lustrated by their diet during spring migration. Ornis Scand. 9:178-192.
MAJOR, R. E. 1990. Stomach flushing of an insectivorous bird: An assessment of differential digestibility of prey and the risk to birds. Aust. Wildl. Res. 17:647-657.
MOODY, D. T. 1970. A method for obtaining food samples from insectivorous birds. Auk 87:579.
RANDALL, R. M., AND I. S. DAVIDSON. 1981. Device for obtaining food samples from the stomachs of Jackass Penguins. S. Afr. J. Wildl. Res. 11:121-125.
ROSENBERG, K. V., AND R. J. COOPER. 1990. Approaches to avian diet analysis. Stud. Avian Biol. 13:80-90.
RYAN, P. G., AND S. JACKSON. 1986. Stomach pumping: Is killing seabirds necessary? Auk 103:427-428.
WILSON, R. P. 1984. An improved stomach pump for penguins and other seabirds. J. Field Ornithol. 55:109-112.
WILSON, R. P., M.-P. WILSON, D. C. DUFFY, B. A. M, AND N. KLAGES. 1989. Diving behaviour and prey of the Humboldt Penguin (*Spheniscus humboldti*). J. Ornithol. 130:75-79.
ZACH, R., AND J. B. FALLS. 1976. Bias and mortality in the use of tartar emetic to determine the diet of Ovenbirds (Aves: Parulidae). Can. J. Zool. 54: 1599-1603.

Received 28 March 1994, accepted 27 May 1994.

The Auk 112(3):782-785, 1995

Intersexual Comparison of Plasma Osmolytes, Kidney Size, and Glomerular Number and Size in Pekin Ducks (*Anas platyrhynchos*)

MARYANNE R. HUGHES[1], ELDON J. BRAUN[2], AND DARIN C. BENNETT[1]
[1]*Department of Zoology, University of British Columbia, 6270 University Blvd., Vancouver, British Columbia V6T 1Z4, Canada; and*
[2]*Department of Physiology, College of Medicine, University of Arizona, Tucson, Arizona 85724, USA*

Increased sodium chloride (NaCl) intake resulted in sexually disparate changes in body mass and plasma osmolality (Osm_{pl}; Hughes et al. 1992) and plasma concentrations of the osmoregulatory hormones arginine vasotocin ($[AVT]_{pl}$) and angiotensin II ($[AII]_{pl}$; Zenteno-Savin 1991) in Pekin ducks (*Anas platyrhynchos*). During gradual increase in drinking water NaCl concentration ([NaCl]), both sexes maintained body mass and changed Osm_{pl}, $[AVT]_{pl}$, and $[AII]_{pl}$ similarly until they drank 300 mM NaCl; at higher [NaCl], males became more dehydrated and their Osm_{pl}, $[AVT]_{pl}$, and $[AII]_{pl}$ increased significantly more than in females. Kidney mass tended to be smaller in male ducks than in female ducks (Hughes et al. 1989, 1992).

In this study, we reexamined body mass, hematocrit (Hct), plasma ion concentrations, and Osm_{pl} of male and female Pekin ducks rapidly acclimated (over three weeks) to 225 mM NaCl and following longer exposure to this [NaCl] (four and seven months). Finally, the ducks were euthanized and dissected to obtain organ masses. We found female ducks had larger kidneys, so we counted glomeruli (nephrons) in kidneys of both sexes to determine if the kidneys of females actually contained more nephrons. We also measured glomerular diameters to determine if glomerular sizes were equally represented in both sexes.

