

SEXUALITY AND OTHER ASPECTS OF REPRODUCTION IN
ANODONTA (PELECYPODA: UNIONIDAE)¹

William H. Heard

Florida State University, Tallahassee, Florida 32306, U.S.A.

ABSTRACT

The unionid subfamily Anodontinae contains several genera, principally Nearctic in distribution, among which occur considerable variations in life histories and sexual conditions. Eight species of *Anodonta* from the U.S.A. (viz., *A. californiensis*, *A. corpulenta*, *A. couperiana*, *A. gibbosa*, *A. hallenbeckii*, *A. imbecilis*, *A. peggyae* and *A. wahlamatisensis*) were studied in order to identify and describe visceral sex, demibranch morphology, possible sex-reversal, age of sexual maturity, seasonal gonad activity, glochidial development and gravid periods, and adult life span.

Anodonta corpulenta, *A. gibbosa* and *A. wahlamatisensis* were uniformly dioecious, whereas *A. californiensis*, *A. couperiana*, *A. hallenbeckii* and *A. peggyae* contained at least 1 kind of hermaphrodite in addition to males and females. Male-hermaphrodites (with a preponderance of testicular tissue over ovarian tissue) occurred in *A. couperiana*, *A. hallenbeckii* and *A. peggyae*, and female-hermaphrodites (with a preponderance of ovarian tissue, or with approximately equal amounts of male and female gonadal tissue) occurred in *A. californiensis* and *A. couperiana*. *A. imbecilis* consisted only of female-hermaphrodites and females; males were entirely lacking.

All 8 species exhibited the typical anodontine gill morphology. The non-marsupial demibranch (viz., the inner 2 in females and female-hermaphrodites, and all 4 in males and male-hermaphrodites) were undivided by secondary septa and had comparatively many filaments between successive primary interlamellar septa. The marsupial demibranchs (the outer 2 in females and female-hermaphrodites) were seasonally divided by secondary septa that produced temporary secondary water-tubes, resulting in a tripartite organization, and they had comparatively few filaments between successive primary interlamellar septa. Consistent correlation between the visceral sex (i.e., type of gonad present) and outer demibranch morphology in animals of different age classes from different seasons indicated an absence of sex-reversal.

Employment of the number of annuli on the shell suggested that species of *Anodonta* may not reach sexual maturity (i.e., become gravid) before 5 years of age. Studies of gonad activity in animals from monthly (*A. gibbosa*, *A. imbecilis* and *A. peggyae*, including 2 populations of the latter) or bimonthly (*A. couperiana*) samples showed that differences in oögenesis were less striking seasonally than those in spermatogenesis. Testicular activity in *A. couperiana* and *A. gibbosa* generated sperm-morulae (i.e., multinucleated figures of atypical spermatogenesis) from late fall into the following summer, produced typical spermatogenesis in late summer and provided mature spermatozoa in the early fall. In contrast, *A. imbecilis* had few sperm-morulae of irregular occurrence, a peak in typical spermatogenesis from summer to fall and abundant mature spermatozoa at all times. Animals of 1 population of *A. peggyae* displayed testicular activity similar to that of *A. imbecilis*, but those of the other population showed 2 consecutive cycles of activity in a year. Examination of the states of glochidial development revealed that *A. couperiana* and *A. gibbosa* are short-term, winter-tachytictic breeders, *A. imbecilis* is a long-term, bradytictic breeder, and that *A. peggyae* can either be bradytictic or undergo 2 consecutive breeding cycles in a year. The adult life spans of the 8 species studied, judged from the number of annuli, varied interspecifically and intraspecifically. Males and females attained the same age in *A. californiensis*, *A. corpulenta*, *A. gibbosa* and 1 population of *A. peggyae*, whereas females reached a greater age in *A. couperiana*, *A. hallenbeckii*, *A. wahlamatisensis* and a 2nd population of *A. peggyae*. Both males and females were older than any type of hermaphrodite in the same species.

Monoecious individuals displayed either of 3 states of gonadal differentiation, although most contained intermingled zones of male and female acini. The occurrence of hermaphroditism, in its various forms of glandular and ctenidial organization, does not coincide with conchological taxonomic groupings, nor do the present findings support views of the monoecious state as an adaptation to environmental conditions. The manifestations of hermaphroditism in mussels may be described, but the underlying genetic basis remains unknown.

¹This study was supported, in part, by research grant 26-034 from the Florida State University Research Council (1964-1965).

INTRODUCTION

Female and some hermaphroditic animals of the numerous species of the fresh-water mussel family Unionidae Fleming undergo internal fertilization and incubate the developing young in the outer, marsupial demibranchs for varying periods of time, and most release glochidial larvae that are temporarily parasitic on the gills or fins of various species of fishes before assuming a free-living existence (Lefevre & Curtis, 1910a; Coker et al., 1921). However, a few exceptions have been reported: (1) Lefevre & Curtis (1911) stated that the glochidia of *Strophitus undulatus* (Say) [= *S. edentulus* (Say)] can metamorphose into juvenile mussels while still in the marsupial demibranchs and thus bypass a subsequent parasitic period, (2) Howard (1914) suggested that *Anodonta imbecilis* Say also displays similar direct development, and (3) Howard (1915) demonstrated the host of *Simpsoniconcha ambigua* (Say) to be not a fish but a salamander, *Necturus maculosus* (Rafinesque) [Proteidae].

Strophitus undulatus, *A. imbecilis* and *S. ambigua* are members of the unionid subfamily Anodontinae Ortmann (1910a), a group characterized in part as possessing marsupia which fill the entire outer demibranchs and form smooth pads without demarcation of the ovisacs (i.e., water-tubes, in which the embryos and subsequent larvae are brooded) by external sulci (Simpson, 1914). In addition, anodontine animals have been stated to be bradytictic breeders² (Ortmann, 1912). Species of the Anodontinae not only possess unique features among unionids, e.g., a tripartite organization of water-tubes and septa in the outer, marsupial demibranchs (cf. Fig. 5) but not in the non-marsupial demibranchs (Fig. 6), but also exhibit greater plasticity in life histories and sexual phenomena than do members of the other unionid subfamilies.

According to Sterki (1898a) and Ortmann (1911), the marsupial demibranchs of *Strophitus edentulus* contain a horizontal system of divided water-tubes and septa, whereas these elements are vertical in position in all other unionids. However, Heard & Fuller (in preparation) recently found that *S. rugosus* (Swainson) [= *S. edentulus*?] has

the typical tripartite anodontine organization oriented vertically, i.e., dorso-ventrally, and that what had been interpreted as a horizontal structure consists of a vertical series of "perforations" in the interlamellar septa (Fig. 7). In addition, the posterior $\frac{1}{10}$ - $\frac{1}{5}$ of the outer demibranchs lacked marsupial organization. Ortmann (1911) also stated that in *S. edentulus* the secondary water-tubes vanish after the glochidia are expelled. Each ovisac was said to house a short, rather cylindrical gelatinous mass which contains the embryos and subsequent larvae. These masses, in which the glochidia (infective or metamorphosed?) are expelled, were termed "placentae" by Sterki and "placentulae" by Ortmann.

Placentulae are not produced in gravid animals of such other anodontine genera as *Alasmidonta* Say, *Anodonta* Lamarck, *Anodontoides* Simpson and *Lasmigona* Rafinesque (Ortmann, 1911), from which the glochidia are released either individually or in small groups in which the members are joined by intertwined larval filaments (Lefevre & Curtis, 1910a; Tucker, 1927), or individually or in groups agglutinated by a mucous secretion from the incubation chamber (Bouillon, 1955). Gravid animals in these genera also contain the tripartite marsupial organization in the outer demibranchs, the division being created by the seasonal appearance of a secondary septum on each side (lateral and medial) of the primary, central water-tubes (Ortmann, 1911). According to Ortmann, only the primary water-tubes serve as the ovisacs during incubation, but this concept will be modified later in this paper.

The contrasting reports concerning the life cycle of *A. imbecilis* are of particular interest. Howard (1914) stated that he found (1) juveniles, i.e., metamorphosed glochidia, in the marsupial demibranchs, (2) no natural "infections" of glochidia on what he considered to be the likely fish hosts, and (3) only difficult and incomplete experimental encystment of glochidia on these fishes. Allen (1924) also claimed to have found metamorphosed glochidia in the marsupial demibranchs, although Tucker (1927, 1928) reported no glochidial metamorphosis except after encystment on the fins of *Lepomis (Apomotis) cyanellus*

²Bradytictic animals, also called long-term or winter breeders, incubate young for most of the year, except for a part of the Nearctic summer. In contrast, tachytictic animals, also known as short-term or summer breeders, are gravid only during a part of the Nearctic summer.

Rafinesque, the green sunfish [Centrarchidae]. According to Allen, *A. imbecilis* has a very short (3-4 weeks), repetitive reproductive cycle, with an incubation period of even less duration than that displayed by tachytictic species (viz., members of the Pleurobeminae and Unioninae *sensu* Heard & Guckert, 1971). Accepting prior reports that the animals of *A. imbecilis* are hermaphroditic (refuted later in this paper) and might self-fertilize, and that their larvae lack a parasitic habit, Allen hypothesized that these features and such a very short reproductive cycle might be correlated with a more rapid growth rate and comparatively early attainment of sexual maturity in this species. None of Allen's contentions were confirmed by the present study.

A survey of "unionids"³ for the occurrence of hermaphroditism has shown the monoecious condition to be common in the Anodontinae, and especially in *Anodonta* (cf. Van der Schalie, 1970; this paper, Table 1), although Bouillon (1955) previously stated that European species of *Anodonta*, none of which were named, are "dioïques mais dans les eaux stagnantes ce genre se montre souvent hermaphrodite." Van der Schalie listed only 4 "dominantly" herma-

phroditic unionids, including 3 in the Anodontinae, while 16 other species were stated to contain "occasional" (or "sporadic") monoecious individuals in samples composed largely of dioecious animals.

Although several kinds of monoecious conditions in pelecypods have been described, much information concerning the nature of hermaphroditism in freshwater mussels (viz., Unionacea and Mutelacea) is still lacking. It is the purpose of this report to provide further data on this phenomenon and other aspects of reproduction in the Holarctic genus *Anodonta*.

LIST OF SPECIES AND LOCALITIES

Animals of 8 North American species of *Anodonta* were studied, 5 species from the southeastern U.S.A. and 3 from the western U.S.A. A total of 840 individuals were taken in 64 collections made between 1963 and 1969.

Subgenus *Anodonta*, s.s.

1. *A. californiensis* Lea. 1 collection (7 individuals): 11 October 1966. Mill Creek at Bartlett Springs Road, Colusa County, California.

³Species of *Amblema* Rafinesque, *Fusconaia* Simpson, *Gonidea* Conrad, *Quadrula* Rafinesque and *Tritogonia* Agassiz, as well as "*Elliptio*" *sloatianus* (Lea) [= *Elliptoideus sloatianus* (Lea)], listed in the Unionidae by Van der Schalie (1970), were considered by Heard & Guckert (1971) to be members of the Amblemidae Rafinesque.

TABLE 1. Recorded occurrences of hermaphroditism in the Unionidae: Anodontinae. All records are from the U.S.A. unless stated otherwise.

Species	Locality	Reference
<i>Alasmidonta marginata</i> (Say) ^a	Michigan & Tennessee	Van der Schalie, 1970
<i>Anodonta anatina</i> (Linnaeus) ^a	England	Bloomer, 1936
<i>A. californiensis</i> Lea ^a	California	this paper
<i>A. corpulenta</i> Cooper ^a	Tennessee	Van der Schalie, 1970
<i>A. couperiana</i> Lea ^a	Florida	this paper
<i>A. cygnea</i> (Linnaeus) ^a	England	Bloomer, 1930, 1934, 1935, 1939
<i>A. grandis</i> f. <i>footiana</i> Lea ^a	Michigan	Van der Schalie & Locke, 1941
<i>A. hallenbeckii</i> Lea ^a	Alabama	this paper
<i>A. henryana</i> Lea ^b	Texas	Ortmann, 1911
<i>A. imbecilis</i> Say, ^{c,d}	Pennsylvania	Sterki, 1898b; Ortmann, 1911
	Missouri	Utterback, 1915
	Michigan	Van der Schalie, 1966, 1970
	Florida & Alabama	this paper
<i>A. peggyae</i> Johnson ^a	Florida	this paper
<i>Lasmigona complanata</i> (Barnes) ^a	Michigan	Van der Schalie, 1970
<i>L. compressa</i> (Lea) ^c	Pennsylvania	Ortmann, 1911
	Michigan	Van der Schalie, 1966, 1970
<i>L. subviridis</i> (Conrad) ^c	eastern U.S.A.	Van der Schalie, 1966, 1970
<i>Strophitus rugosus</i> (Swainson) ^a	Michigan	Van der Schalie, 1970

^aWith males and females.

