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HISTOLOGICAL ANALYSIS OF THE OVARIES IN TENCH (*TINA TINA* L.) FROM LAKE DRWĘCKIE

ANALIZA HISTOLOGICZNA JAJNIKÓW SAMIC LINA *TINCA TINCA* (L.) W JEZIORZE DRWĘCKIM

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Histological analysis of the ovaries of 108 tench (*Tinca tinca* L.) females confirmed asynchronic development of the oocytes in this fish. Vitellogenesis commenced in May in all years but it depended on water temperature (> 10°C). Oocyte resorption was observed throughout the year, most intensive during the reproduction period. Oogonia were also present all year round, most numerous in the post-spawning period. Ovaries of tench remained in stage III of development since September till April (inclusive).

INTRODUCTION

Studies on the biology of tench reproduction are quite numerous. Nevertheless, there are only a few papers dealing with the changes taking place in tench ovaries in an annual cycle (Kazanskij 1949, Monič 1953, Solewski 1957, Zubenko 1973, Šichšabekov 1977, Brylińska and Długosz 1978, Epler and Bieniarz 1979, Epler et al. 1981).

Studies on tench reproduction in cold- and warm-water ponds revealed strict dependence between water temperature and processes taking place in the ovaries (Epler and Bieniarz 1979, Epler et al. 1981, Horoszewicz 1981, 1983, Horoszewicz et al. 1981, Morawska 1981, 1982, 1984). On the other hand, little is known of the dependence between water temperature and oocyte maturation, ovulation and resorption in natural temperatures of our inland waters.

The aim or the studies consisted of a histological analysis of tench ovaries in an annual cycle, with special reference to the effect of temperature during the reproductive period (May-August) on changes taking place in the ovaries.

MATERIALS AND METHODS

Materials were collected from 108 tench females originating from commercial catches since April 1978 till June 1980 (Tab. 1). 6 samples were collected from 3 parts of each ovary: anterior, middle and posterior (right and left). Samples were preserved in AFA liquid, passed through alcohol and immersed in paraffin. Histological scraps 10–40 μm thick were stained with Delafield's haematoxylin with eosine.

In order to determine development phases of the reproductive cells, scale of Sakun and Buckaja (1968) was used, as modified by Epler et al. (1981). Attention was paid to morphological features of cells representing particular stages (Fig.1).

Post-spawning stages were denoted as VI/II-III, VI/III, VI/III-IV, etc. depending on maturity of the oocytes predominating in the ovary, in a similar way as before the first spawning.

In order to determine oocyte and follicular membrane resorption, Khoo's (1975) nomenclature was used, and 3 resorption stages were distinguished: α, β and γ.

Water temperatures were obtained from the Meteorological Station in Iława. Water temperature was measured at 7.00 A.M., 1 m below the surface. Water temperatures during tench reproduction (May-August) are presented in Fig. 2.

Table 1

Number of the examined tench females

Year \ Month	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	I–XII
1978	–	–	–	3	3	8	3	5	3	2	4	–	31
1979	–	4	–	–	11	13	12	2	4	5	3	4	58
1980	4	4	–	4	2	5	–	–	–	–	–	–	19
Total	4	8	–	7	16	26	15	7	7	7	7	4	108