Methods and materials.—Twelve Pekin ducks (6 male, 6 female) were reared for six months in adjacent enclosures on *ad libitum* tap water and duck pellets (Buckerfield's, Abbottsford, British Columbia; sodium, $[Na^+]$, potassium, $[K^+]$, and chloride, $[Cl^-]$, concentrations, 83, 153.5, and 99 $mM \cdot Kg^{-1}$, respectively). They were then acclimated in three equal weekly increments to 225 mM NaCl. Drinking this solution at the minimum rate measured for Pekin ducks (Hughes et al. 1991) should replace at least one-half of the Na^+ in the extracellular fluid each day. This Na^+ intake should have been sufficient to increase the secretory potential of the salt glands allowing Osm_{pl} to be maintained (Hughes et al. 1992). At the end of each week, birds were weighed and 5.0 mL blood samples were taken. The birds were held on 225 mM NaCl for seven months and blood samples were taken at the end of the fourth and seventh months. Triplicate Strumia microhematocrit tubes were immediately filled from each blood sample and centrifuged simultaneously with the remaining blood for 3 min at 15,600 × g. After the last blood sampling, four female and three male ducks were intravenously infused with Alcian blue for 30 min to stain the glomeruli (Bankir and Hollenberg 1983) and then euthanized with a lethal dose of sodium pentobarbitol. The heart, liver, kidneys, and salt, adrenal, and Harderian glands were removed and weighed with a Mettler top-loading balance with an accuracy of 1 mg. Experiments conformed to guidelines of the Canadian Council on Animal Care.

All analyses were done at least in duplicate. De-

TABLE 1. Body mass, hematocrit (Hct), plasma osmolality, and plasma ion concentrations of freshwater (FW) male and female Pekin ducks after rapid acclimation (SW) and prolonged exposure (SW4, four months; SW7, seven months) to 225 mM NaCl. Given as $\bar{x} \pm$ SE.

Group	Body mass (g)	Hct (percent)	Ion concentration (mM) Na	K	Cl	Osmolality (mOsm·kg⁻¹)
			Male (n = 6)			
FW	3,217 ± 104	43.5 ± 0.6	148.2 ± 0.7	3.1 ± 0.1	108.4 ± 1.9	289.5 ± 1.6
SW	3,058 ± 96*	44.9 ± 0.4*	148.3 ± 1.4	3.3 ± 0.2	109.1 ± 1.2	290.3 ± 2.2
SW4	3,058 ± 11	41.5 ± 0.6*	152.6 ± 3.1	2.7 ± 0.1	104.6 ± 4.4	295.3 ± 5.8
SW7	3,134 ± 17	—	148.4 ± 1.0	2.0 ± 0.1*	—	—
			Female (n = 6)			
FW	2,941 ± 185	48.4 ± 0.1^B	147.4 ± 0.6	3.3 ± 0.1	107.1 ± 1.7	283.3 ± 1.1^B
SW	3,075 ± 203	43.9 ± 1.1*	143.8 ± 1.2*^A	3.0 ± 0.2	106.4 ± 1.9	284.9 ± 1.6
SW4	2,712 ± 112	39.5 ± 0.7*	147.4 ± 3.5	2.6 ± 0.1	107.0 ± 3.3	291.1 ± 6.6
SW7	2,555 ± 98^A	—	148.6 ± 1.9	2.7 ± 0.7	—	—

* $P < 0.05$. Significant difference between designated value and immediately preceding value (Bonferroni-adjusted t-test).
^A, $P < 0.05$. ^B, $P < 0.05$. Comparison of males and females on same drinking water (t-test).

terminations were made of: plasma sodium, $[Na^+]_{pl}$, and potassium, $[K^+]_{pl}$ concentrations with an Instrumentation Laboratory Model 943 flame photometer (Instrumentation Laboratory S.p.A. Milan, Italy); Osm_{pl} with a Wescor Model 5500 osmometer (Wescor, Inc., Logan, Utah); and plasma chloride ($[Cl^-]_{pl}$) with a Buchler digital chloridometer (Searle Buchler Instruments, Fort Lee, New Jersey). Glomeruli were prepared by the method of Niznik et al. (1985). The acid-digested kidneys were fragmented in distilled water in 2-L flasks. We counted 1-ml aliquots using Cue-2 Planomorphometry image-analysis software (Olympus Corporation, New York).

Data are given as means ± SE and analyzed statistically using SYSTAT (Wilkinson 1990). Data for the sexes were compared with independent sample t-tests (with pooled variances). Sequential samples were compared using repeated paired t-tests with Bonferroni-adjusted probabilities. An arcsin transformation was applied to organ mass, as percent body mass, before t-tests were applied.