^bMonoecious condition based only on inference, because of its alleged relationship to *A. imbecilis*.

^cAs hermaphrodites only.

^dWith females only.

2. *A. wahlamatis* Lea. 1 collection (7 animals): 10 October 1966. Upper Blue Lake at State Highway 20, Lake County, California.

Subgenus *Pyganodon* Crosse & Fischer

3. *A. corpulenta* Cooper. 1 collection (14 individuals): 9 November 1969. Lake Mary, about 10 miles NE of Flagstaff, Coconino County, Arizona.
4. *A. gibbosa* Say. 10 collections (total of 53 individuals): monthly, except in June and August, 1965. Holmes Creek at Federal Highway 90, 4 miles W of Chipley, Holmes County, Florida.
5. *A. hallenbeckii* Lea. (a) 1 collection of 5 animals: 24 October 1963. Patsaliga Creek at Federal Highway 331, N edge of Luverne, Crenshaw County, Alabama. (b) 1 collection of 4 animals: 25 October 1963. Big Swamp Creek at State Highway 21, 1 mile SW of Hayneville, Lowndes County, Alabama. (c) 1 collection of 7 animals: 27 October 1963. Big Swamp Creek at Federal Highway 80, Lowndes County, Alabama. (d) 1 collection of 6 animals: 28 November 1963. Catoma Creek, 6 miles SW of Pike Road (village), Montgomery County, Alabama. (e) 1 collection of 9 animals: 1 December 1963. Cubahatchee Creek at Federal Highway 80, about 20 miles W of Tuskegee, Macon County, Alabama.

Subgenus *Utterbackia* F. C. Baker

6. *A. couperiana* Lea. (a) 6 bimonthly collections in 1965 (total of 372 individuals surveyed for gravidity; 10 per collection were sectioned). Myakka River at Myakka River State Park, about 18 miles SE of Sarasota, Sarasota County, Florida. (b) 2 collections (total of 16 animals): 16 August and 3 November 1968. Apalachicola River at Ocheese Landing, about 6 miles N of Blountstown, Calhoun County, Florida.
7. *A. imbecilis* Say. (a) 9 collections (total of 70 individuals): monthly, except in January, April, June and August, 1964-1965. Lake Talquin (= reservoir of the Ochlockonee River) at Coe's Landing, about 13 miles W of Tallahassee, Leon County, Florida. (b) 5 collections (total of 41 animals): in March, May, June, July and Novem-

ber 1965. Gantt Lake (= reservoir of the Conecuh River) at Federal Highway 29, Clearview, Covington County, Alabama.

8. *A. peggyae* Johnson. (a) 12 monthly collections (total of 124 animals), 1963-1964. Lake Talquin (see under *A. imbecilis*, above), the type-locality. (b) 12 monthly collections (total of 102 individuals), 1964-1965. Holmes Creek (see under *A. gibbosa* above).

METHODS

One of the populations of *A. couperiana* was sampled every 2 months, and attempts were made to obtain animals of *A. gibbosa*, *A. imbecilis* and *A. peggyae* (both populations) each month for a calendar year in order to provide material for the study of seasonal aspects of reproduction.

All animals were narcotized with 10% sodium nembutal (= Diabotal), fixed either in Bouin's fluid or 10% formalin, and preserved in 70% alcohol or 1% propylene phenoxetol. Wedge-shaped pieces of the visceral mass containing the gonads were removed, dehydrated through an alcohol series, cleared in xylol and embedded in paraffin. Sections were cut at a thickness of 10 μ m, stained in Harris' hematoxylin and counterstained with alcoholic eosin, and mounted with Canada balsam. Samples of the outer and inner demibranchs of each animal received the same treatment, but were immersed in a 1% solution of hydrochloric acid in 70% alcohol prior to embedding in order to decalcify any shelled larvae present. Gonadal sections were cut parasagittally from the outside inward so as to survey the width of the gonads for possible regional separation of the ovarian and testicular tissues in any hermaphrodites present. The demibranch sections were cut frontally from the axis (dorsal) downward, also to provide for examination of any possible regional variation. Five slides of each gonadal and demibranch sample were prepared, each containing sections from different locations.

The total length, height and width of each shell was measured (cf. Cvanara, 1963), and the occurrence of gravidity was recorded. The age of each individual was determined by counting the number of annuli on a single valve (see Chamberlain,

1931; Crowley, 1957; Ökland, 1963; Stansbery, 1967).

The incubating young in each gravid animal (about 3,000,000 in European species of *Anodonta*, *vide* Bouillon, 1955) were classified according to their comparative state of development. The seasonal occurrences of the general morphogenic stages (described on p. 96-97) were employed in determining the period of fertilization, the duration of incubation, the period of larval discharge, and the number of broods per year.

VISCERAL SEX

The sex of most unionids can be determined by the morphology of the outer demibranchs (see p. 87-88), or more precisely by the nature of the gonads⁴. The "visceral sex," defined by the kinds of gonads present, can be male, female or hermaphroditic. Coe (1943) arranged hermaphroditic conditions in pelecypods into 4 categories, principally according to the sequence of reproductive events (Table 2). In addition, monoecious unionids can be grouped as "♂ hermaphrodites" and "♀ hermaphrodites," depending in part on the relative preponderance of one type of

gonadal tissue over the other, and in part on the morphology of the outer demibranchs (Table 2).

In the present investigation, the determination of the visceral sex of each individual showed that *A. imbecilis* (both populations) contained ♀ hermaphrodites (Fig. 1) and females; males and ♂ hermaphrodites were lacking (Table 3). *A. californiensis* was largely dioecious, although one animal was a ♀ hermaphrodite. *A. hallenbeckii*, and *A. peggyae* (Figs. 2-4) in both populations, were principally dioecious, but both also contained ♂ hermaphrodites. Two populations of *A. couperiana* were investigated; that from the Myakka River possessed only males and females (hermaphrodites were absent), whereas that from the Apalachicola River had females, ♀ hermaphrodites and ♂ hermaphrodites (males were lacking). *A. gibbosa* was entirely dioecious. *A. corpulenta* and *A. wahlamatisensis* also lacked hermaphrodites, but the number of specimens (14 and 7, respectively) from a single sample each was small, and possibly rare monoecious animals may have escaped collection. Indeed, Van der Schalie (1970) previously reported the occurrence of hermaphrodites in *A. corpulenta*.

That *A. imbecilis* was found to contain females in addition to [♀] hermaphrodites is

⁴*Anodonta cygnea* (Linnaeus), the type-species of the genus, is the only reported unionid in which the outer demibranch morphology and visceral sex may not always coincide (cf. Bloomer, 1934; p. 87 here).

TABLE 2. Classifications of pelecypod hermaphroditism (types 1-4 after Coe, 1943; types 5-6 used in this paper).

Type	Characteristics
1. Functional hermaphroditism a. normal ^{a,b} b. accidental or developmental ^{a,c}	1. Eggs and sperm produced simultaneously a. typically in monoecious species b. typically in dioecious species
2. Consecutive sexuality	2. Single sex-reversal, usually protandrous
3. Rhythmical sexuality	3. > 1 sex-reversal, usually protandrous
4. Alternative sexuality ^{a?}	4. Adults function seasonally as separate sexes; they may or may not reverse by the next reproductive season
5. ♂ hermaphroditism ^{a,b,d}	5. Predominance of testicular tissue; animals not gravid, with non-marsupial outer demibranch morphology
6. ♀ hermaphroditism ^{a,e}	6. Ovarian tissue slightly or greatly exceeding quantity of testicular tissue; animals with marsupial outer demibranch morphology, may become gravid

^aExamples known in the Unionidae, including *Anodonta* (type 4 in *A. cygnea*?).

^bSynonymous with "usual" and "dominant" *sensu* Van der Schalie (1966, 1970).

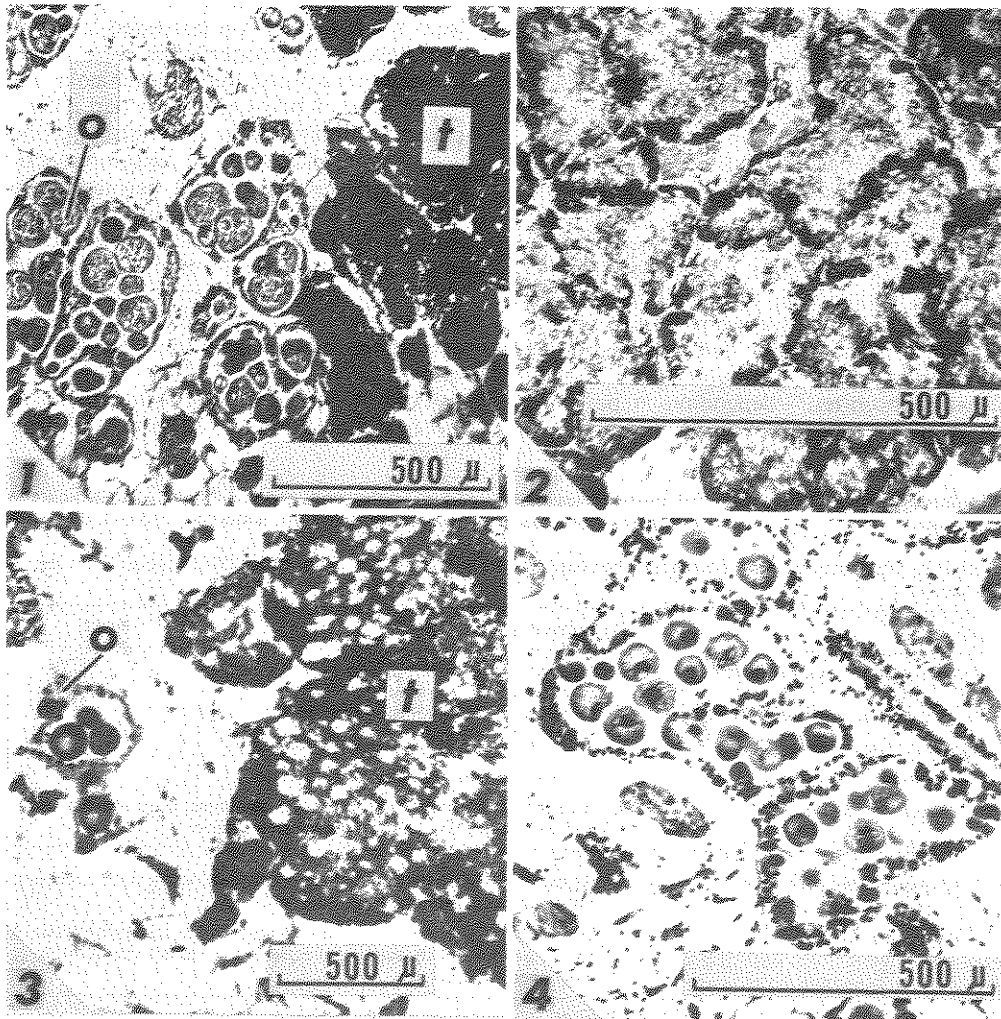
^cSynonymous with "occasional," "partial" and "sporadic" *sensu* Van der Schalie (1966, 1970).

^dSynonymous with accidental or developmental hermaphroditism (1.b.) in *Anodonta* spp.