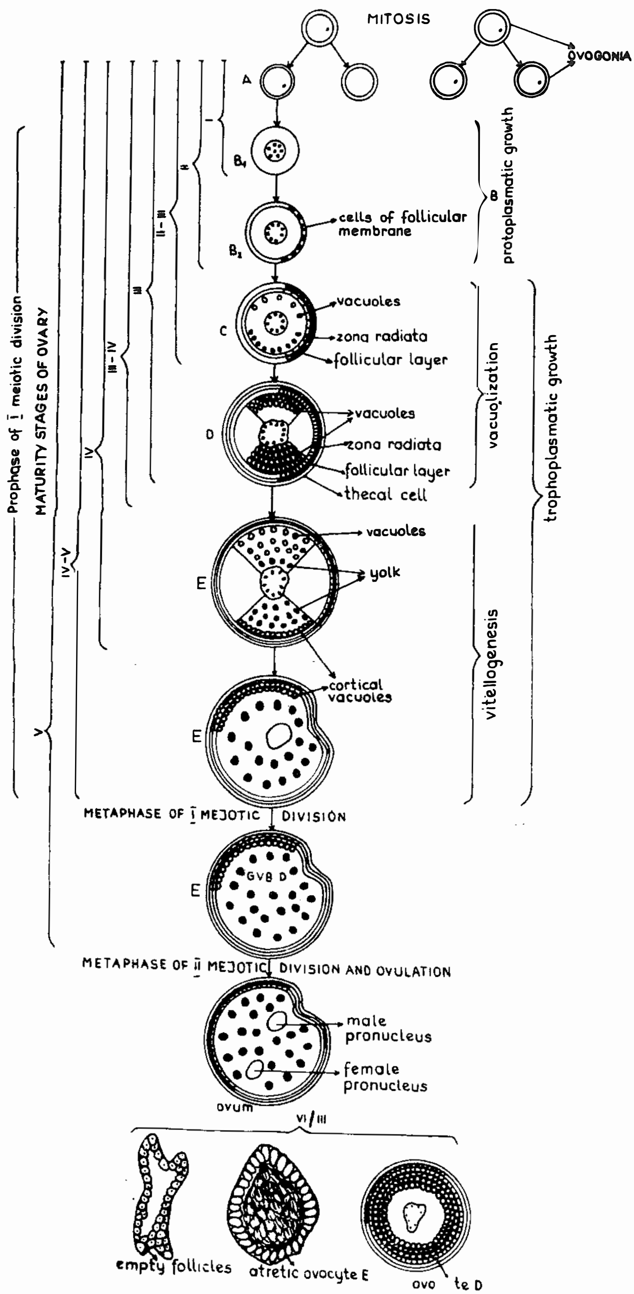


Fig. 1. Scheme of tench ovogenesis

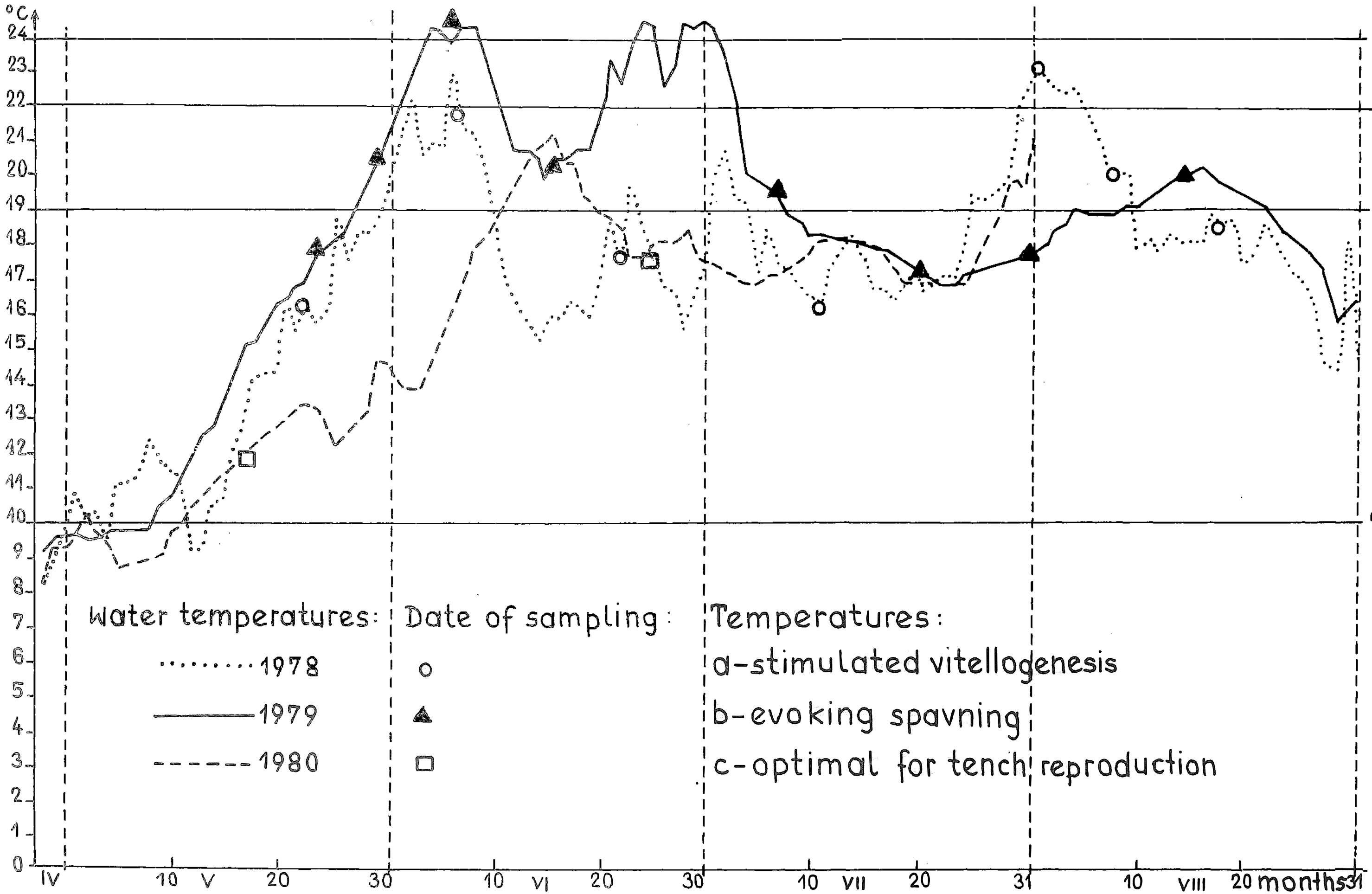


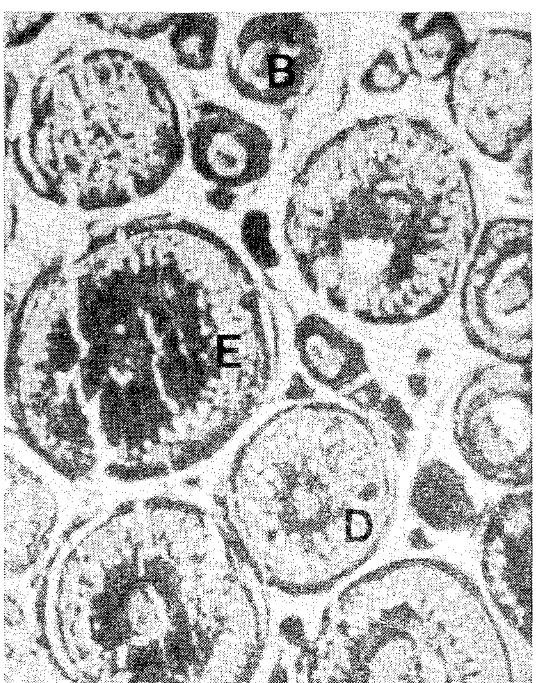
Fig. 2. Water temperatures in the period of tench reproduction

RESULTS

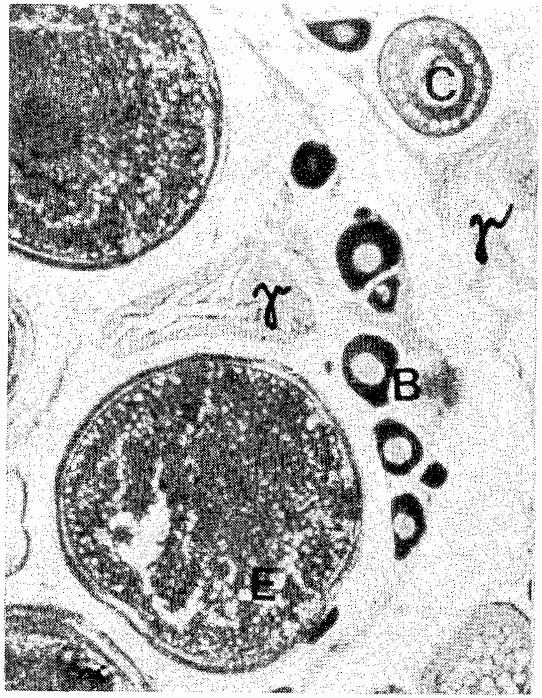
Observations on changes taking place in the ovaries were initiated in April i.e. at the time of intensive vitellogenesis which determines females fecundity in the coming reproductive season. April 1978 and 1980 was cold, with temperatures below 10°C (Fig. 2). Hence, vitellogenesis was not observed and the ovaries were only in stage III of maturity (Fig. 3). Numerous oocytes of protoplasmatic growth (B) and in various stages of vacuolization (C, D) were observed in the histological picture. Only in a few D cells vacuolization reached the nucleus.