Results.—When tap water was drunk, females had higher Hct ($P < 0.01$) and lower Osm_{pl} ($P < 0.01$) than males (Table 1). When drinking water [NaCl] was increased to 225 mM over three weeks, males lost mass ($P < 0.05$) and increased Hct ($P < 0.05$), but females maintained mass and decreased Hct ($P < 0.05$) and $[Na^+]_{pl}$ ($P < 0.05$).

By the end of the fourth month on 225 mM NaCl, male Hct had returned (increased) to the FW value, but had decreased further ($P < 0.05$) in females. Other blood parameters did not change significantly (Table 1). By the end of the seventh month, $[K^+]_{pl}$ was decreased only in males. The Hct, $[Cl^-]_{pl}$, and Osm_{pl} were not measured. Although females did not lose mass when initially exposed to saline (as males did),

TABLE 2. Body mass and mass of osmoregulatory organs in male and female Pekin ducks that had drunk 225 mM NaCl for seven months. Given as $\bar{x} \pm$ SE.

	Male Mass (g)	Male Percent body mass	Female Mass (g)	Female Percent body mass	P[a]
Body	3,134 ± 174	—	2,555 ± 98	—	0.016
Heart	25.5 ± 0.8	0.82 ± 0.04	23.9 ± 3.3	0.97 ± 0.18	0.464
Liver	78.0 ± 8.0	2.58 ± 0.41	116.6 ± 8.4	4.58 ± 0.32	0.003
Kidneys	22.0 ± 0.8	0.71 ± 0.05	27.4 ± 1.7	1.07 ± 0.05	0.001
Glands					
Salt	1.06 ± 0.11	0.035 ± 0.005	0.86 ± 0.04	0.034 ± 0.003	0.921
Harderian	1.59 ± 0.15	0.051 ± 0.003	1.27 ± 0.09	0.050 ± 0.004	0.924
Adrenal	0.27 ± 0.15	0.006 ± 0.004	0.28 ± 0.10	0.010 ± 0.096	0.575

[a] Comparison of body mass or organ mass (as percent of body mass) in males and females. Six birds of each sex (t-test).

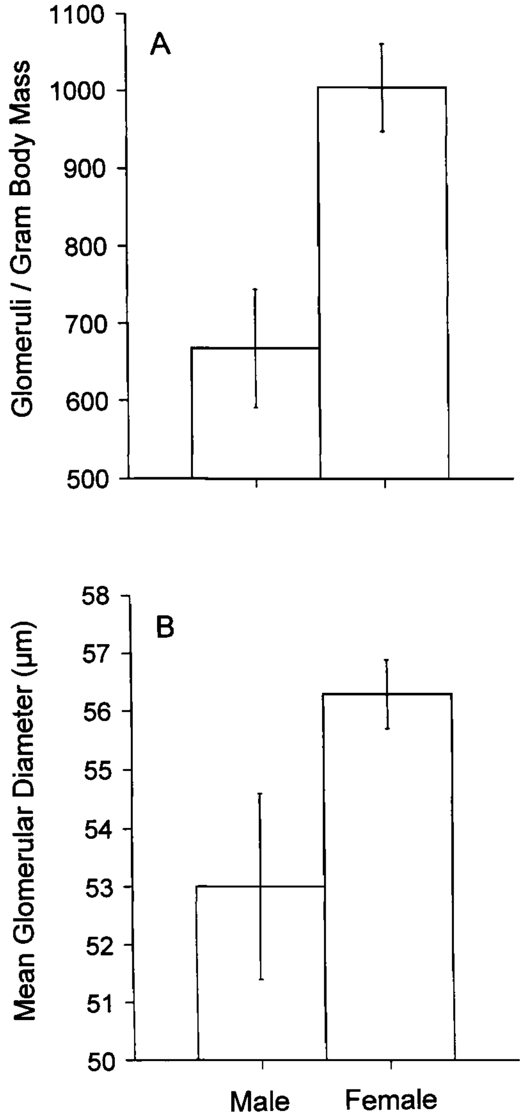

Fig. 1. (A) Number of glomeruli per gram body mass (difference between sexes; $P < 0.02$); (B) mean glomerular diameter in male and female Pekin ducks (difference suggestive but not significant; $P < 0.08$). Given as $\bar{x} \pm$ SE.

they gradually lost body mass with the result that their mass was significantly ($P < 0.05$) less than that of males at the end of the seventh month (Table 1).