^eSynonymous with normal hermaphroditism (1.a.) in *A. imbecilis*, and with accidental or developmental hermaphroditism (1.b.) in other species of *Anodonta*.

in disagreement with the view, prompted by Sterki (1898a, 1898b) and perpetuated since by others (e.g., Ortmann, 1910a, 1911; Utterback, 1915; Allen, 1924; Baker, 1927, 1928; Van der Schalie, 1966, 1970), that this species is "normally" or "dominantly" hermaphroditic. Although Utterback considered that to be the case, he inconsistently listed data on males and females. Van der Schalie (1966) originally suggested that "...*Anodonta imbecillis* [sic] had representatives in Florida which were dioecious, indicating that such a wide ranging species

may not be consistently hermaphroditic." These dioecious animals from the Hillsborough River in Hillsborough County, Florida, seen by me, actually belonged to *A. peggyae*. Van der Schalie (1970: 98) later stated that "...the difference in the sexual condition observed between the northern [Michigan] and southern [Florida] forms [of *A. "imbecillis"*], thought to be one of geographical strain, might be specific, a moot point." However, characteristic shell features, a distinctive geographical range and the different sexual composition support the



FIGS. 1-4. Histological sections of different visceral sexes in *Anodonta*. FIG. 1. ♀ hermaphrodite of *A. imbecillis* (gravid); Lake Talquin, 20 July 1963. FIG. 2. ♂ *A. peggyae*; Lake Talquin, 18 Jan. 1964. FIG. 3. ♂ hermaphrodite of *A. peggyae* (non-gravid); Holmes Creek, 24 Dec. 1964. FIG. 4. ♀ *A. peggyae* (non-gravid); Lake Talquin, 20 July 1963. o, ovarian tissue; t, testicular tissue.

TABLE 3. Numbers of animals according to their observed visceral sex. The numbers in parentheses represent gravid animals.

Species	Males	Hermaphrodites		Females	Total no. examined
		♂	♀		
<i>A. californiensis</i>	5		1(0)	1(1)	7
<i>A. corpulenta</i>	7			7(4)	14
<i>A. couperiana</i>					
Myakka River	22			38(5)	60
Apalachicola River		5(0)	1(0)	10(0)	16
<i>A. gibbosa</i>	35			18(12)	53
<i>A. hallenbeckii</i>	10	1(0) ^a		20(18)	31
<i>A. imbecilis</i>					
Lake Talquin			34(12)	36(10)	70
Gantt Lake			24(9)	17(6)	41
<i>A. peggyae</i>					
Lake Talquin	80	1(0)		42(24)	123 ^b
Holmes Creek	45	10(0)		47(38)	102
<i>A. wahlamatensis</i>	4			3(3)	7

^aWith 2 males and 3 females from Catoma Creek, Alabama.

^bOne animal, with 2 annuli on each valve, was sexually undifferentiated.

systematic validity of *A. peggyae*. Furthermore, through disc electrophoretic studies, Dr. John B. Burch of the University of Michigan (personal communication) found only a 75% similarity between foot muscle proteins of *A. imbecilis* from Michigan and *A. peggyae* from Florida (Lake Talquin topotypes).

DEMIBRANCH MORPHOLOGY

In describing anatomical features of several species of *Anodonta*, Ortmann (1911) contrasted the morphological organization common to all 4 demibranchs of males and the inner demibranchs of females with that of the outer demibranchs of females. Only the latter, which carry the incubating young and are therefore called "marsupial demibranchs," were stated to possess tripartite water-tubes; the non-marsupial demibranchs lacked this divided system. These observations, previously briefly noted by Ortmann (1910a, 1910b), were confirmed in the present study (Figs. 5-6), further refuting the opinion of Lefevre & Curtis (1910b) that such divided water-tubes do not exist.

It was also found here that as the marsupial demibranchs became laterally distended with near-infective glochidia, the secondary water-tubes united with the primary water-tubes (see Figs. 8-10, in a

seasonal series). This finding is in contrast to Ortmann's (1911) report that the secondary water-tubes vanished *after* the discharge of the glochidia, and that only the primary, central water-tubes served as ovisacs.

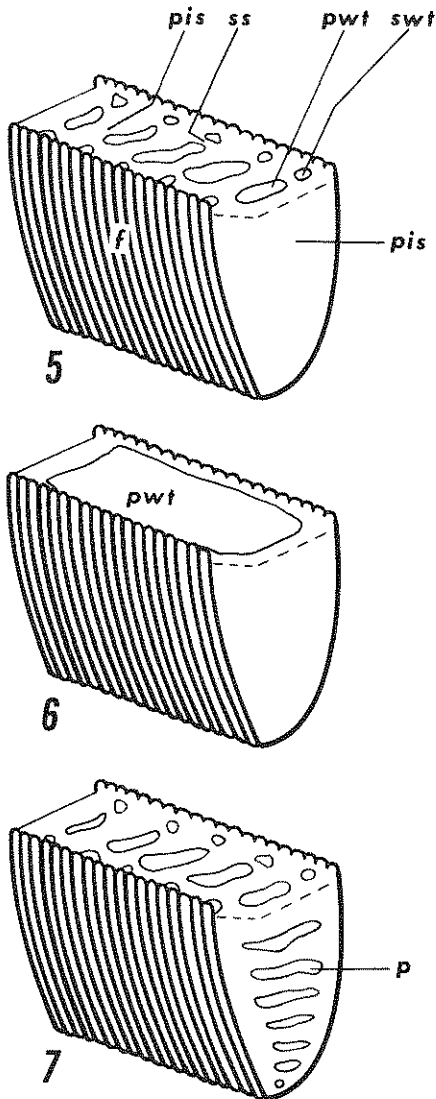
Ortmann (1911, 1912) and Bloomer (1934, 1936) noted that, in contrast to their more distant spacing in non-marsupial demibranchs, the primary interlamellar septa of marsupial demibranchs were comparatively close together. Indeed, Utterback (1915), among others, referred to the marsupial demibranchs as having "crowded septa." This dimorphism, apparently first detected by Peck (1877: shown in illustrations, but not described in the text), was confirmed here by determining the number of filaments between consecutive primary interlamellar septa: greater in non-marsupial demibranchs (Table 4; also compare Figs. 8 and 11).

Bloomer (1934, 1935, 1939) reported various hermaphroditic conditions in *A. cygnea*. At opposite ends of his monoecious series (visceral sex criterion) were forms in which the outer demibranchs were marsupial, and forms in which these demibranchs were non-marsupial⁵. The possible significance of these findings is discussed in the section on Sex-Reversal (p. 89).

Two kinds of monoecious animals were encountered in the present study: ♂ hermaphrodites and ♀ hermaphrodites, both initially identified by the visceral sex criterion (p. 85; Tables 2-3). None of the ♂ herma-

⁵Bloomer's distinction of the type of outer demibranchs present was based on septal spacing. Identification by the presence or absence of tripartite organization was not made.

phrodites were gravid; both non-gravid and gravid ♀ hermaphrodites were found, the non-gravid condition presumably reflecting age or seasonal variations in animals that probably were previously gravid or would



FIGS. 5-7. Diagrammatic representations of anodontine demibranch morphologies. FIG. 5. Tripartite marsupial organization in *Anodonta*. FIG. 6. Non-marsupial organization in *Anodonta* and *Strophitus*. FIG. 7. Tripartite marsupial organization in *Strophitus rugosus* (Zukey Lake inlet, Livingston Co., Michigan, U.S.A.). f, filaments; p, perforation; pis, primary interlamellar septum; pwt, primary water-tube; ss, secondary septum; swt, secondary water-tube.

have subsequently become gravid. All animals of *A. imbecilis*, ♀ hermaphrodites as well as females, possessed marsupial outer demibranchs (tripartite; crowded septa) and non-marsupial inner demibranchs (non-tripartite; distant septa) (Table 4). The single non-gravid ♀ hermaphrodite of *A. californiensis* had marsupial outer demibranchs. Males and females of *A. couperiana* from the Myakka River population had non-marsupial and marsupial outer demibranchs, respectively; the ♂ hermaphrodites from the Apalachicola River population had non-marsupial outer demibranchs, and although the single non-gravid ♀ hermaphrodite was discarded before a sample could be obtained it is hypothesized that such animals would have marsupial outer demibranchs as did the females. In *A. hallenbeckii* and *A. peggysæ* only the females had marsupial outer demibranchs; the ♂ hermaphrodites and males had 4 non-marsupial demibranchs. The demibranchs of *A. corpulenta* and *A. gibbosa* were not examined histologically, and it is only assumed that the males and females had non-marsupial and marsupial outer demibranchs, respectively; no hermaphrodites were found among the animals of either species. The inner demibranchs were consistently non-marsupial in all males, ♂ hermaphrodites, ♀ hermaphrodites and females of each species.

Van der Schalie's (1970) report on hermaphroditism in unionacean mussels was concerned only with the visceral sex condition, and no reference to the outer demibranch morphology of the monoecious animals was made. However, the legends to his photomicrographs of gonad sections of hermaphrodites often contain information on the relative proportions of ovarian and testicular tissue, and usually a notation about whether or not the animal was gravid, and some deductions can be made from his findings. For example, gravid hermaphrodites of *Anodonta corpulenta* and *Lasmigona complanata* were recorded, the latter "mostly female but with scattered spermatogenesis," both animals were probably ♀ hermaphrodites. Also listed were non-gravid hermaphrodites of *Alasmidonta marginata*, one of which was "evidently a female with patches of sperm," and *Anodonta grandis* form *footiana* that was "mostly male with only small amount of female tissue," the former may have been a ♀ hermaphrodite, and the latter a ♂ hermaphrodite. Van der Schalie consider-

TABLE 4. Dimorphism between the spacing of primary interlamellar septa in marsupial (tripartite) outer demibranchs and that in non-marsupial outer demibranchs of animals of different sexes in 6 species of *Anodonta*; based on numbers of filaments between consecutive interlamellar septa. Values for the inner, non-marsupial demibranchs of individuals of all visceral sex conditions were similar to those for the non-marsupial outer demibranchs. g, gravid animals; N, number of water-tubes examined; ng, non-gravid animals.

Species	Visceral sex	Tripartite organization	N	No. of filaments	
				Range	Mean
<i>A. californiensis</i>	male	no	57	6-20	12.3
	female; g	yes	15	4-9	6.4
	♀ hermaphrodite; ng	yes	10	2-8	4.3
<i>A. couperiana</i> Apalachicola River	female; ng	yes	10	1-4	2.5
	♀ hermaphrodite; ng	?	—	—	—
	♂ hermaphrodite; ng	no	10	10-18	12.7
Myakka River	male	no	20	10-30	17.9
	female; ng	yes	12	2-4	2.6
	female; g	yes	12	3-6	4.5
<i>A. hallenbeckii</i>	male	no	18	14-27	19.1
	♂ hermaphrodite; ng	no	5	17-25	19.2
	female; g	yes	15	2-6	4.1
<i>A. imbecilis</i> (all Lake Taiquin)	female; g	yes	27	1-6	2.4
	female; ng	yes	10	1	1.0
	♀ hermaphrodite; g	yes	50	1-8	3.7
<i>A. peggyae</i> Holmes Creek	male	no	25	8-30	17.8
	♂ hermaphrodite; ng	no	20	8-22	14.1
	female; g	yes	24	3-7	5.0
	female; ng	yes	8	3-6	4.9
Lake Taiquin	male	no	15	10-20	16.4
	female; g	yes	26	2-10	5.5
	female; ng	yes	23	2-9	3.7
<i>A. wahlamatisensis</i>	male	no	43	6-20	9.6
	female; g	yes	45	2-11	5.8

*The soft-parts of the single animal were discarded before a sample could be taken.