In 1978 three females were collected on May 23 (Fig. 2). Two of them had ovaries in stage III–IV of maturity, and one in stage III (Fig. 3). In the ovaries of the first two females, the oldest E cells were almost totally filled with yolk (Phot. 1), while the youngest had still 3–4 rows of vacuoles at the circumference.

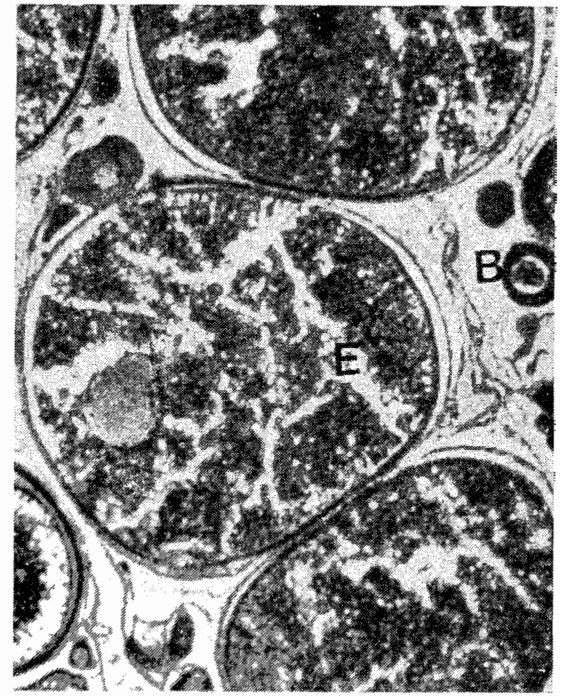
In the warmest year (1979, Fig. 2) a sample collected on May 24 contained eight females, of which seven had ovaries in stage III of maturity. One had ovaries in stage III–IV (Fig. 3). The oldest oocytes in the ovaries of seven females had four to five rows of vacuoles, and in some vacuolization reached the nucleus. Atretic oocytes D were observed in three females. Ovary in stage III–IV of maturity had the oldest oocytes in the initial phase of yolk accumulation, with three of four rows of vacuoles at the cell circumference.



Phot. 1. Ovary in stage III–IV of development – 23.05.1978 (x60)



Phot. 2. Ovary in stage VI/IV of development – 30.05.1979 (x60)



Phot. 3. Ovary in stage IV–V of development – 30.05.1979 (x60)

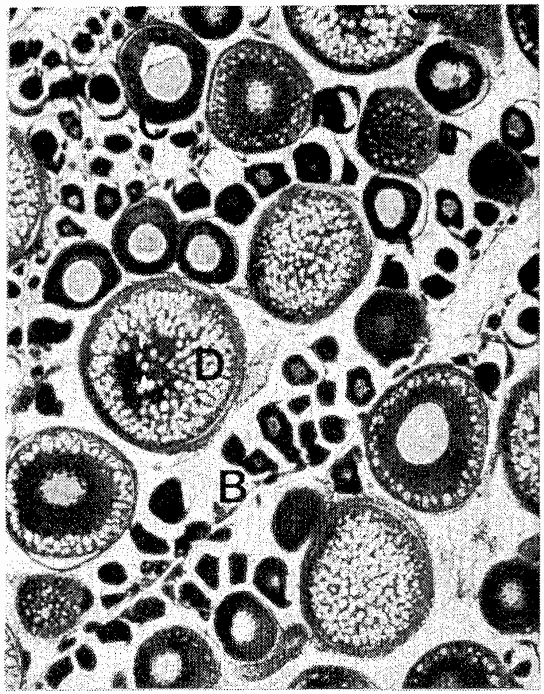
Next sample was collected on May 30 of the same year, when temperature increased to 19°C (Fig. 2). Histological sections of the ovaries of two remales had follicles after ovulation (Phot. 2), oocytes E in various stages of yolk accummulation, and resorbed oocytes. In the biggest oocytes D vacuolization reached the nucleus. Ovaries of these females were in stage VI/IV of development, suggesting that some eggs had already been spawned (Fig. 3). Ovary of the third female was in stage IV–V (Fig. 3). Many cells filled with yolk were observed, with the nucleus moving toward the circumference (Phot. 3). Cells of protoplasmatic growth were noticeable only in a few fragments of the section.

In May 1980 ovaries attained stage III or maturity (Fig. 3, Phot. 4). In the oldest D cells, vaculization reached the nucleus. Low degree of development was connected with low temperature in April and May 1980. Temperature stimulating vitellogenesis was noted as late as May 12 (Fig. 2).

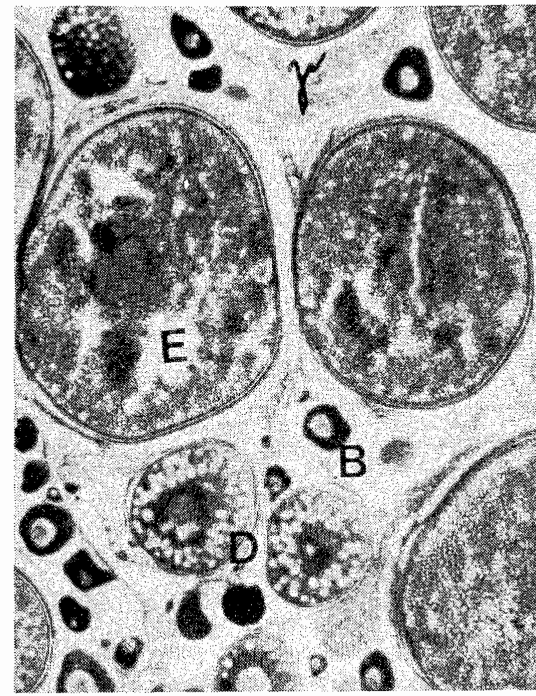
In June 1978 samples were collected twice (on 7 and 22 June, Fig. 2). The first was collected when temperature increased to the level inducing tench spawning. One female in this sample had ovaries in stage IV--V (Fig. 3). The ovary contained numerous cells filled with yolk, with nucleus translocated toward the micropyle. Two females were caught during spawning (first egg portion). Their ovaries contained numerous oocytes E filled with yolk, with formed micropyle and migrating nucleus. Empty follicles were also observed (stage γ). Ovaries of these females were in stage VI/IV/V of development (Phot. 5, Fig. 3). Ovaries of the other two females were in stage VI/III–IV of maturity, suggesting post-spawning stage. Histological picture of these ovaries revealed oocytes E filled with yolk in varying degree. Part of oocytes E as well as oocytes D (advanced vacuolization) underwent resorption. Ooocytes C, D and B and various oogonia were also noticeable.