At euthanasia, both sexes had the same relative masses of the heart, salt glands, adrenal glands, and Harderian glands. Absolute ($P < 0.01$) and relative ($P < 0.01$) liver mass and absolute ($P < 0.02$) and relative ($P < 0.001$) kidney mass were larger in females than in males (Table 2). Left kidneys (male, 10.9 ± 0.4 g; female, 14.0 ± 1.1 g) and right kidneys (male, 11.1 ± 0.4 g; female, 13.4 ± 0.8 g) had the same mass and number of glomeruli. It is suggestive, although not statistically significant ($P < 0.09$), that the total number of glomeruli in kidneys of females ($26.99 \pm 2.19 \times 10^5$; $n = 4$) was greater than in males ($20.68 \pm 1.84 \times 10^5$; $n = 3$); the number of glomeruli per gram body mass was significantly greater ($P < 0.02$) in females than in males (Fig. 1A). While not statistically significant ($P < 0.08$), there was a suggestion that glomerular diameter (Fig. 1B) was greater in females (56.3 ± 0.6 μm) than in males (53.0 ± 1.6 μm).

Discussion.—A sexual disparity in the responses of male and female ducks to saline drinking water (Hughes et al. 1989, 1992) was confirmed by the different changes in Hct and plasma concentration in the two sexes as drinking water [NaCl] increased (Table 1). When the [NaCl] of the drinking water was rapidly increased to 225 mM NaCl, males decreased body mass and increased Hct, but females maintained body mass and decreased Hct (and $[Na^+]_{pl}$). This suggests males became dehydrated, but that females actually retained water. In a similar study, Hughes et al. (1992) found females decreased $[Na^+]_{pl}$, but neither sex altered body mass or Hct. We found that prolonged exposure to saline was associated with decreased Hct in both sexes, but body mass decreased only in females, verifying the sexually disparate response to increased saline intake recorded by Hughes et al. (1992). Some of the changes recorded in these two studies were different and may reflect seasonal constraints on the osmoregulatory physiology of these birds. This saline acclimation occurred during the winter, whereas Hughes et al. (1992) acclimated ducks during the summer.

Renal mass of Pekin ducks differs between the sexes. When ducks drank tap water, females had heavier kidneys ($P < 0.05$; M. R. Hughes, E. A. Ayre, and K. M. Cheng unpubl. data); however, when they drank saline for a short time (Hughes et al. 1989, Hughes et al. 1992), the sexual disparity in kidney mass was not statistically significant ($P < 0.1$ and < 0.3, respectively). The ducks we studied drank saline for a protracted period (Table 1). The sexual difference in absolute renal mass was significant ($P < 0.02$); the relative renal mass of females was significantly greater ($P < 0.001$; Table 2). Not only were the kidneys of female ducks heavier than those of males, but they contained more nephrons (Fig. 1A), which had larger glomeruli (Fig. 1B). Female Pekin ducks that drank tap water had a higher glomerular filtration rate (GFR, $ml[min \cdot Kg]^{-1}$) than males, presumably reflecting the greater renal mass of females, since the GFR per gram kidney was the same in the two sexes (Hughes et al. 1989). When the ducks were given saline, females decreased GFR, but males did not (Hughes et al. 1989). Drinking-water salinity also did not affect GFR of male Pekin ducks in the studies of Holmes et al. (1968). Salt-gland secretion of male Pekin ducks may be more concentrated than that of females (Hughes et al. 1992).

Pekin ducks are the birds most often used for studies of renal (Holmes et al 1968, Gerstberger and Gray 1993) and extrarenal (Peaker and Linzell 1975) salt and water excretion in birds with salt glands. Since several studies (Hughes et al. 1989, 1992, this study) suggest that sex may influence osmoregulation in Pekin ducks, especially under conditions of saline stress, it would seem necessary to include sex as a variable when evaluating osmoregulatory responses.

