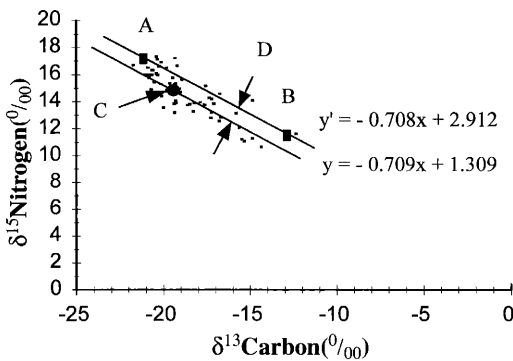


FIG. 4. A comparison of (1) the linear regression ( $y$ ) of whole blood carbon and nitrogen stable isotope values from combined 1993 and 1994 late-winter Chesapeake Bay Canvasbacks ( $n = 59$ , data points shown), and (2) the straight line ( $y'$ ) drawn between two end-point loci as determined from penned feeding trials with single Canvasbacks held on treatment diets of Baltic clams (locus A) and Baltic clams and corn (locus B). C is the mean value of 59 winter-sampled Canvasbacks and D is the observed offset between the lines.

be  $\sim 10\times$  more rapid than that of the cellular blood fraction. In hindsight, use of the plasma fraction of centrifuged whole blood should be considered in applications where rapid turnover rate is desired.

*Significance of diet to winter energetics of Canvasbacks.*—Wintering on a low-benefit shellfish diet places Canvasbacks at energetic risk in balancing nutrient reserves, dietary energy intake, and energy expenditure under cold weather conditions (Perry et al. 1986, Lovvorn 1994, Jorde et al. 1995, de Leeuw et al. 1999). Because of its high carbohydrate content, corn is a calorically dense food that is highly digestible in a winter environment where calorically dense natural foods, such as the tubers of wild celery, are virtually unavailable. Predictable availability of corn at feeding sites can thus be of great benefit to wintering Canvasbacks that otherwise must invest considerable foraging costs in diving to obtain large quantities of relatively low-benefit shellfish. For example, in pen studies de Leeuw et al. (1999) found Tufted Ducks (*Aythya fuligula*) consumed daily up to  $3\times$  their body mass (600 g) in zebra mussels (*Dreissena polymorpha*) and Jorde et al. (1995) found Canvasbacks to consume as much as twice their body mass (1200 g) in Baltic clams. Such high rates of consumption can deplete natural stocks of shellfish and result in starvation under harsh winter weather conditions (Lovvorn 1989, Suter and van Eerden 1992). Corn therefore is a ready source of digestible energy that plays an important role in mediating that dependency.

As an integration of assimilated elements from winter diet, mean isotopic signature of late-winter Canvasbacks combined for 1993 and 1994 ( $n = 59$ ) can be used to estimate percentage of foods consumed. We considered mean isotopic value to be a simple arithmetic sum of a proportion of two diets: clams and corn, and clams only. To estimate relative contribution of carbon and nitrogen from dietary components, we transferred the mean 'C' to line A-B (Fig. 4) and calculated proportions. Results indicated that for carbon 73.5% came from the clam component of the diet, whereas 26.5% came from the corn and clam component; for nitrogen, 71.8% came from the clam diet, whereas 28.2% came from the corn and clam diet.

An important question with regard to winter energetics is how much energy does the corn

component of the diet contribute to the daily energy budget of wintering Canvasbacks? On the basis of the mean daily consumption of corn for the Baltic clam and corn *ad libitum* diet of 65 g per day, we estimated consumption based on 26.5%, that is, assuming a linear proportioning of food intake and constant assimilation rate. Thus, on average, Canvasbacks were receiving  $\sim 17.2$ g dry weight of corn per bird per day. Assuming metabolizable energy content of corn is 3.80 kcal/g (Sibbald 1979), the corn component of the diet would contribute  $\sim 65.4$  kcal to the daily energy budget of Canvasbacks. Using an allometric equation provided by Kendeigh et al. (1977) for daily existence metabolism (EM) for nonpasserine birds at  $0^\circ\text{C}$  and a  $10 \pm$  h photoperiod during winter, EM can be calculated for a 1200 g Canvasback as 183.5 kcal per day. Field metabolic rate (FMR) has been estimated as  $2\times$  EM following Nilsson (1980). Thus, Canvasbacks are estimated to have a daily energy requirement in winter of 367 kcal per day. On the basis of those calculations, mean daily corn component of the diet would provide  $\sim 35.6\%$  of EM and 17.8% of FMR for a wintering Canvasback. At that indicated feeding rate, corn consumption for an estimated 15,000 Canvasbacks frequenting feeding sites in the upper bay, where we sampled Canvasbacks, would be 258 kg per day. That figure seems very realistic in terms of amount of corn potentially available to Canvasbacks in that region—so realistic in fact that corn alone could easily account for the observed enrichment in the carbon signature of Canvasback blood, leaving strong doubt that SAV has any significant role in the diet of Canvasbacks in that region of the bay.