Next sample was collected 2 weeks later (22 June, Fig. 2). At this time water temperature decreased rapidly, and then increased (Fig. 2). Two females had the ovaries in stage VI/III–IV of development (Fig. 3). Rare, very small follicular membranes (stage γ) were found, and some atretic oocytes (stage α). Oocytes E filled with yolk were also present. These were accompanied by some oocytes C and D as well as by oocytes B of protoplasmatic growth. Third female in this sample had ovaries in stage IV–V (Fig. 3).

In June 1979 two samples were collected: on 5 and 15 (Fig. 2). On 5 June eight females were caught immediately after an increase of water temperature (Fig. 3). Ovaries of these females contained numerous atretic oocytes E in stages α and γ, some atretic oocytes D, and numerous follicular membranes or atretic oocytes in stage γ (Phot. 6). Apart from the cells which were being resorbed, the ovaries contained also oocytes C and some oocytes D, and a number of oocytes B. Hence, it can be assumed that strong atresion of the oocytes D and E will not permit these females to spawn any more. Maturity of these ovaries was determined as stage VI/II–III (Fig. 3). Three other females possessed numerous follicular membranes in their ovaries as well as oocytes E filled with yolk to a varying degree. These ovaries reached stage VI/IV of maturity (Fig. 3), i.e. the females had just spawned. Ovaries of two other females were in stage IV–V of maturity



Phot. 4. Ovary in stage III of development – 17.05.1980 (x60)



Phot. 5. Ovary in stage VI/IV–V of development – 5.06.1979 (x60)

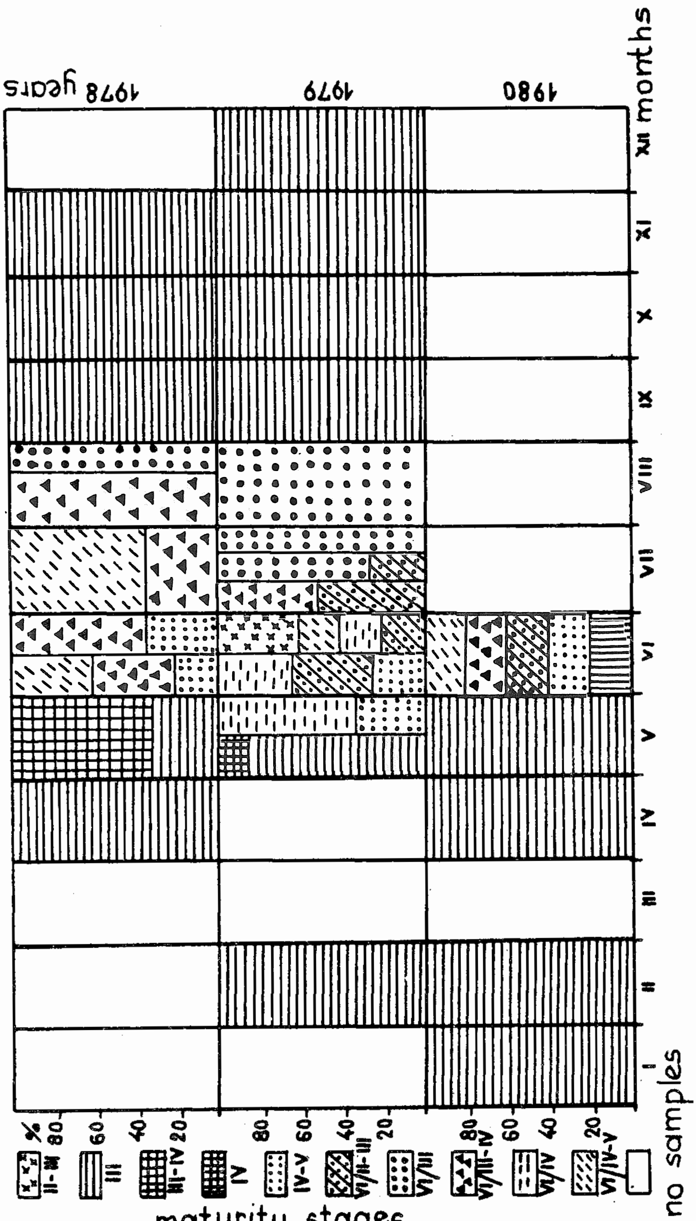
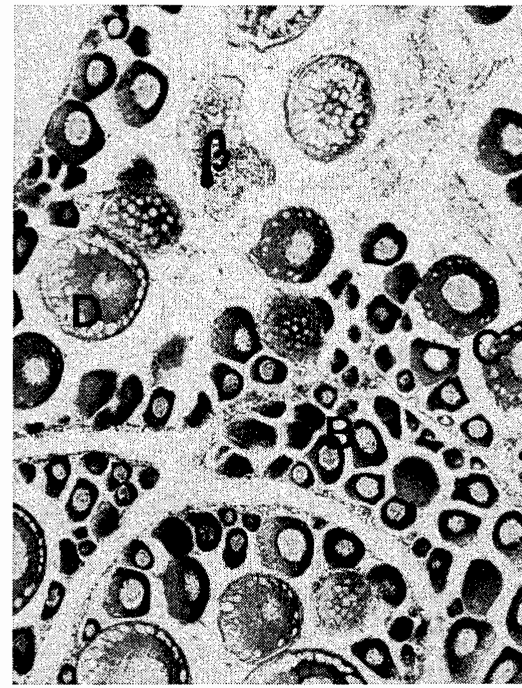


