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FORMATION OF FECUNDITY OF TENCH, *TINCA TINCA* (L.) FEMALES IN LAKE DRWĘCKIE

FORMOWANIE SIĘ PŁODNOŚCI SAMIC LINA – *TINCA TINCA* (L.) W JEZIORZE DRWĘCKIM

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Analyses were made of 108 ovaries of tench females from Lake Drwęckie. Measurement of oocyte diameter revealed that in order to calculate absolute fecundity of tench females it was necessary to include oocytes with diameter of from 0.2 mm onwards. In these oocytes the trophoplasmatic growth had been initiated. Quantitative analyses showed that the pool of oocytes of trophoplasmatic growth in the annual cycle reached 4.9-31.9% of all oocytes. This pool was 2.1-19.4-fold lower than the pool of oocytes of protoplasmatic growth. However, number of trophoplasmatic cells was sufficient to form the oocytes which could be spawned in the given season. Absolute fecundity calculated before fish spawning was much higher than the physiological fecundity. This was due to the resorption of maturing oocytes. The process was especially intensive when water temperature decreased rapidly.

INTRODUCTION

Absolute fecundity of tench is usually expressed as the number of oocytes of trophoplasmatic growth present in the ovaries before the first egg batch has been spawned (Kaj et al. 1964, Skóra 1964, Moroz 1968, Brylińska et al. 1979, Pimpicka 1981). So far, estimates of absolute fecundity have been based on counts of cells bigger than 0.2 mm (Vibrickas 1968), 0.3 mm (Moroz 1968, Morawska 1981, 1982, Pimpicka 1986), 0.31 mm (Brylińska et al. 1979) or even 0.4 mm (Monič 1953, Čeban

Table 1

Sampling times and studied characteristics of tench females in Lake Drwęckie during 1978–1980

Date	Number (n)	Ovary			Body length (L_c)	Age (A)	Gonadosomatic index (GSI)
		Maturity stage	Percent oocytes in trophoplasmatic growth (% CDE)	Percent oocytes in protoplasmatic growth (% B)			
	a) 1978						
24 April	3	III	10.1–17.6	82.4–89.9	27.0–30.0	5–6	2.7–3.7
23 May	2	III–IV	17.5–22.3	77.7–82.5	32.4–33.1	6	5.1–6.6
23 May	1	III	19.8	80.2	27.5	5	6.1
22 June	2	IV–V	23.9–31.9	68.1–76.1	26.2–27.3	5	13.4–16.1
	4	VI/III–IV	12.9–25.4	74.6–87.1	27.3–31.8	5–6	6.3–14.9
	2	VI/IV–V	17.1–23.6	76.4–82.9	24.8–25.8	3–4	5.5–14.9
11 July	1	VI/III–IV	19.9	80.1	24.5	4	11.5
	2	VI/IV–V	13.1–16.6	83.4–86.9	32.2–35.4	6–7	12.8–27.1
1,8, 17 Aug.	3	VI/III	9.6–17.4	82.6–90.4	21.8–35.1	4–7	1.4–4.4
	2	VI/III–IV	15.9–17.5	72.5–74.1	27.2–32.4	5–7	5.1
29 Sep.	3	III	17.1–19.1	81.9–82.9	31.2–33.8	5–6	3.1–4.0
24 Oct.	2	III	21.2–21.4	78.6–78.8	26.7–33.2	5–6	1.8–3.0
22 Nov.	4	III	9.9–16.9	83.1–90.1	21.6–24.9	3–4	1.5–4.2
	b) 1979						
27 Feb.	4	III	14.7–18.2	81.8–85.3	27.9–32.6	4–6	2.5–4.3

21, 24, 30 May	7	III	7.0–22.8	77.2–93.0	20.3–34.0	3–7	0.7–6.4
	1	III–IV	19.5	80.5	22.4	4	6.4
	1	IV–V	23.4	76.6	22.8	5	12.6
	2	VI/IV	23.0–23.4	77.0–76.6	27.3–29.8	5–6	11.8–12.3
5, 15 June	2	II–III	8.3–9.6	90.4–91.7	21.9–23.6	3–4	1.1–1.8
	2	IV–V	24.4–27.3	72.7–75.6	24.2–26.6	4–5	10.0–18.2
	3	VI/II–III	4.9–9.4	90.6–95.1	20.8–26.5	4–5	2.6–4.7
	1	VI/III	18.9	81.1	22.8	4	5.8
	4	VI–IV	17.4–25.5	74.5–82.6	21.3–34.7	4–7	4.9–7.6
	1	VI/IV–V	28.6	71.4	33.9	7	13.9
7,20,30 July	3	VI/II–III	9.2–11.4	88.6–90.8	29.2–37.0	5	2.9–6.0
	7	VI/III	6.8–19.3	80.7–93.2	26.0–36.1	4–8	2.9–11.8
	2	VI/III–IV	9.1–18.7	90.9–81.3	29.9–31.6	6	9.0–16.4
14 Aug.	2	VI/III	10.1–12.9	87.1–89.9	22.8–27.1	3–5	2.1–4.0
19 Sep.	4	III	11.7–26.0	74.0–88.3	28.2–36.0	5–8	3.2–4.8
9 Oct.	5	III	7.7–24.2	75.8–92.3	26.1–37.0	4–8	1.4–6.1
19 Nov.	3	III	13.4–19.2	80.8–86.6	31.5–38.7	6–8	3.0–4.4
21 Dec.	4	III	15.0–17.9	82.1–85.0	26.3–29.7	4–6	0.7–4.4
	c) 1980						
25 Jan.	4	III	15.4–23.4	76.6–84.6	30.8–33.9	6	3.1–4.4
19 Feb.	4	III	9.4–21.4	78.6–90.6	22.0–32.0	5–6	2.9–9.4
19 Apriö	4	III	11.0–19.0	81.0–89.0	26.8–38.5	4–6	2.8–5.1
17 May	2	III	6.4–18.5	81.5–93.6	23.8–33.6	4–6	2.5–5.8
25 June	1	IV	23.7	76.3	21.2	3	14.5
	1	IV–V	30.2	69.8	25.1	4	13.6
	1	VI/II–III	9.0	99.0	19.4	3	1.9
	1	VI/III–IV	26.6	73.4	24.5	4	3.4
	1	VI/IV–V	27.4	72.6	31.2	5	21.1