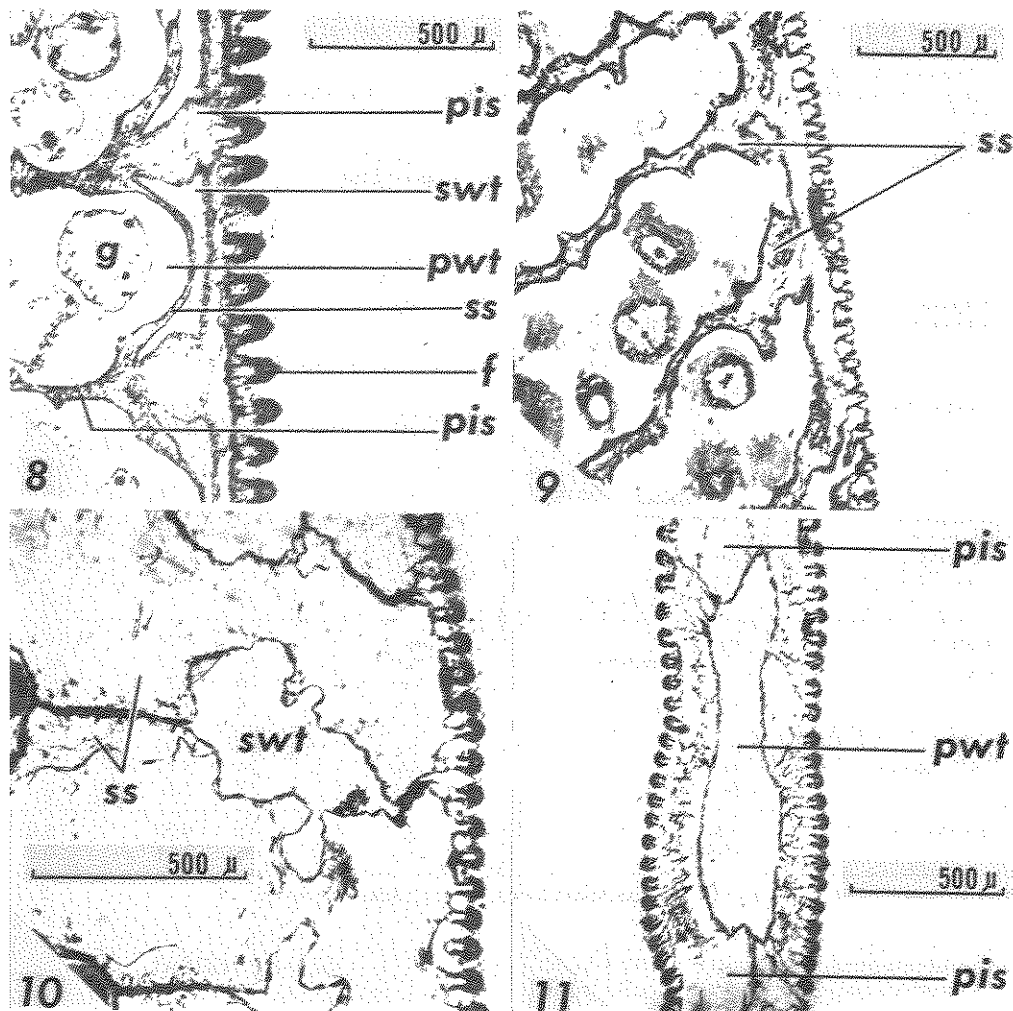
ed the monoecious individuals of these 4 taxa, and that of *Strophitus rugosus* ("mostly male with only a small amount of female tissue," but gravidity uncertain; perhaps a ♂ hermaphrodite), to be "occasional" hermaphrodites among largely dioecious samples. He listed *Anodonta imbecilis*, *Lasmigona compressa* and *L. subviridis*, as well as *Carunculina parva* (Barnes) [Unionidae: Lampsilinae], as the only species "in which hermaphroditism is the dominant condition"⁶.

SEX-REVERSAL

Bloomer (1934) found that in *A. cygnea* the morphology of an individual's outer demibranchs was not always correlated with its visceral sex⁷. Although all females evidently had marsupial outer demibranchs, only some males had non-marsupial outer demibranchs; these demibranchs were marsupial in some individuals called males. The outer demibranchs in hermaphrodites were either marsupial (gravid), non-marsupial

⁶Among other unionaceans, no species of the Amblemidae is known to be at least predominantly monoecious. And although Van der Schalie (1966) reported just "occasional hermaphrodites" in the margaritifera *Cumberlandia monodonta* (Say) [not cited by Van der Schalie, 1970] and *Margaritifera margaritifera* (Linnaeus), Heard (1970) found only hermaphrodites (visceral sex criterion) in a collection of 26 animals of *M. falcata* (Gould).

⁷Bloomer made several histological sections of gonad samples of *A. cygnea*, but most of his findings on the visceral sex of individuals were based on smear preparations. The latter is a less reliable technique for determining the presence of a hermaphroditic state, and especially for assessing the comparative abundance of ovarian and testicular tissues.



FIGS. 8-11. Frontal sections of *Anodonta* demibranchs showing the occurrence (FIG. 8), subsequent rupture (FIG. 9) and reappearance (FIG. 10) of secondary septa and secondary water-tubes in marsupial demibranchs, and the constant, non-tripartite organization of non-marsupial demibranchs (FIG. 11). FIG. 8. ♀ hermaphrodite of *A. imbecilis*; Lake Talquin, 20 July 1963. FIG. 9. ♀ *A. peggyae*; Lake Talquin, 16 May 1964. FIG. 10. ♀ *A. peggyae*; Holmes Creek, 15 May 1965. FIG. 11. ♂ hermaphrodite of *A. peggyae*; Lake Talquin, 18 Jan. 1964. g, developing glochidium; other abbreviations as in FIGS. 5-7.

(non-gravid) or "intermediate" (non-gravid), and Bloomer concluded that sex-reversal had occurred in this species.

Bloomer did not determine the ages of the specimens that he investigated, but he did record the total lengths of the shells. It was therefore possible to survey his data for indirect evidence of sex change or reversal with growth, an index of age. It appears that

if sex-reversal did occur in Bloomer's animals, at least in some cases, it was independent of age; hermaphrodites in evidently varying stages and kinds, males and females of all sizes were reported⁸, although most hermaphrodites were of larger size. In addition, Bloomer's (1946) observations on the seasonal sequence of events in the production and occurrence of sperm-morulae (see

⁸Such a condition might be expected in cases where more than one sex-reversal occurs in the life of an individual (see "Rhythmical sexuality" in Table 2).

below) and mature spermatozoa suggest that the possible sex-reversal is independent of seasonal influence.

Another aspect to be considered in examining the possibility of sex-reversal in *A. cygnea* is the occurrence of different sex compositions (i.e., associations of members of the different kinds of visceral sex conditions, apart from sex ratios) in different populations. Bloomer (1939) found the following associations, each characteristic of a separate population: males, females and hermaphrodites; males and hermaphrodites; females and hermaphrodites; and hermaphrodites only. Males and females never occurred together without hermaphrodites, and hermaphrodites were present in all populations. These relationships were apparently constant through at least several consecutive seasons; it is unknown whether the ratios changed with time. In the present study, *A. imbecillis* and *A. peggyae* showed no intraspecific variation in sex composition of the populations, although the ratios of the kinds of animals were different. However, a considerable difference in the sexual associations occurred between the 2 populations of *A. couperiana* (cf. Table 3). Van der Schalie's (1966, 1970) implication that *A. peggyae* may be a southern race of *A. imbecillis* is considered to be erroneous, although it may be that the northern (hermaphroditic only?) and southern (females and ♀ hermaphrodites) populations of *A. imbecillis* vary in sexual composition.

If none of the animals of *A. cygnea* had undergone sex change(s), Bloomer's observations can be explained in either of 2 ways. (1) A population of relatively stable size, without significant emigration and immigration, might establish and maintain a characteristic sexual composition (regardless of possible changes in ratios), even through self-fertilization by hermaphrodites as concluded by Bloomer (1940, 1943)⁹. (2) It may be that *A. cygnea* contains males, ♂ hermaphrodites, ♀ hermaphrodites and females, with at least some, or even all, of these 4 kinds of individuals occurring in different populations.

The entire problem of the sexual nature of *A. cygnea* should be restudied, the hermaphroditic states described by Bloomer more

precisely defined, and the mechanism of sex-reversal (if present) identified. The latter is made difficult by the lack of knowledge of this mussel's fish host(s), nature of genetic sex-determination and population genetics, and further by the time involved: several years between glochidial metamorphosis into a juvenile and the first "breeding."

It was found in the present study that the visceral sex of all individuals was consistently correlated with the morphology of the outer demibranchs, regardless of the season and the age of the animals. In those hermaphrodites with marsupial outer demibranchs the amount of ovarian tissue slightly (*A. imbecillis*) or conspicuously (*A. californiensis*, and *A. couperiana*?) exceeded the amount of testicular tissue in the sections, whereas the mass of testicular tissue greatly predominated in those hermaphrodites with non-marsupial outer demibranchs (*A. couperiana*, *A. hallenbeckii* and *A. peggyae*). It is therefore concluded that sex-reversal did not occur in the populations investigated.

SEASONAL ASPECTS OF REPRODUCTION

Sperm-Morulae

Organelles termed "sperm-morulae," named for their structural resemblance to the morular stage of ontogenetic development, may be observed in the testicular tissue of male (Fig. 12), ♂ hermaphroditic and ♀ hermaphroditic unionaceans. These multinucleate structures have previously been reported to occur in the marine pelecypod *Mya arenaria* Linnaeus [Myidae] (Coe & Turner, 1938; Stroganova, 1963; Ropes & Stickney, 1965; Shaw, 1965), as well as in the unionids *Anodonta anatina* (Bloomer, 1936; Stroganova, 1963), *A. cygnea* (Bloomer, 1930-1946), *A. grandis* form *footiana* (Van der Schalie & Locke, 1941), and *Unio pictorum* (Linnaeus) and *U. tumidus* (Philipsson) (both Stroganova, 1963). They are of widespread occurrence in the Unionacea, having been found in 43 species of 17 genera in a survey of the families Amblemidae, Hyriidae, Margaritiferidae and Unionidae (Heard, unpublished).

⁹On the other hand, the same concept might apply toward describing a characteristic sex composition for a population should at least some of the animals reverse their sexual condition at a constant rate, possibly with selection for or against one or more kinds of visceral sex if an imbalanced rate is present.

Sperm-morulae are seasonal in occurrence, being least abundant during the period of "typical" spermatogenesis¹⁰ (Ropes & Stickney, 1965; confirmed here). Their origin has been attributed to "atypical" spermatogenesis (Coe & Turner, 1938; Stroganova, 1963; Ropes & Stickney, 1965), although there has been disagreement as to their fate. Coe & Turner (1938) reported that most sperm-morulae in *Mya arenaria* underwent cytolysis to provide nutrient supplies for the ensuing "typical" spermatogenesis, whereas some metamorphosed into mature spermatozoa; both conclusions were based on circumstantial evidence, and neither was documented. Bloomer (1946) also concluded that sperm-morulae in *Anodonta cygnea* generate mature spermatozoa, but without providing direct evidence.

Precise details of the origin, development, morphology and function of sperm-morulae are not yet clearly understood, although electron microscopic and cytochemical studies on *Villosa villosa* (Wright) [Unionidae: Lampsilinae] and *Anodonta peggyae* suggest that they do not undergo cytolysis in these species but instead complete metamorphosis to provide spermatozoa that are indistinguishable in morphology and size from those generated in the "typical" spermatogenic pathway, i.e., that containing "typical" meiotic figures (Heard & Thomas, in preparation). Nevertheless, the male germinal cycle in *A. couperiana* and *A. gibbosa* was found here to be seasonally different from that in *A. imbecilis* and *A. peggyae* (see next section), and because the former 2 species contained many more sperm-morulae than did the latter 2, possibly not all of these structures generated mature spermatozoa. Whether or not the spermatozoa derived from sperm-morulae are viable is unknown at this time.