Fig. 3. Maturity stages of female gonads of tench from Drwęckie Lake

Phot. 6. Ovary in stage VI/II–III of development – 5.06.1979 (x60)



Phot. 7. Ovary in stage II–III of development – 15.06.1979 (x60)

(Fig. 3) and contained oocytes E, in which the nucleus had moved toward the circumference and micropyle was well visible. These females were ready to spawn their first egg portion. Next sample was collected 10 days later (June 15), at water temperature above 19.5°C (Fig. 2). Follicles of different sizes were visible in the histological sections of three ovaries, suggesting that these females reached stage VI/IV, VI/IV/V and VI/II–III of maturity (Fig. 3). In the latter ovary, atretic oocytes E and D were found. Ovaries of two young females (4+ and 3+) contained oocytes in the initial stage of vacuolization (C and D) as well as oocytes B of protoplasmatic growth and numerous oogonia A. Atretic oocytes D were found in some places. Ovaries of these females were in stage II–III of development (Phot. 7, Fig. 3). Probably these fishes will not participate in spawning during this reproductive season.

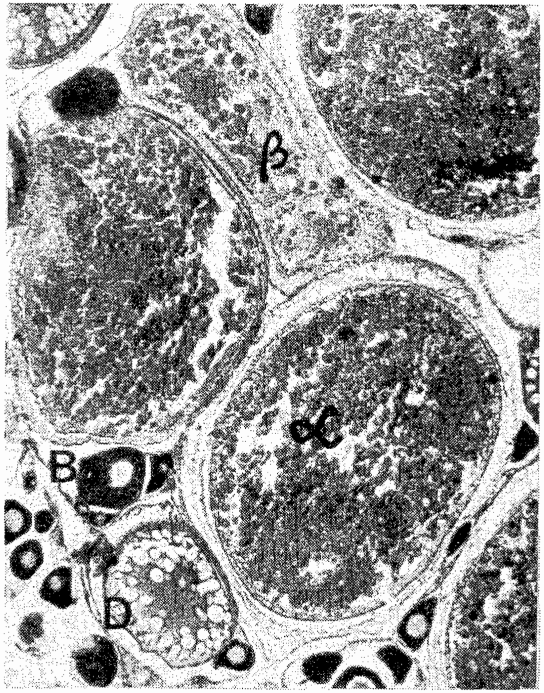
On June 25, 1980, after a 5-day decrease of temperature from 21°C to 17°C (Fig. 2), five females were caught. Two of them possessed ovaries in stage IV and IV–V of maturity (Fig. 3), containing many oocytes E filled with yolk to a varying degree, and oocytes C and D. Oocytes B of protoplasmatic growth were observed close to lamella side as also numerous oogonia A. No empty follicles were found. Kazanskij (1949) stated that resorption of follicles lasted for 15–30 days. Hence, it can be assumed that at the observed temperature range (Fig. 2) these females had not spawned yet. Ovaries of three females contained small follicles; their ovaries were in stage VI/IV–V, VI/III–IV and VI/II–III of maturity (Fig. 3). Percentage of oocytes C, D and E differed. Ovaries of two females contained also many oocytes B of protoplasmatic growth and oogonia A. Numerous atretic oocytes D were also observed. State of follicular membranes did not allow for stating whether these were some residues after ovulation or stage γ of atretic oocytes.

July samples were collected only in two years (1978, 1979). Ovaries of all females contained follicles suggesting that they had spawned at least one egg portion. Preparation of the ovaries for next portion differed in particular females.

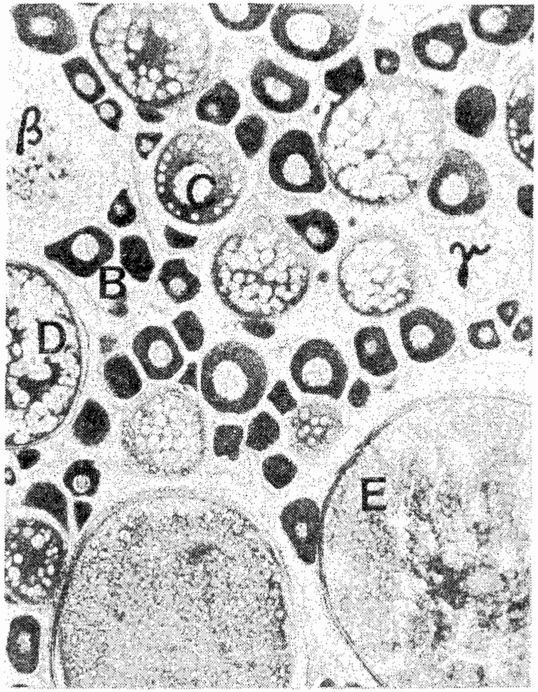
On July 11, 1978, three females were caught (Fig. 2), of which two had ovaries in stage VI/IV–V (Fig. 3), with numerous oocytes E totally filled with yolk, containing micropyle, a few oocytes C and D, and oocytes B of protoplasmatic growth. Some atretic oocytes and follicular membranes were also noticed. One female was ready to lay another portion of eggs; its ovary was in stage VI/III–IV (Fig. 3).

In July 1979 three samples were collected, the first on July 7 (Fig. 2). Before sampling the temperature decreased rapidly after a period of very warm weather (24.5°C) at the turn of June and July. Two females from this sample had ovaries in stage VI/III–IV, containing many oocytes E almost totally filled with yolk, and atretic oocytes E in stage α. It can be assumed that the females were ready to spawn but decrease of temperature inhibited reproduction and resorption of oocytes E commenced. Ovaries of the other two females were in stage VI/II–III and these fishes had probably completed reproduction in this season.

Next samples (20 and 30 July) were collected in cool weather, the temperature being lower than the threshold level (Fig. 2). Ovaries of eight females contained atretic oocytes



Phot. 8. Ovary in stage VI/III of development – 30.07.1979 (x60)



Phot. 9. Ovary in stage VI/III–IV of development – 1.08.1978 (x60)

E at different stages of resportion (Phot. 8) as well as fully vacuolized oocytes D. The latter were also being resorbed. Small follicular membranes (stage γ) were also noticed. Some parts of the ovaries contained numerous oogonia concentrated in nests or strings, among which oocytes B of protoplasmatic growth were also noticed. Ovaries of seven females were in stage VI/III, and one female had the ovaries in stage VI/II–III (Fig. 3).