Acknowledgments.—We gratefully acknowledge the conscientious animal care of Arthur Van der Horst and the technical expertise of: Cristina de Sobrino, who did some of the ion analyses; Sharon Fong, who helped with the saline acclimation experiment; and Yoshie Valadez, who did the glomerular analyses. We also thank Howard Haines, Peter Stettenheim, and a third anonymous reviewer for helpful critiques of the manuscript. This work was supported by a grant from the Natural Sciences and Engineering Council of Canada (A3442; M.R.H.) and a grant from the National Science Foundation (IBN; E.J.B.).

Literature Cited

BANKIR, L., AND N. K. HOLLENBERG. 1983. *In vivo* staining of the kidney with alcian blue: An adjunct to morphological and physiological studies. Renal Physiol. 6:151-155.

GERSTBERGER, R., AND D. A. GRAY. 1993. Fine structure, innervation, and functional control of avian salt glands. Intl. Rev. Cytol. 144:129-215.

HOLMES, W. N., G. L. FLETCHER, AND D. J. STEWART. 1968. The patterns of renal electrolyte excretion in the duck (*Anas platyrhynchos*) maintained on fresh water and hypertonic saline. J. Exp. Biol. 48:487-508.

HUGHES, M. R., D. KOJWANG, AND T. ZENTENO-SAVIN. 1992. Effects of caecal ligation and saline acclimation on plasma concentration and organ mass in male and female Pekin ducks, *Anas platyrhynchos*. J. Comp. Physiol. B Biochem. Syst. Environ. Physiol. 162:625-631.

HUGHES, M. R., J. R. ROBERTS, AND B. R. THOMAS. 1989. Renal function in freshwater and chronically saline-stressed male and female Pekin ducks. Poult. Sci. 68:408-416.

HUGHES, M. R., T. ZENTENO-SAVIN, AND D. KOJWANG. 1991. Effects of saline acclimation and cecal ligation on body water and water flux in male and female Pekin ducks. Can. J. Zool. 69:771-775.

NIZNIK, R. A., R. F. WIDEMAN, B. S. COWEN, AND R. E. KISSELL. 1985. Induction of urolithiasis in single-comb white leghorn pullets: Effect on glomerular number. Poult. Sci. 64:1430-1437.

PEAKER, M., AND J. L. LINZELL. 1975. Salt glands in birds and reptiles. Cambridge Univ. Press, Cambridge.

WILKINSON, L. SYSTAT: 1990. The system for statistics. SYSTAT, Inc. Evanston, Illinois.

ZENTENO-SAVIN, T. 1991. Plasma arginine vasotocin and angiotensin II concentrations during saline acclimation in birds with salt glands. M.Sc. thesis, Univ. British Columbia, Vancouver.

Received 12 January 1995, accepted 10 April 1995.

The Auk 112(3):785-790, 1995

Method for Sexing Fledglings in Cory's Shearwaters and Comments on Sex-ratio Variation

VINCENT BRETAGNOLLE[1] AND JEAN-CLAUDE THIBAULT[2]
[1]*Centre d'Etudes Biologiques de Chizé, Centre National de la Recherche Scientifique, F-79360 Beauvoir sur Niort, France; and*
[2]*Parc Naturel Régional de la Corse, B.P. 417, F-20184 Ajaccio, Corse, France*

Equal parental investment in the two sexes is a necessary consequence of natural selection (Fisher 1930). Studies of sex ratios at birth (or, in birds, at fledging) have focused particularly on sexually dimorphic species to test the prediction that there should be a bias towards the smaller, "cheaper" sex (Trivers 1972; also, see reviews in Clutton-Brock and Albon 1982, Clutton-Brock 1986, Breitwisch 1989). The prediction has been upheld in mammals, but not in birds (Clutton-Brock 1986, Breitwisch 1989, Richner 1991) and not even in species that are strongly dimorphic in size, such as raptors (Newton 1979, Edwards et al. 1988, Negro and Hiraldo 1992). In bird species that are not dimorphic in size, offspring sex ratios are poorly documented, probably because chicks are difficult to sex in the field. Methods of sexing chicks at or before fledging, therefore, are of considerable interest. In this paper, we describe the calls given by chicks in Cory's Shearwater (*Calonectris diomedea*). Using sexual dimorphism in voice, we derive a discriminant function based on morphometric measurements that allows the sex of fledglings to be determined. The sex ratio at fledging departed significantly from parity (male biased).