Spermatogenesis

According to Bloomer (1946), animals of some English populations of *A. cygnea* contained comparatively few sperm-morulae and spermatozoa from January to May, showed a decline in the numbers of spermatozoa in May and June, underwent "develop-

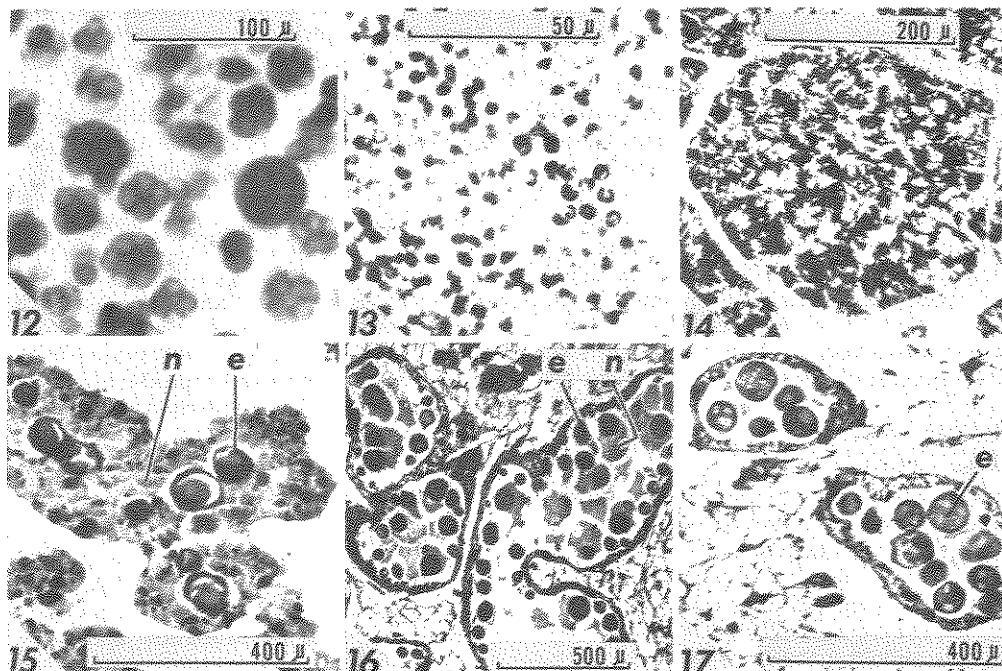
ment" of sperm-morulae in June and July, and contained large numbers of spermatozoa in July and August. The period of occurrence of "typical" spermatogenesis was not indicated. Bloomer (1940, 1943) previously stated that the release of spermatozoa was stimulated by direct sunlight and warmer water temperatures.

Possible interspecific and intraspecific seasonal variation in the occurrence of sperm-morulae (Fig. 12), typical spermatogenesis (Fig. 13) and mature spermatozoa (Fig. 14) was investigated here among 4 species: *A. couperiana*, *A. gibbosa*, *A. imbecilis* and *A. peggyae* (both populations). Although the cells of these 3 events were not absolutely mutually exclusive, they often appeared in distinct peak periods of abundance that were in seasonal sequence (see Table 5).

The "typical" spermatogenesis in *A. couperiana* and *A. gibbosa* was found to be confined to August and September, and spermatozoa appeared only in September. Sperm-morulae were predominant at all other times, usually alone filling the testicular tissue. In contrast, *A. imbecilis* and *A. peggyae* each showed considerable typical spermatogenesis and numerous spermatozoa throughout the year, with animals of both species containing many fewer sperm-morulae than occurred in *A. couperiana* and *A. gibbosa*. However, the sperm-morulae, also present throughout the year, were observed to be more common in males and ♂ hermaphrodites of *A. peggyae* (both populations) than in the ♀ hermaphrodites of *A. imbecilis*.

No seasonal variation in the occurrences of sperm-morulae was detected between males (total of 125 specimens from both populations) and ♂ hermaphrodites (total of 11) of *A. peggyae*, although both kinds of animals from the Lake Talquin population consistently contained more sperm-morulae than did those from Holmes Creek. The Lake Talquin animals displayed 2 periods of typical spermatogenic, spermatozoan and sperm-morular abundance, whereas those from Holmes Creek showed only one (Table 5). The 2 peak periods of typical spermatogenesis in Lake Talquin animals occurred

¹⁰Typical and atypical spermatogenic pathways, giving rise to 2 different kinds of spermatozoa (viz., eupyrene, and oligopyrene or apyrene, respectively) that are functionally and usually morphologically distinct from each other, are known to occur simultaneously in most prosobranch gastropods (cf. Nishiwaki, 1964).



FIGS. 12-17. States of gonad activity in *Anodonta*. FIG. 12. Sperm-morulae in a ♂ *A. couperiana*; Myakka River, 30 Jan. 1965. FIG. 13. Typical spermatogenesis in a non-gravid ♂ hermaphrodite of *A. couperiana*; Apalachicola River, 16 August 1968. FIG. 14. Mature spermatozoa in a ♂ *A. gibbosa*; Holmes Creek, 21 Sept. 1963. FIG. 15. Early oögenesis in a non-gravid ♀ *A. couperiana*; Myakka River, 27 March 1965. FIG. 16. Intermediate oögenesis in a gravid ♀ hermaphrodite of *A. imbecilis*; Lake Talquin, 20 July 1963. FIG. 17. Mature eggs in a gravid ♀ *A. peggyae*; Lake Talquin, 20 July 1963. e, egg; n, nutrient matter.

TABLE 5. Seasonal events in the male germinal cycle in 4 species of *Anodonta* from North America.

Species	Locality	Peak occurrences		
		Typical spermatogenesis	Mature spermatozoa	Sperm-morulae
<i>A. couperiana</i>	Myakka River	Aug.—Sept.	Sept.	Dec.—July
<i>A. gibbosa</i>	Holmes Creek	Aug.	Sept.	Oct.—June
<i>A. imbecilis</i>	Lake Talquin	June—Nov.	year-around	irregular
<i>A. peggyae</i>	Lake Talquin	March—April,	May—June,	July—Aug.,
	Lake Talquin	July—Sept.	Oct.—Feb.	irregular
	Holmes Creek	April—Sept.	Oct.—Feb.	irregular

within the more extensive single period in Holmes Creek animals, but only the second period of spermatozoan and sperm-morular abundance in Lake Talquin coincided with the single one in Holmes Creek. The bimodal activity exhibited within the Lake Talquin population was not associated with differences in the ages of the animals, and was not related to differences between males (80 specimens) and ♂ hermaphrodites (only 1); it instead reflected 2 consecutive breeding cycles per year (see p. 96).

Bloomer's (1946) inference that in *A. cygnea* the sperm-morulae metamorphosed into mature spermatozoa was based on the observed disappearance of sperm-morulae just prior to the appearance of large numbers of spermatozoa (temporal occurrence of typical spermatogenesis not cited). This sequence was not observed here in *A. couperiana* and *A. gibbosa* (cf. Table 5). The evidence is less clear in *A. imbecilis* and *A. peggyae*, but it may be that in the animals of these 2 species the comparatively few

sperm-morulae found rapidly metamorphosed into spermatozoa independently of the season.

A comparison of the seasonal male germinal cycle in *A. gibbosa* and *A. peggyae* from Holmes Creek, and in *A. imbecilis* and *A. peggyae* from Lake Talquin, showed that these congeneric species were not simultaneously and identically synchronized with, or influenced by, environmental influence on gonad activity. The cycle in Holmes Creek was much more seasonally limited in *A. gibbosa* than in *A. peggyae*, and in Lake Talquin there was 1 cycle per year in *A. imbecilis* but 2 cycles per year in *A. peggyae*.

Oögenesis

The comparative state of ovarian activity was classified into several arbitrary categories: spent or seasonally inactive, early oögenesis (a few oögonia and much nutritive matter; Fig. 15), "intermediate" oögenesis (primary oöcytes and some nutritive matter; Fig. 16) and mature eggs¹¹ (primary oöcytes [?] only, nutritive matter vanished; Fig. 17).

Anodonta couperiana females from the Myakka River were inactive in January, showed early oögenesis in March, May and July, displayed intermediate oögenesis and mature eggs in September, and were spent in November. In the Apalachicola River population, females and the single ♀ hermaphrodite showed early oögenesis in early August; the females were spent in mid-November.

No females of *A. gibbosa* were found between mid-May and early October, and only inactive ovarian tissue occurred in the samples from the remaining months. The absence of mature eggs for most of the year coincided with the preponderance of sperm-morulae and lack of spermatozoa for much of that period.

In *A. imbecilis* from Lake Talquin, only the intermediate oögenic stage and mature eggs were found. Each occurred throughout the year in the ovarian tissue of both females and ♀ hermaphrodites, although there appeared to be a slight preponderance of mature eggs from December into March in

members of the 2 sexual conditions. The Gantt Lake animals, available in fewer collections, coincided in ovarian activity with those from Lake Talquin.

Based on the seasonal sequence of spermatogenesis and larval development (see next section), it is concluded that Lake Talquin animals of *A. peggyae* underwent 2 reproductive cycles per year. However, although displaying only intermediate oögenesis and mature eggs throughout the year, females of that population showed a slightly greater activity in only 1 period: January into February. In *A. peggyae* from Holmes Creek, with just 1 reproductive cycle per year, intermediate oögenesis and mature eggs also occurred at all times; a slight peak in abundance appeared from late December into April.

The Incubation Period

Gravid animals in *Anodonta* have long been reported to exhibit a prolonged, bradyctictic period of incubation, and sometimes to show a non-gravid interval between consecutive, annual breeding cycles (cf. Table 6).

Anodonta cataracta Say (Conner, 1907; Ortmann, 1909, 1912), *A. corpulenta* (Surber, 1912), *A. grandis* Say (Baker, 1928; Van der Schalie, 1938) and *A. implicata* Say (Conner, 1909) were found to have, during the Nearctic summer, a short but distinct non-gravid interval between consecutive "breeding seasons." In comparison, Baker (1928) and Van der Schalie (1938) reported the occurrence of gravid animals of *A. imbecilis* throughout the year, this species apparently lacking a non-gravid interval. Non-gravid animals of *A. cygnea* were reported only in July (Bloomer, 1930-1939), and the data may not be complete. *A. anatina* was reported to be gravid in July through December in England (Bloomer, 1936), and either this species has a comparatively long non-gravid interval, or the observations are perhaps again incomplete.

All of these reports concern only the seasonal presence of gravid animals, and such information does not reveal the number of

¹¹Lillie (1901) demonstrated that in *Elliptio complanatus* (Dillwyn) [Unionidae: Pleurobeminae *sensu* Heard & Guckert (1971)] diploid primary oöcytes and not mature haploid ova were released from the ovaries, and that reduction-division of these cells did not occur until after sperm penetration (in the marsupial demibranchs). Because it is presently unknown whether this phenomenon is universal in the Unionidae, the most mature cells in the sections of ovarian tissue studied here will be referred to simply as "eggs."

TABLE 6. Known gravid periods in 13 species of *Anodonta* (from Baker, 1928; Bloomer, 1934, 1935, 1936; Conner, 1907, 1909; Ortmann, 1909; Surber, 1912; Van der Schalie, 1938; present observations).

Species	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
<i>A. anatina</i>							X	X	X	X		X
<i>A. californiensis</i>										X		
<i>A. cataracta</i>	X	X	X	X	X	(... interval ^a ...)			X	X	X	X
<i>A. corpulenta</i>	X	X	X	X		(..... interval ^a)				X	X	X
<i>A. couperiana</i>											X	
<i>A. cygnea</i>	X	X	X	X	X	X		X	X	X	X	X
<i>A. gibbosa</i>										X		X
<i>A. grandis</i>	X	X	X	X	(. . interval ^a . . .)			X	X	X	X	X
<i>A. hallenbeckii</i>										X	X	X
<i>A. imbecilis</i>	X	X	X	X	X	X	X	X	X	X	X	X
<i>A. implicata</i>	X		X	X	X	(... interval ^a ...)			X	X	X	X
<i>A. peggyae</i>	X	X	X	X	X	X	X	X	X	X	X	X
<i>A. wahlamatisensis</i>										X		

^aInterval, called "interim" by various authors, represents a non-gravid period between 2 consecutive reproductive cycles.

breeding cycles within a year. For instance, whereas *A. imbecilis* was found by Baker (1928) and Van der Schalie (1938) to be gravid throughout the year, Allen (1924) reported not just one but several reproductive cycles per year in that species.