In August 1978 and 1979 thermal conditions varied considerably. Females caught in the first days of August 1978 (on August 1 and 8, Fig. 2) had the ovaries in stage VI/III–IV (Fig. 3). They contained empty follicular membranes and unlaid oocytes E in various stages of resorption (β and γ , Phot. 9), but also oocytes D and C, and oocytes B of protoplasmatic growth as well as oogonia A concentrated in nest-like formations. These females had probably laid consecutive egg portion, while next portion was just developing.

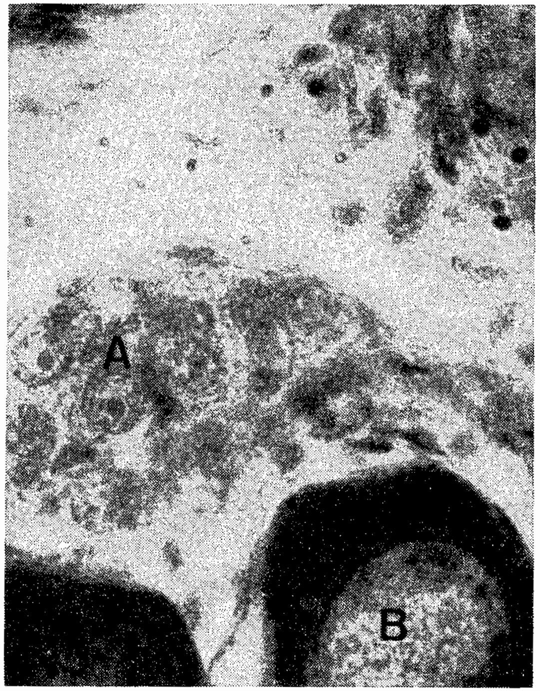
Tench females caught on 17 August (Fig. 2) had ovaries in stage VI/III of maturity (Fig. 3). Oocytes D with maximally three or four rows of vacuoles were the oldest cells observed in the histological picture. Follucilar membranes (stage γ) of different sizes were observed; some were very small. Atretic oocytes E were also present as well as some oocytes D undergoing resorption. Oogonia A were numerous (Phot. 10).

Ovaries collected on 14 August 1979 were in stages VI/III. One female possessed oocytes E undergoing resorption (α and β). Numerous oocytes C were observed in the histological picture and less numerous oocytes D with only two rows of vacuoles. Oocytes B of protoplasmatic growth were very numerous as well as oogonia A.

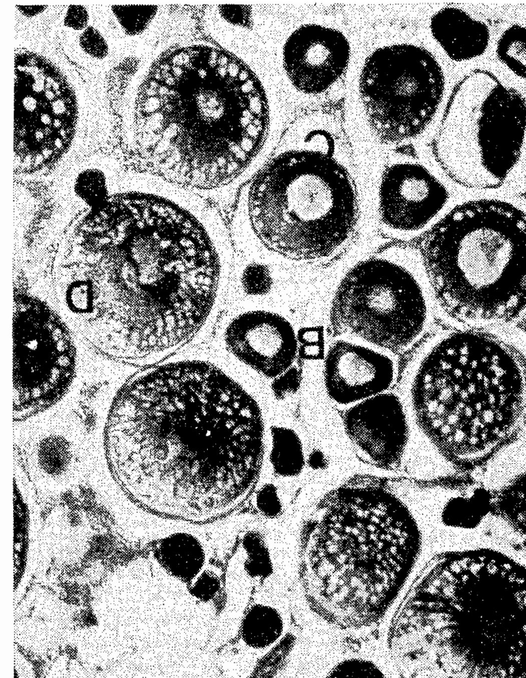
Picture of the ovaries collected in the second half of August 1978 and in August 1979 suggested that spawning had been completed and the gonads commenced to re-establish oocyte pool for the next season. This was confirmed by the presence of oogonia.

In September 1978 and 1979 seven females were collected (Tab. 1). Their ovaries were in stage III of maturity (Fig. 3, Phot. 11). Numerous follicular membranes almost completely closed (stage γ) were still observed; only a few contained yolk residues (stage γ). Vacuolization reached the nucleus in some oocytes D; part of them were being resorbed. Number and size of oocytes B of protoplasmatic growth increased considerably, as if B_1 oocytes developed rapidly and passed into stage B_2. A few oogonia A were also noticed.

In October 1978 and 1979 seven females were collected (Tab. 1) with the ovaries in stage III of development (Fig. 3). Very numerous oogonia A were observed in the histological picture as well as numerous oocytes D with maximally three rows of vacuoles. These were accompanied by oocytes B of protoplasmatic growth and oocytes C. Some atretic oocytes E and small follicular membranes were also noticed (stage γ). Oogonia A in various growth phases and small oocytes B_1 of protoplasmatic growth were more numerous than in September. Also number of vacuolized oocytes D increased. Ovary of one female was, however, noticeably different. It contained numerous oocytes B of protoplasmatic growth, which formed compact groups. Oocytes C and D undergoing vacuolization were rare (these bore signs of degeneration). Oogonia were numerous, especially in the transition stage between A and B_1. Large rosy areas were observed over



Phot. 10. Nest of oogonia in an ovary section, stage VI/III of development – 17.08.1978 (x60)



Phot. 11. Ovary in stage III of development – 29.09.1978 (x100)