Validity of our estimate depends on how well penned Canvasbacks are a true parallel of wild Canvasbacks with regard to metabolic processes during winter. We suggest that the greatest potential difference lies in narrow limits imposed on daily energy balance by the combined effect of higher metabolic costs and low-benefit diet in wild Canvasbacks which leads ultimately to a dependence on stored reserves. Differential use of stored reserves, both fat and lean body mass, between penned and wild birds can bias our measurements of blood isotopic signature and our interpretation of diet (Hobson et al. 1993, Hobson and Stirling 1997). Whereas regulation of stored reserves can be critical to

survival during periods of winter stress (Lovvorn 1994), our field knowledge of Canvasbacks on Chesapeake Bay suggests that only during periods of subfreezing conditions and ice cover, which limits access to food, are ducks unable to meet their energetic needs through daily foraging and revert to dependence on body reserves (Nichols and Haramis 1980, Jorde et al. 1995). We therefore would expect isotopic signatures of penned ducks to most closely represent those of wild ducks when energetic needs of wild ducks are met through daily foraging with minimal dependence on stored reserves. If those conditions are assumed in our isotopic evaluation of Chesapeake Bay Canvasbacks, then our estimate of corn consumption is conservative in proportion to daily metabolic requirements of penned versus free-ranging ducks. Some evidence of effects of cold stress on Canvasbacks appeared in our study when a significant difference in mean isotopic values occurred (Table 2) between wild Canvasbacks sampled in February 1993, a generally mild winter with no icing, versus February 1994, a winter with an extended period of ice cover. A depletion in  $\delta^{13}\text{C}$  of 1.34‰ and enrichment in  $\delta^{15}\text{N}$  of 2.05‰ in the colder 1994 winter suggests a greater dependence on benthic food resources, although it also might result from catabolism of body reserves under cold stress. The latter, however, seems unlikely because experimental evidence has shown that both  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  become enriched in blood of birds under nutritional stress (Hobson et al. 1993, Hatch et al. 1995). Thus, we conclude that our observation of a depletion in carbon and enrichment of nitrogen signature of Canvasbacks under stressful winter conditions is more consistent with a dietary shift to a greater dependence on benthic food resources, rather than catabolic processes. We suggest that shift was caused in part by the reduced availability of corn as a result of near-shore icing.

Another possible bias that might influence our results is differential metabolic use of dietary corn by penned and wild Canvasbacks. That difference follows a general pattern that omnivores that have both animal (protein-rich) and plant (carbohydrate-rich) components in the diet, generally use the animal component for tissue synthesis and the plant component for energy (Hobson and Stirling 1997, Hobson et al. 1999). Because digestible energy is a nu-

tritional priority for wintering ducks, and bay Canvasbacks receive limited amounts of corn, it is likely that a disproportionate amount of readily digestible corn carbohydrate would be used to fuel metabolism, rather than be utilized for tissue synthesis. Although unconfirmed for wintering Canvasbacks, that mechanism seems likely and would result in having a conservative effect on our estimate of corn consumption from isotopic measures of whole blood.

Finally, we must address the possibility of capture heterogeneity for Canvasbacks trapped with corn as bait. Certainly our results could be influenced by sampling ducks that are entrained to feeding on corn versus ducks that are less dependent on such food. However, we would argue that corn has been such a predictable part of this environment for so many decades that recognition by Canvasbacks must be universal if not nearly so. We conclude that our findings are most specific to Canvasbacks wintering along the upper western shore of Chesapeake Bay but likely have general applicability to all birds within that region.

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