In addition to recording the observed occurrences of gravid animals of *A. californiensis*, *A. corpulenta*, *A. couperiana*, *A. gibbosa*, *A. hallenbeckii*, *A. imbecilis*, *A. peggyae* and *A. wahlamatisensis* in the present study (cf. Table 6), the incubating young in each of these animals were identified according to their comparative state of development (morphological stages described in next section).

The presence of gravid animals of *A. couperiana* and *A. gibbosa* only in the fall and winter suggests a "winter-tachytictic" breeding season hitherto unreported in the Unionidae. The indication of such a phenomenon is more reliable from *A. couperiana* (52 gravid Myakka River animals, all in November, among a total of 372 specimens examined from all 6 collections) than from *A. gibbosa* (12 of only 18 females were gravid: October and December). However, all gravid animals of both species contained only mature larval stages; no embryos or earlier larvae were found, and the rates of development and the more precise duration of incubation are not accurately known. Nevertheless, the seasonally confined occurrences of mature spermatozoa in both species (cf. Table 5) point to an autumnal period of fertilization, and thus suggest a rapid rate of larval development.

The presence of gravid Lake Talquin animals of *A. imbecilis* in all collections confirmed Baker's (1928) and Van der Schalie's (1938) earlier reports. Embryos appeared in March to May, early larvae from June to September, intermediate larvae in September to December, and mature larvae in January and February. This seasonal sequence, alike in the fewer Gantt Lake collections, seems to refute (at least for these populations) Allen's (1924) claim of several very short breeding cycles per year in this species. Nevertheless, the peculiar phenomenon of large numbers of mature eggs and sperm throughout the year in Lake Talquin (cf. Table 5) suggests that, although these gametes may not be chemically/physiologically differentiated or viable at certain seasons in that population, these cells may be functional during several consecutive periods per year in other populations (perhaps especially in other latitudes, Allen's study having been conducted in Iowa). Individuals of these other populations might thus undergo more than one reproductive cycle in a year. Such variation was found here in *A. peggyae* (see below). Seasonal investigation of gravid and non-gravid females and ♀ hermaphrodites of *A. imbecilis* revealed that there were no significant variations in gravid periods and developmental stages of incubating young between the Lake Talquin and Gantt Lake populations. Similarly, it was found that once the age of sexual maturity¹² was reached there was no significant variation in these features between animals of different age classes, re-

¹²"Sexual maturity" is defined here in terms of age when first gravid, rather than in terms of gonad development and activity (see p. 92-94).

ardless of whether the individuals were females or ♀ hermaphrodites.

Anodonta peggyae was gravid throughout the year in both populations. However, whereas the Holmes Creek animals displayed 1 breeding cycle in the year, the Lake Talquin individuals had 2. The latter group showed these 2 cycles both in terms of seasonal spermatogenesis (cf. Table 5) and comparative stages of embryonic and larval development. Gravid females undergoing one cycle contained embryos in March, early larvae in April, intermediate larvae in May and June, and mature larvae in July and August; those in the subsequent cycle contained embryos in September and October, early and intermediate larvae in November and December, and mature larvae in January and February. One individual from the September collection contained both embryos and mature larvae; all other gravid animals, in all species, contained only one developmental stage at a time, and most animals from the same month carried the same stage. In contrast to the Lake Talquin population, the gravid Holmes Creek females undergoing a single breeding cycle in the year contained embryos in March into July, intermediate larvae in August into December, and mature larvae in January and February.

Although there were differences in the number of breeding cycles per year between the Holmes Creek and Lake Talquin populations of *A. peggyae*, breeding cycles may not be influenced entirely by such conspicuous environmental factors as water temperature and the presence or absence of current because there were also such differences between the Lake Talquin populations of *A. imbecilis*, with 1 cycle per year, and *A. peggyae* with 2 cycles per year (see Tables 5 and 6).

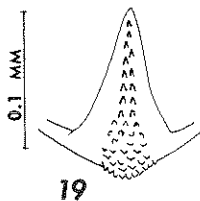
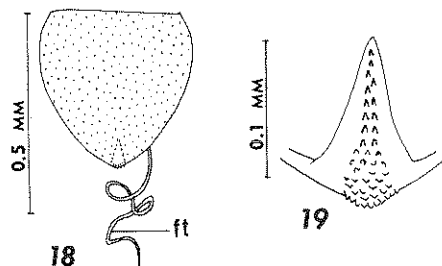
Seasonal observations were not possible for *A. californiensis*, *A. corpulenta*, *A. hallenbeckii* and *A. wahlamatisensis*, although the presence of gravid animals of these species is noted in Table 6. *Anodonta californiensis* and *A. wahlamatisensis* carried intermediate larvae in October, and *A. hallenbeckii* contained only mature larvae in late October, November and early December. The November animals of *A. corpulenta* were discarded before the development stage of incubating young could be made.

GLOCHIDIA

Glochidia, i.e., the mature and infective larvae, in *Anodonta* have been described and illustrated by Tucker (1927) and Bouillon (1955), among others, but most thoroughly by Inaba (1941). The subtriangular shell is punctated (Fig. 18), and the ventrally situated hooks bear numerous small spines (Fig. 19). Triangular, hooked glochidia are a feature of all species of the Anodontinae (Ortmann, 1912), although the prominent hooks are not a unique characteristic of this group of unionaceans, also occurring on the glochidia of some amblemids (at least the Indian species of *Indonaia* Prashad; cf. Prashad, 1918) and throughout the Hyriidae (Ortmann, 1921; McMichael & Hiscock, 1958; Parodiz & Bonetto, 1963), as well as in some species of the Unionidae: Unioninae s. s. (Ortmann, 1912, 1918). Another feature of the glochidia of many if not all species of *Anodonta* is the late appearance of a larval "thread" or "filament" (called a byssal thread by Bouillon, 1955); whether it occurs in other groups of the Anodontinae is not known.

The developmental continuum of incubating young observed in the present study was divided into several arbitrary stages in order to assist in determining the season of fertilization, the incubation period, the season of glochidial discharge and the number of broods generated per year. *Embryos* included those individuals from the zygote to the distinct appearance of the single adductor muscle, before the formation of the rudimentary shell; *early larvae* were characterized by the presence of a shell that lacked punctae and hooks; *intermediate larvae* possessed a punctated shell with only rudimentary hooks; and *mature larvae*, i.e., *glochidia*, displayed a punctated shell with well developed and spined hooks, as well as larval thread, and had broken free from the fertilization membrane that surrounded the individuals of the previous stages.

The seasonal observations indicate that in *A. couperiana* and *A. gibbosa* fertilization occurred in the late summer, and the short incubation period may not extend over the entire winter. In *A. imbecilis* and *A. peggyae* (Holmes Creek only, with 1 reproductive cycle per year), fertilization occurred in late winter, and the long incubation period lasted



FIGS. 18-19. Glochidial features of *Anodonta gibbosa*. FIG. 18. Mature larva, with filament-thread (ft) and punctated, subtriangular, ventrally hooked shell. FIG. 19. Ventral surface of spined hook.

until the following late winter. In the Lake Talquin population of *A. peggyae*, with 2 cycles per year, one fertilization occurred in the early spring, and incubation lasted until mid-summer; the second fertilization occurred in late summer, with incubation lasting until late winter. A search for mature glochidia in species of *Anodonta* in the southern United States should thus be made during the winter season, i.e., November-February.

In all species investigated here, the dorsal margin of the glochidial shell was straight, or nearly so, and its length was less than the total length which was found slightly above the middle of the valves (cf. Fig. 18). Inaba (1941) reported the sizes of the glochidial shells in *A. beringiana* Middendorff (0.296 mm total length \times 0.296 mm high), *A. japonica* Clessin (0.258 \times 0.232), *A. woodiana calipygos* Kobelt (0.298 \times 0.297), *A. woodiana lauta* von Martens (0.303 \times 0.268) and *A. "woodiana lauta tumens* Haas" (0.296 \times 0.258), and in most of these species the total length was greater than the height. That relationship was found here only in *A. hallenbeckii* (0.285 \times 0.266) and *A. peggyae* (0.261 \times 0.256); the height exceeded the total length in *A. couperiana* (0.294 \times 0.336), *A. gibbosa* (0.378 \times 0.399) and *A. imbecilis* (0.228 \times 0.233). The glochidia of *A. gibbosa* are the largest, and those of *A. imbecilis* the smallest, among those of all 10 taxa. The same relationship was found for the size of the hooks: 0.084 mm long in *A. couperiana*, 0.126 in *A. gibbosa*, 0.099 in *A. hallenbeckii*, 0.062 in *A. imbecilis* and 0.095 in *A. peggyae*.

¹³The spines, correctly depicted by Inaba (1941), were directed medially, and clearly do not act as barbs to anchor the glochidium against dislodging. Although they would make penetration of the hooks more difficult, they might serve to further lyse the host tissue to stimulate cyst formation, and perhaps initiate rapid supply of nutrients.

There was no variation in the relative number or location of the punctae, none in the size and number of spines on the hooks¹³, and apparently none in the nature of the larval thread among the glochidia of the different species studied here. Although the larval thread was loosely coiled, its comparative length could not be determined with confidence. There was also no difference in size or proportions between glochidia of the 2 populations of *A. peggyae*, and none between these larvae in females and ♀ hermaphrodites of both populations of *A. imbecilis*.

No metamorphosed glochidia, i.e., juveniles, were found in the marsupial demi-branches of *A. imbecilis* or the other species surveyed here.

AGE OF SEXUAL MATURITY AND LIFE SPAN

Van der Schalie & Locke (1941) concluded that animals of *A. grandis* form *footiana* did not become "sexually mature" until they had reached their second year of growth. In that instance, sexual maturity was defined in terms of gonad development while identifying the visceral sex of the animals.

In characterizing the Anodontinae, Stansbery (1967) reported that the mussels of this group "...are typically rapid growing, early maturing species having a relatively short life span. The extreme of this group in Lake Erie [Ohio, U.S.A.] is apparently *Anodonta imbecilis* Say, which matures in its second year but rarely lives to be 5 years of age." The criterion of maturity was not described in this example; the life span was estimated by counting the annuli on the shell.

Table 7 shows the age at the earliest observed sexual maturity (gravidity criterion) in animals of 15 populations representing 8 species. The number of annuli present in the youngest gravid animals ranged from 3 to 5 (4-6 years of age). This observation suggests that species of *Anodonta* mature at a greater age and state of growth than that reported by Stansbery (1967) for *A. imbecilis*. Although, as previously noted, there was no significant seasonal difference in the gravid periods of females and ♀

TABLE 7. Ages of youngest gravid animals and observed life span in 8 North American species of *Anodonta*. Age in years = number of annuli + 1.

Species	No. of annuli in youngest gravid	No. of annuli in oldest animals			
		Males	Hermaphrodites		Females
			♂	♀	
<i>A. californiensis</i>	4 ^a	4		2 ^b	4
<i>A. corpulenta</i>	5	7			7
<i>A. couperiana</i>					
Myakka River	4	9			12
<i>A. gibbosa</i>	5 ^c	15			15
<i>A. hallenbeckii</i>	4	13			15
<i>A. imbecilis</i>					
females					
Lake Talquin	5				12
Gantt Lake	4				8
hermaphrodites					
Lake Talquin	4			8	
Gantt Lake	3			9	
<i>A. peggyae</i>					
Lake Talquin	4	10	6		14
Holmes Creek	5	10	8		10
<i>A. wahlamatisensis</i>	8 ^d	9			10

^aThe only female in the collection.

^bNon-gravid.

^cCollections contained total of 2 non-gravid females with 4 annuli; none younger.

^dYoungest animal available was a male with 6 annuli.

hermaphrodites of *A. imbecilis*, the majority of the youngest gravid animals were monoecious.