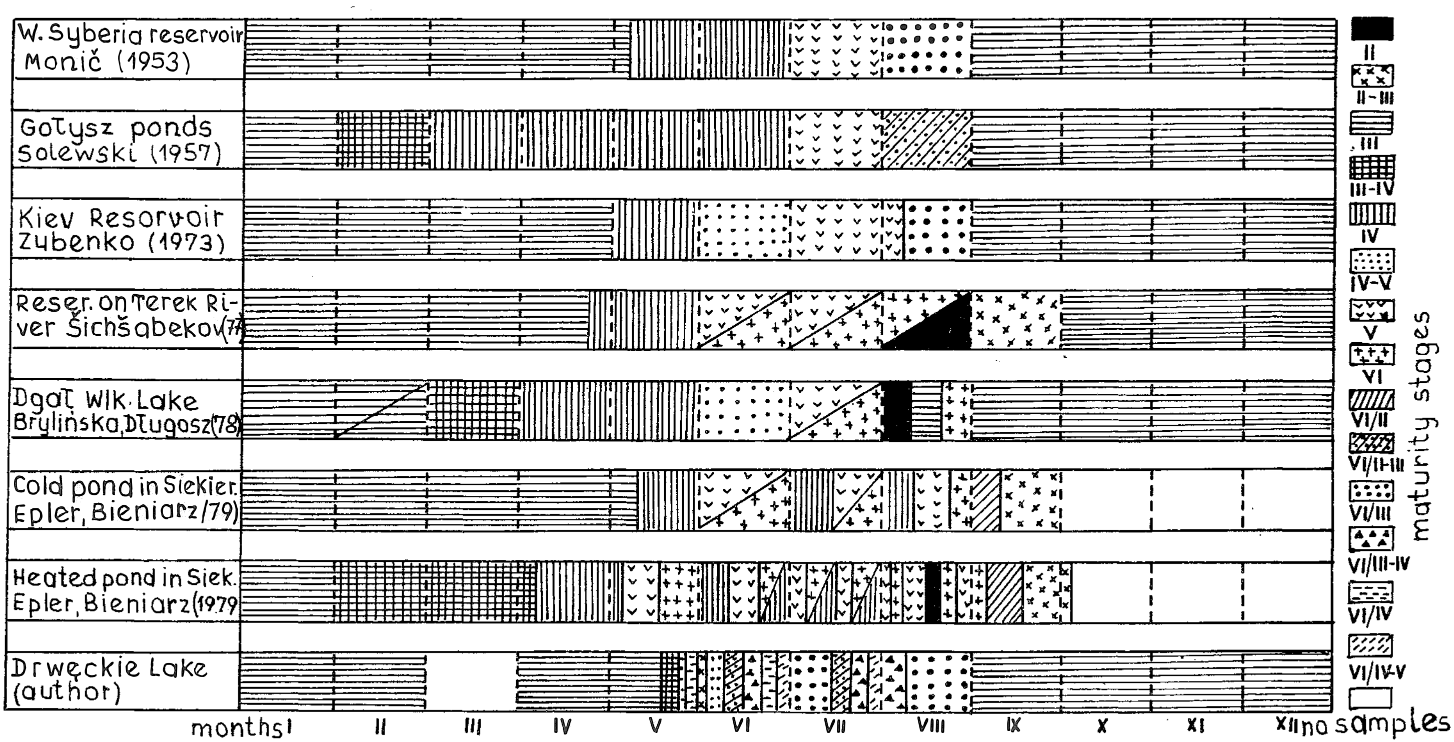


Fig. 4. Maturity stages of female gonads of tench from Drwęckie Lake in comparison to the data from the literature

Phot. 12. Mass oocyte resorption – 19.11.1979 (x60)

the whole section, especially at the edges (Phot. 12). They represented places of mass resorption of the unspawned eggs.

Seven females were caught in November 1978 and 1979 (Tab. 1). Ovaries of these fishes were in stage III of maturity. Very numerous oogonia A were observed in the histological picture and a number of oocytes B of protoplasmatic growth. Their number was higher than in October. Vacuolization was not advanced in three females – oocytes D had maximally three rows of vacuoles. All ovaries contained atretic oocytes in various stages (α, β, γ). Gonads of one female were characterized by mass resorption of oocytes, and the ovary looked as in October samples.

Four females were collected in December 1979 (Tab. 1). Their ovaries were in stage III of development (Fig. 3), but more advanced than those examined in November. Most D cells possessed three full rows of vacuoles and the fourth row was being formed. A few oogonia were observed and numerous oocytes B of protoplasmatic growth, less oocytes C, more D as well as atretic oocytes (stage β and γ).

Four females were collected in January 1980 (Tab. 1) with the ovaries in stage III of maturity. Oocytes D were very numerous in all ovaries; many of them had four rows of vacuoles, and in some vacuolization reached the nculeus. Oogonia were also noticed in all sections but their number varied from a few to many. Some atretic oocytes in stage α and β were observed, and more numerous in stage γ.

Eight females were collected in February 1979 and 1980 (Tab. 1). Their ovaries contained numerous oogonia A and oocytes B of protoplasmatic growth. Vacuolization of oocytes D differed in particular females. These oocytes contained three to five rows of vacuoles, and sometimes vacuolization reached the nucleus. Atretic oocytes in stage α, β and γ were found in the ovaries of two females. Ovaries of all fishes were in stage III of development.

DISCUSSION

Studies on tench population in Lake Drwęckie revealed strict relationship between water temperature in particular years and histological picture of the ovaries. Level and seasonal variations of water temperature affected rate of development of the reproductive cells and ovulation period.

As soon as temperatures reached threshold value for the vitellogenesis (> 10°C), nutritive substances commenced to accumulate in the oocytes, and consecutive egg portions were formed. Vitellogenesis commenced in May in all years but this was not true of all females. Ovaries of some fishes were still in stage III of maturity (Figs 3 and 4).

Also in lakes of West Siberia (Monič 1953) and in a cold-water pond in Siekierki (Epler and Bieniarz 1979) vitellogenesis commenced late in the season and not in all females at the same time (Fig. 4). On the other hand, in Kiev Reservoir (Zubenko 1973) vitellogenesis commenced in May in all tench females. In a fish pond in Gołdysz, yolk accumulated in the oocytes as early as February (Solewski 1957), and a little later (March) in Lake Dgał Wielki (Brylińska and Długosz 1978) and in a heated pond in Siekierki (Epler and Bieniarz 1979).

In Lake Drwęckie vitellogenesis lasted from about 21 days in case of tench females with more developed gonads and at rapidly growing temperature in 1979 to about 35 days in 1980. Similar results were obtained by Epler and Bieniarz (1979) for tench reared in a cold-water pond in Siekierki, and by Šichšabekov (1977) for tench from the rivers Terek and Sułaka. Vitellogenesis lasted for a longer period in those water bodies in which it commenced earlier: in Gołysz ponds – 5 months, in Lake Dgał Wielki – 3 months, in warm-water ponds in Siekierki – 3 months (Fig. 4). The shortest vitellogenesis was observed in a cold-water pond in Siekierki in 1975 – only 18 days at constantly growing temperature (Epler et al. 1981).

In my studies I have observed first follicular membranes in the ovaries of two females on 30 May 1979. In most fishes they appeared in June. This means that the females had laid first egg portion. Period of spawning the first and the last egg portion depended on water temperature (Fig. 2). It can be assumed that in 1978 tench females laid 3 and in 1979 – 2 egg portions.

In water bodies cited in the literature, tench females also commenced spawning in June, ponds in Gołysz and West Siberian waters being the only exception (Fig. 4). In Lake Drwęckie, tench spawning in 1978 ended probably in August, when water temperature decreased rapidly. In 1979 spawning must have ended even earlier, at the begining of July, and the eggs were subsequently resorbed.

Also other authors observed that tench spawned until July (Monič 1953, Solewski 1957, Brylińska and Długosz 1978). Moroz (1968) observed that spawning ended in mid-July notwithstanding high temperatures of 22–26°C. Zaharova (1955), Šichšabekov (1977) stated that tench ceased to spawn in the first days of August, and Papadopol and Wienberger (1971) – in mid-August. Epler et al. (1981) and Horoszkiewicz et al. (1981) stated that in cold- and warm-water ponds in Siekierki tench ceased to spawn in the second half of August (in 1974 and 1975) independently of water temperature.

Histological analysis of tench ovaries from Lake Drwęckie revealed asynchronic, continuous growth of oocytes during the spawning season. These findings agree with the observations by other authors: Kazanskij (1949), Solewski (1957), Koŝelev (1962, 1971), Vibrickas (1968), Papadopol (1970), Papadopol and Weinberger (1971), Šichšabekov (1977), Brylińska and Długosz (1978), Epler et al. (1981), Morawska (1981, 1982). Morawska (1982) states that asynchronic growth of the oocytes may be determined by water temperatures. Monič (1953) observed a special type of asynchronic vitellogenesis in tench from West Siberian waters, of a transitory character between broken and continuous asynchronic growth.

Resorption of the oocytes in different phases of development was observed throughout the year. The same was observed by Kazanskij (1949). Egg resorption in fishes of portion spawning does not inhibit the development of other oocytes and may occur at the same time as the development and ovulation of consecutive egg portions (Kazanskij 1949, Šichšabekov 1974, 1985, Oven 1976, Morawska 1982).

Parallel to resorption processes, oogonia were observed in the histological sections of the ovaries throughout the whole period of studies. They were most numerous in the post-spawning period. Also Kazanskij (1949) in tench and Crossland (1977) in *Chrysophrys auratus* observed increased numbers of oogonia in the post-spawning period.

Period of restoration (August) and rest (since September till the end of April) lasted in tench ovaries from Lake Drwęckie for 9 months. In this period all ovaries were in stage VI/II–III or VI/III, and then in stage III of maturity. Similar results were obtained by Monič (1953) and Zubenko (1973) (Fig. 4).

CONCLUSIONS

1. Analysis of the development of tench ovaries in an annual cycle confirmed asynchronic oocyte development in this fish.

2. Vitellogenesis commenced in May. Its begining was determined by water temperature (> 10°C). The shortest vitellogenesis lasted for 21–32 days. As vitellogenesis commenced, individual differences were observed between the females with respect to ovary development.

3. In Lake Drwęckie the first egg portions were spawned by the end of May or the begining of June. Begining of spawning depended on thermal conditions and occurence of threshold temperatures for tench spawning (> 19°C).

4. Spawning period of tench population in Lake Drwęckie ended at the begining of August in 1978, and at the begining of July in 1979.

5. Rest period of tench ovaries lasted since September till April. Ovaries were in stage III of maturity.

6. Oocyte resorption was observed throughout the year. It was most intensive during fish reproduction, especially in 1979 when water temperature showed considerable variations.

7. Oogonia were noticed in histological pictures of the ovaries throughout the year. They were most numerous in the post-spawning period suggesting re-establishment of repeoductive cell pool in course of intensive oogonial divisions.

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ANALIZA HISTOLOGICZNA JAJNIKÓW SAMIC LINA (*TINCA TINCA* L.) W JEZIORZE DRWĘCKIM

STRESZCZENIE

Materiał do badań pozyskano od 108 samic lina w okresie od kwietnia 1978 roku do czerwca 1980 roku (tab. 1) z Jeziora Drwęckiego. Skrawki jajnika utrwalono w płynie AFA, po przeprowadzeniu przez alkohole zatopione w parafinie. Skrawki histopatologiczne grubości 10–40 μm barwiono hematoksyliną Delafielda z eozyną i przeglądano w mikroskopie świetlanym typu Ergaval. W celu określenia faz rozwoju komórek rozrodczych posługiwano się skalą dojrzałości opracowaną przez Sakun, Buckaja (1968), zmodyfikowaną przez Epler i inni (1981) (rys. 1). W przeprowadzonych badaniach zauważono ścisły związek między przebiegiem temperatur wody w różnych latach badań, a obrazem histologicznym badanych jajników lina (rys. 2,3).

Rozpoczęcie procesu witellogenezy w 1978 i 1979 roku nastąpiło w maju, natomiast w 1980 roku dopiero w czerwcu (rys. 3), co warunkowane było termiką wody (> 10°C). Wraz z rozpoczęciem procesu witellogenezy obserwowano indywidualne różnice w dojrzałości jajników u poszczególnych samic.

Zakończenie sezonu rozrodczego nastąpiło na początku sierpnia w 1978 roku i na początku lipca w 1979 roku. Okres spoczynku jajników trwał od września do końca kwietnia i jajniki w tym czasie znajdowały się w III stadium dojrzałości. Podczas całego cyklu rocznego obserwowano resorbowane owocyty różnych stadiów rozwoju (najwięcej w okresie potarłowym) oraz występowanie owogonii (najliczniejsze w okresie potarłowym). Przeprowadzone badania potwierdziły asynchroniczny rozwój owocytów w jajnikach samic lina. Otrzymane wyniki analizy histologicznej cyklu rocznego jajników samic lina z Jeziora Drwęckiego porównano z cyklem rocznym samic lina.

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