Also listed in Table 7 are the observed maximum numbers of annuli on the shells of the animals examined here. These values for most species varied between 7 and 15 (8-16 years in age¹⁴); the shells of *A. californiensis* were much smaller (and younger?) than those in museum collections. Animals of the southern (Florida, Alabama) populations of *A. imbecilis* lived longer than the northern one (Ohio) described by Stansbery (1967). Whereas there was little difference in the observed greatest ages between ♀ hermaphrodites of both populations of *A. imbecilis*, the females from Lake Talquin apparently lived 4 years longer than the hermaphrodites (and 4 years longer than the Gantt Lake females). A similar relationship occurred in *A. peggyae*, with no difference in the maximum age attained by males and females in the Holmes Creek population, and none between males from Lake Talquin and Holmes Creek; Lake Talquin females lived 4 years longer than Holmes Creek females, and 4 years longer than the males of both populations. There was no difference in the greatest ages between males and females of *A. cali-*

forniensis, *A. corpulenta* and *A. gibbosa*, and but little difference between those of *A. wahlamatisensis*. Females survived longer than males in *A. couperiana* (by 3 years in the Myakka River) and *A. hallenbeckii* (by 2 years; composite values from all populations). In *A. peggyae* (both populations), the ♂ hermaphrodites were not as old as either the males or females. The ages of none of the animals of *A. couperiana* from the Apalachicola River could be determined with confidence, but there was no significant difference between the sizes of the females, ♀ hermaphrodite and ♂ hermaphrodites (males were lacking).

It must be noted, however, that relatively few younger and older animals were found during this study, and the data on earliest gravidity and greatest age may be incomplete. Furthermore, these features may not be species-specific, perhaps varying intra-specifically.

Even the oldest animals of all species displayed active gametogenesis (many with mature gametes, depending on the season), and most of the oldest females and ♀ hermaphrodites were gravid. These findings suggest the absence of a post-reproductive (or senility) period, mentioned by Stansbery

¹⁴Bouillon (1955) claimed a life span of 20-30 years in European species of *Anodonta*, none of which were identified by name.

(1967) as a feature of the Unionidae: Lampsilinae, as well as a potentially longer life span within the population.

SEXUAL DIMORPHISM OF THE SHELL

The possibility of sexual dimorphism of the shells of *Anodonta* was investigated by von Siebold (1837) and Weisensee (1916) for *A. cygnea* and by Hazay (1881) for *A. anatina*. It was concluded that females were significantly wider than males. Inasmuch as total length is often used as an index of size, Brummer (1932) and Brander (1954) employed the ratio of D:L (i.e., "diameter" [= width] to total length) in reporting dimorphic shell differences between males and females of *A. complanata* Rossmässler and *A. piscinalis* Nilsson. These investigations utilized size-frequency polygrams rather than statistical tests.

In the present study, mean sizes (based on width/total length $\times 100$) of shells of individuals of different age classes and sexual conditions in *A. couperiana*, *A. gibbosa*, *A. imbecilis* and *A. peggyae* were determined, and the *t*-test for correlated means (cf. Snedecor & Cochran, 1967) was employed to assess the values. At the 5% rejection level, there was no statistically significant difference in size between the males and females of *A. gibbosa*, and none between females and $\text{\textcircled{f}}$ hermaphrodites of *A. imbecilis* (Lake Talquin). Myakka River females of *A. couperiana* were significantly larger than males. The similarities and differences were constant for animals common to each age class; the same relationship was also found for *A. peggyae*.

In the Holmes Creek population of *A. peggyae*, there was no significant difference in size between males and females, and none between $\text{\textcircled{m}}$ hermaphrodites and females, although the $\text{\textcircled{m}}$ hermaphrodites were larger (though younger) than males. In contrast, Lake Talquin females were significantly larger than males. Also, Lake Talquin males and females were significantly larger than Holmes Creek males and females, respectively.

Although some sexual dimorphisms were found in the shells of *Anodonta*, none were of the conspicuous type well known in the Unionidae: Lampsilinae, in which the posterior part of the female shell is expanded in size and shape. Sexual dimorphism in

Anodonta shells can be determined only by measurement and statistical testing.

DISCUSSION OF HERMAPHRODITISM

Glandular Differentiation

Pelseneer (1895) listed 4 "morphological forms" of hermaphroditism: (1) gonads with monoecious acini in which both eggs and sperm are produced, (2) gonads with intermingled zones of male and female acini, (3) gonads with male and female acini in regionally distinct and separate zones, and (4) distinct and separate male and female gonads in the same individual. A survey of Van der Schalie's (1970) recent report, and the present findings, indicates that the majority of the known hermaphroditic unionids belong to the first 2 categories, and it suggests that most of the monoecious animals are $\text{\textcircled{f}}$ hermaphrodites (cf. Table 8).

The present study revealed that the $\text{\textcircled{m}}$ hermaphrodites of *A. couperiana*, *A. hal-lenbeckii* and *A. peggyae*, as well as the $\text{\textcircled{f}}$ hermaphrodites of *A. californiensis* and *A. couperiana*, were of Pelseneer's 2nd type, displaying intermingled dioecious acini. Only the $\text{\textcircled{f}}$ hermaphrodites of *A. imbecilis* showed the regional differentiation of the 3rd type. Truly monoecious acini, reported in *A. corpulenta* by Van der Schalie (1970), were not found here.

Although he did not find monoecious acini in *A. cygnea*, Bloomer (1934), in his concept of sex-reversal in this species, suggested that acini containing sperm-morulae would subsequently be occupied by eggs. Should this type of transformation be documented, *A. cygnea* would exhibit an "asynchronous successive hermaphroditism" (cf. Bacci, 1951; Portmann, 1960) which, according to Pelseneer (1894), is a more primitive condition than that of "functional hermaphroditism" in which eggs and sperm are produced simultaneously.

Van der Schalie (1969, 1970) noted that in most hermaphroditic unionids one type of gonadal tissue was in a more advanced state of development and activity than the other, and that functional hermaphroditism was observed only in *Actinonaias ellipsiformis*, *Carunculina parva*, *Villosa iris* (all Lampsilinae) and *Anodonta imbecilis*. Functional hermaphroditism was verified here for *A. imbecilis*, and it was found that the eggs in $\text{\textcircled{m}}$

TABLE 8. List of unionids displaying the monoecious states distinguished by Pelseener (1895). Data drawn from Van der Schalie (1969, 1970: legends to photomicrographs) and the present study.

1. Hermaphroditic acini	2. (continued)
* <i>Actinonaias ellipsiformis</i> (Conrad) ^a	* <i>Anodonta peggyae</i> Johnson ^b
* <i>Alasmidonta marginata</i> (Say) ^a	<i>Elliptio productus</i> (Conrad) ^b
* <i>Anodonta corpulenta</i> Cooper ^a	* <i>Lasmigona compressa</i> (Lea) ^a
<i>Carunculina parva</i> (Barnes) ^a	<i>Leptodea laevis</i> (Lea) ^a
<i>Elliptio dilatatus</i> (Rafinesque) ^a	<i>Pleurobema cordatum</i> (Rafinesque)
* <i>Lasmigona complanata</i> (Barnes) ^a	<i>Pleurobema c. coccineum</i> (Conrad) ^a
<i>Lampsilis cariosa</i> (Say)	<i>Proptera alata</i> (Say) ^a
<i>Ptychobranchus subtentum</i> (Say) ^a	* <i>Ptychobranchus fasciolaris</i> (Raf.) ^a
<i>Villosa iris</i> (Lea) ^a	* <i>Strophitus rugosus</i> (Swainson) ^b
2. Intermingled zones of ♂ and ♀ acini	3. Regionally distinct and separate ♂ and ♀ acini
* <i>Anodonta californiensis</i> Lea ^a	* <i>Anodonta imbecilis</i> Say ^a
* <i>Anodonta couperiana</i> Lea ^a + b	4. Separate ♂ and ♀ gonads in the same individual: none.
* <i>Anodonta grandis</i> f. <i>footiana</i> Lea ^b	
* <i>Anodonta hallenbeckii</i> Lea ^b	

^aGravid or non-gravid, but with principally ovarian tissue; known or probably ♀ hermaphrodites.

^bNon-gravid, and with principally testicular tissue; known or probable ♂ hermaphrodites.

*Members of the subfamily Anodontinae Ortmann.

hermaphrodites of *A. peggyae* were always smaller (= "poorly developed" or "suppressed" *sensu* Van der Schalie?) than those in females collected at the same time. However, observations on the monoecious animals of the other species is less conclusive. In *A. californiensis* and *A. couperiana* only sperm-morulae and early oögenesis were simultaneously present in the hermaphrodites, whereas in *A. hallenbeckii* both testicular and ovarian tissues were inactive in the single ♂ hermaphrodite.

Significance of Hermaphroditism

The known monoecious conditions in *Anodonta* do not coincide with conchological subgeneric groupings, although verification of these taxa as natural units is needed. However, the presence of hermaphroditism in various other unionaceans suggests that its occurrence, in its different forms, does not reflect phylogeny. Other explanations of the monoecious state have considered hermaphroditism to be an adaptation to environmental conditions.

Weisensee (1916) stated that in *A. cygnea* a dioecious relationship occurred in streams, whereas increased proportions of hermaphrodites appeared in standing waters. Unionids are, in general, stream- rather than lake-dwellers, and animals from lakes are typically smaller than animals of the same species from streams. A number of exceptions are known, however, including species of *Anodonta* which survive very well in standing water and not infrequently reach larger sizes in such environments.

Two populations of *A. peggyae* were studied here, one from a lotic environment (Holmes Creek) and one from a lentic environment (the Lake Talquin impoundment). The proportions of *male* hermaphrodites are in contrast to Weisensee's conclusion of ecological adaptation: the Lake Talquin population had a smaller proportion of monoecious animals (0.8%) than did the Holmes Creek population (9.8%). Both populations of *A. imbecilis* were from impoundments, and whether the proportion of the *female* hermaphrodites in a stream environment would be lower or higher than those observed here (48.5% in Lake Talquin; 58.5% in Gantt Lake) is not known.

Species of *Anodonta* have been referred to by Baker (1928) and others as "floaters" because these thin-shelled and thus comparatively light-weight animals can float in the water if an adequate amount of air becomes trapped between the valves. This phenomenon might be an adaptation to dispersal, especially if an individual was a ♀ hermaphrodite that had the capacity to self-fertilize and consequently serve as the beginning of a new population (assuming the presence of a suitable host for the glochidia). Nevertheless, such an event would not be advantageous unless it occurred in a stream.

According to Cole (1954:104), "Parthenogenesis, hermaphroditism and purely asexual reproduction may clearly offer some advantages under conditions that restrict the probability of contacts between the sexes." However, it may be noted that internal fertilization, even without copula-

tion, requires close proximity of members of a breeding population.

Ghiselin (1969), in reviewing hermaphroditism in parazoans and metazoans (freshwater mussels briefly noted on p. 98), stated that hermaphroditism "should" evolve under one or more of the following conditions: difficulty in finding a mate (for which a low density model was presented), beneficial size dimorphism between different sexes (size advantage model), and occurrence of small, genetically isolated populations (gene dispersal model). Too little information on unionid population dynamics is presently available for the use of any of these models in definitively interpreting the occurrence and nature of hermaphroditism in *Anodonta*.

Finally, Van der Schalie (1969) suggested genetic, hormonal or cytogenetic controls as reasons for the occurrence of hermaphroditism in its variable manifestations, but the nature of such possible mechanisms (if present) has yet to be investigated.

ACKNOWLEDGMENTS

I thank J. J. Landye for supplying animals of *A. corpulenta*, and D. W. Taylor for aid in obtaining *A. californiensis* and *A. wahlamatisensis*; R. H. Guckert and V. A. Vail helped in making several collections from Florida and Alabama. Invaluable assistance in the laboratory was provided by S. S. Hendrix and C. C. Swift. Richard I. Johnson kindly verified the identification of *A. couperiana* from the Apalachicola River.

LITERATURE CITED

- ALLEN, E., 1924, The existence of a short reproductive cycle in *Anodonta imbecilis*. *Biol. Bull.*, 46: 88-94.
- BACCI, G., 1951, Ermafroditismo ed intersessualità nei Gasteropodi e Lamellibranchi. *Arch. zool. ital.*, suppl. 7: 57-151.
- BAKER, F. C., 1927, On the division of the Sphaeriidae into two subfamilies; and the description of a new genus of Unionidae, with descriptions of new varieties. *Amer. Midl. Natur.*, 10: 220-223.
- BAKER, F. C., 1928, The fresh water Mollusca of Wisconsin. Part II. Pelecypoda. *Bull. Wis. Geol. Natur. Hist. Surv.*, 70: 1-495, pls. 29-105.
- BLOOMER, H. H., 1930, A note on the sex of *Anodonta cygnea*. *Proc. malacol. Soc. Lond.*, 19: 10-14.
- BLOOMER, H. H., 1934, On the sex, and sex-modification of the gill, of *Anodonta cygnea*. *Proc. malacol. Soc. Lond.*, 21: 21-28, pl. 3.
- BLOOMER, H. H., 1935, A further note on the sex of *Anodonta cygnea* L. *Proc. malacol. Soc. Lond.*, 21: 304-321.
- BLOOMER, H. H., 1936, A note on the sex of *Anodonta anatina*. *Proc. malacol. Soc. Lond.*, 22: 129-134, pl. 14.
- BLOOMER, H. H., 1939, A note on the sex of *Pseudanodonta* Bourguignat and *Anodonta* Lamarck. *Proc. malacol. Soc. Lond.*, 23: 285-297.
- BLOOMER, H. H., 1940, Experiments on self-fertilization in *Anodonta cygnea* (L.). *Proc. malacol. Soc. Lond.*, 24: 113-121.
- BLOOMER, H. H., 1943, A further note on experiments on self-fertilization in *Anodonta cygnea* (L.). *Proc. malacol. Soc. Lond.*, 25: 192-200.
- BLOOMER, H. H., 1946, The seasonal production of spermatozoa and other notes on the biology of *Anodonta cygnea* (L.). *Proc. malacol. Soc. Lond.*, 27: 62-68.
- BOUILLON, J., 1955, La reproduction des Anodontes. *Naturalistes Belges*, 36: 118-121.
- BRANDER, T., 1954, Über Geschlechtsdimorphismus bei europäischen Unionazeen. *Arch. Molluskenk.*, 83: 163-172.
- BRUMMER, K., 1932, Tutkimuksia Joutsenselän ja Kirmusten järven isosimpukoista, Unionidae. Ms. Helsinki [not seen; cited by Brander (1954)].
- CHAMBERLAIN, T., 1931, Annual growth of fresh-water mussels. *Bull. U.S. Bur. Fish.*, 46: 713-739, and 4 unnumbered plates containing Figs. 1-2, 3-4, 14 and 18.
- COE, W. R., 1943, Sexual differentiation in mollusks. I. Pelecypods. *Quart. Rev. Biol.*, 18: 154-164.
- COE, W. R. & TURNER, H. J., 1938, Development of the gonads and gametes in the soft-shell clam (*Mya arenaria*). *J. Morphol.*, 62: 91-111.
- COKER, R. E., SHIRA, A. F., CLARKE, H. W. & HOWARD, A. D., 1921, Natural history and propagation of fresh-water mussels. *Bull. U.S. Bur. Fish.*, 37: 77-181, pls. 5-21.
- COLE, L. C., 1954, The population consequences of life history phenomena. *Quart. Rev. Biol.*, 29: 103-137.
- CONNER, C. H., 1907, The gravid periods of unios. *Nautilus*, 21: 87-89.
- CONNER, C. H., 1909, Supplementary notes on the breeding seasons of the Unionidae. *Nautilus*, 22: 111-112.
- CROWLEY, T. E., 1957, Age determination in *Anodonta*. *J. Conchol.*, 24: 201-207.
- CVANCARA, A. M., 1963, Clines in three species of *Lampsilis* (Pelecypoda: Unionidae). *Malacologia*, 1: 215-225.
- GHISELIN, M. T., 1969, The evolution of hermaphroditism among animals. *Quart. Rev. Biol.*, 44: 189-208.
- HAZAY, J., 1881, Die Mollusken-Fauna von Budapest. *Malakozool. Blätt.*, 3: 1-69; 4: 43-224.
- HEARD, W. H., 1970, Hermaphroditism in *Margaritifera falcata* (Gould) (Pelecypoda: Margaritiferidae). *Nautilus*, 83: 113-114.
- HEARD, W. H. & GUCKERT, R. H., 1971, A re-evaluation of the Recent Unionacea (Pelecypoda) of North America. *Malacologia*, 10: 333-355.

- HOWARD, A. D., 1914, A second case of metamorphosis without parasitism in the Unionidae. *Science*, 51: 353-355.
- HOWARD, A. D., 1915, Some exceptional cases of breeding among the Unionidae. *Nautilus*, 29: 4-11.
- INABA, S., 1941, A preliminary note on the glochidia of Japanese freshwater mussels. *Annot. zool. jap.*, 21: 14-23.
- LEFEVRE, G. & CURTIS, W. C., 1910a, Reproduction and parasitism in the Unionidae. *J. exp. Zool.*, 9: 79-115, pls. 1-5.
- LEFEVRE, G. & CURTIS, W. C., 1910b, The marsupium of the Unionidae. *Biol. Bull.*, 19: 31-34.
- LEFEVRE, G. & CURTIS, W. C., 1911, Metamorphosis without parasitism in the Unionidae. *Science*, 33: 863-865.
- LILLIE, F. R., 1901, The organization of the egg of *Unio*, based on a study of its maturation, fertilization, and cleavage. *J. Morphol.*, 17: 227-292, pls. 24-27.
- McMICHAEL, D. F. & HISCOCK, I. D., 1958, A monograph of the freshwater mussels (Mollusca: Pelecypoda) of the Australian Region. *Austr. J. mar. freshw. Res.*, 9: 372-503, pls. 1-19.
- NISHIWAKI, S., 1964, Phylogenetical study on the type of the dimorphic spermatozoa in Prosobranchia. *Sci. Rep. Tokyo Kyoiku Daigaku*, sect. B, 11: 237-275.
- ÖKLAND, J., 1963, Notes on population density, age distribution, growth and habitat of *Anodonta piscinalis* Nilss. (Moll., Lamellibr.) in a eutrophic Norwegian lake. *Nytt Mag. Zool.*, 11: 19-43.
- ORTMANN, A. E., 1909, The breeding season of Unionidae in Pennsylvania. *Nautilus*, 22: 91-95, 99-103.
- ORTMANN, A. E., 1910a, A new system of the Unionidae. *Nautilus*, 23: 114-120.
- ORTMANN, A. E., 1910b, The marsupium of the Anodontinae. *Biol. Bull.*, 19: 27.
- ORTMANN, A. E., 1911, A monograph of the naiades of Pennsylvania. Parts I and II. *Mem. Carnegie Mus.*, 4: 279-347, pls. 86-89.
- ORTMANN, A. E., 1912, Notes upon the families and genera of the naiades. *Ann. Carnegie Mus.*, 8: 222-365, pls. 18-20.
- ORTMANN, A. E., 1918, The anatomy of two African nayades, *Unio caffer* and *Spatha wahlbergi*. *Nautilus*, 31: 75-78.
- ORTMANN, A. E., 1921, South American naiades; a contribution to the knowledge of the freshwater mussels of South America. *Mem. Carnegie Mus.*, 8: 451-668, pls. 34-48.
- PARODIZ, J. J. & BONETTO, A. A., 1963, Taxonomy and zoogeographic relationships of the South American naiades (Pelecypoda: Unionacea and Mutelacea). *Malacologia*, 1: 179-213.
- PECK, R. H., 1877, The minute structure of the gills of lamellibranch Mollusca. *Quart. J. microsc. Sci.*, 17: 43-66, pls. 4-7.
- PELSENEER, P., 1894, Hermaphroditism in Mollusca. *Quart. J. microsc. Sci.*, 37: 19-46, pls. 1-2.
- PELSENEER, P., 1895, L'hermaphroditisme chez les Mollusques. *Arch. Biol.*, 14: 33-62, pls. 3-5.
- PORTMANN, A., 1960, Embranchement des Mollusques (p. 1623-2164), in GRASSE, P.-P. (ed.). *Traité de Zoologie. Anatomie, Systématique, Biologie*. Tome 5: 1053-2219. Masson et Cie, Paris.
- PRASHAD, B., 1918, Studies on the anatomy of Indian Mollusca. 2. The marsupium and glochidium of some Unionidae and on the Indian species hitherto assigned to the genus *Nodularia*. *Rec. Indian Mus.*, 15: 143-148.
- ROPES, J. W. & STICKNEY, A. P., 1965, Reproductive cycle of *Mya arenaria* in New England. *Biol. Bull.*, 128: 315-327.
- SHAW, W. N., 1965, Seasonal gonadal cycle in the male soft-shell clam, *Mya arenaria*, in Maryland. *U. S. Fish. Wildlife Serv., Spec. sci. Rep., Fish.*, no. 508: 1-5.
- SIEBOLD, C. T. von, 1837, Über den Unterscheid der Schalenbildung der männlichen und weiblichen Anodonten. *Arch. Naturgesch.*, 3: 415-416.
- SIMPSON, C. T., 1914, *A descriptive catalogue of the naiades, or pearly fresh-water mussels*. Bryant Walker, Detroit. xi + 1540 p.
- SNEDECOR, G. W. & COCHRAN, W. G., 1967, *Statistical Methods*, 6th ed. Iowa State Univ. Press, Ames, Iowa. xiv + 593 p.
- STANSBERRY, D., 1967, Growth and longevity of the naiades from Fishery Bay in Lake Erie. *Amer. malacol. Union, ann. Repts.*, 1967: 10-11. [Abstract.]
- STERKI, V., 1898a, Some observations on the genital organs of the Unionidae, with reference to classification. *Nautilus*, 12: 18-21, 28-32.
- STERKI, V., 1898b, *Anodonta imbecilis*, hermaphroditic. *Nautilus*, 12: 87-88.
- STROGANOVA, N. S., 1963, Peculiarities of spermatogenesis in certain bivalve mollusks. *Vestn. Mosk. Univ., Ser. Biol. Soil Sci.*, no. 6: 25-34. [In Russian.]
- SURBER, T., 1912, Identification of the glochidia of freshwater mussels. *U.S. Bur. Fish., Doc. no. 771: 1-10*, pls. 1-3.
- TUCKER, M. E., 1927, Morphology of the glochidium and juvenile of the mussel *Anodonta imbecilis*. *Trans. Amer. microsc. Soc.*, 46: 286-293.
- TUCKER, M. E., 1928, Studies on the life cycles of two species of fresh-water mussels belonging to the genus *Anodonta*. *Biol. Bull.*, 54: 117-127.
- UTTERBACK, W. I., 1915-1916, The naiades of Missouri. *Amer. Midl. Natur.*, 4: 41-53, 97-152, 181-204, 244-273 (1915); 311-327, 339-354, 387-400, 432-464, pls. 1-27 (1916).
- VAN DER SCHALIE, H., 1938, The naiad fauna of the Huron River, in southeastern Michigan. *Misc. Publs. Mus. Zool., Univ. Mich.*, no. 40: 1-83, pls. 1-12.
- VAN DER SCHALIE, H., 1966, Hermaphroditism among North American freshwater mussels. *Malacologia*, 5: 77-78. [Abstract.]
- VAN DER SCHALIE, H., 1969, Two unusual unionid hermaphrodites. *Science*, 163: 1333-1334.
- VAN DER SCHALIE, H., 1970, Hermaphroditism among North American freshwater mussels. *Malacologia*, 10: 93-112.

ANODONTA REPRODUCTION

103

VAN DER SCHALIE, H. & LOCKE, F., 1941, Hermaphroditism in *Anodonta grandis*, a freshwater mussel. *Occ. Paps. Mus. Zool., Univ. Mich.*, no. 432: 1-7, pls. 1-3.

WEISENSEE, H., 1916, Die Geschlechtsverhältnisse und der Geschlechtsapparat bei *Anodonta*. *Z. wiss. Zool.*, 115: 262-325.

.....

.....