1975, Zubenko 1975). Inclusion or omission of the oocytes of different size may lead either to overestimation or to underestimation of the absolute fecundity.

Oocytes which had attained the phase of trophoplasmatic growth before the first egg-batch was spawned, were likely to attain full maturity and be spawned in the given season. The oocytes of protoplasmatic growth left in the ovaries constituted a reserve pool for the next years.

The objectives of the work were:

- to determine diameter of oocytes of trophoplasmatic growth which should be taken into account in the estimates of fecundity from microscopic analyses, the latter allowing for precise determination of cell developmental stage;
- to follow up seasonal changes taking place in the ratio between oocytes of trophoplasmatic growth and those of protoplasmatic growth;
- to check whether the pool of trophoplasmatic oocytes was supplemented with oocytes of protoplasmatic growth when consecutive egg batches were spawned;
- to study the effect of egg resorption on the estimates of absolute (potential) fecundity made before spawning.

MATERIAL AND METHODS

Materials were collected from commercial catches performed from April 1978 till June 1980 (Tab. 1a, b, c).

The following measurements were made: total female weight (W), weight after gutting (W_o), and weight of ovaries (O_w). Gonadosomatic index (GSI) was calculated as a ratio of ovary weight to degutted fish weight. Age (A) of tench females was determined from scales.

Ovaries for histological examinations were preserved in AFA liquid, passed through alcohol, and immersed in paraffin. Histological scraps 10-40μm thick were stained with Delafield's haematoxylin with eosine.

In order to determine the phases of reproductive cell development, advantage was taken of the scale by Sakun and Buckaja (1968), as modified by Epler et al. (1981) and Pimpicka (1986, 1989). In order to distinguish the resorbed oocytes (atretic) and follicles, the nomenclature of Khoo (1975) was used. Histological sections were randomly selected from all sections made of a given gonad. The procedures used were determined following a pilot study (Pimpicka 1986). Counts were made in 30 squares of a net 0.1 mm x 0.1 mm, overposed on the gonad section (Fot. 1) using 100 x magnification. The following were counted:

1. Oocytes of protoplasmatic (B) and trophoplasmatic (C, D, E) growth. Attention was given only to the oocytes which possessed a visible nucleus. Ratio between the number of oocytes of protoplasmatic and trophoplasmatic growth was used, to determine their percentage in the whole ovary.

Table 2

Diameter of protoplasmatic (B) and trophoplasmatic (C, D, E) growth oocytes of tench gonads in annual cycle (in μm);

a – range of oocytes diameter measured on histological sections
b – range of oocytes diameter expressed as a standard deviation (X ± SD)
c – range of estimated oocytes diameter preserved in formalin for description see Methods

Month	B				C			
	a	X ± SD	b	c	a	X ± SD	b	c
Jan.	0.020–0.140	0.068±0.030	0.038–0.098	0.054–0.140	0.130–0.225	0.178±0.030	0.148–0.208	0.211–0.297
Feb.	0.020–0.150	0.071±0.034	0.037–0.105	0.053–0.150	0.140–0.220	0.180±0.018	0.162–0.198	0.231–0.283
April	0.012–0.190	0.074±0.044	0.003–0.118	0.043–0.169	0.114–0.275	0.191±0.034	0.157–0.225	0.224–0.321
May	0.012–0.165	0.066±0.033	0.033–0.099	0.047–0.141	0.100–0.285	0.176±0.034	0.142–0.210	0.200–0.300
June	0.012–0.136	0.064±0.028	0.036–0.092	0.051–0.131	0.095–0.250	0.170±0.028	0.142–0.198	0.200–0.280
July	0.014–0.132	0.062±0.029	0.033–0.091	0.047–0.130	0.100–0.230	0.178±0.022	0.156–0.200	0.223–0.286
Aug.	0.016–0.128	0.065±0.031	0.034–0.096	0.049–0.137	0.125–0.230	0.175±0.022	0.153–0.197	0.219–0.281
Sep.	0.020–0.130	0.068±0.030	0.038–0.098	0.054–0.140	0.130–0.225	0.179±0.020	0.159–0.199	0.227–0.284
Oct.	0.020–0.136	0.067±0.033	0.034–0.100	0.049–0.143	0.130–0.215	0.173±0.019	0.154–0.192	0.220–0.274
Nov.	0.020–0.136	0.072±0.033	0.039–0.105	0.056–0.150	0.130–0.200	0.173±0.018	0.155–0.191	0.221–0.273
Dec.	0.020–0.134	0.069±0.033	0.036–0.102	0.051–0.146	0.150–0.230	0.183±0.020	0.163–0.203	0.233–0.290
Month	D				E			
	a	x ± SD	b	c	a	x ± SD	b	c
Jan.	0.240–0.430	0.332±0.057	0.275–0.389	0.293–0.556				
Feb.	0.215–0.410	0.311±0.050	0.261–0.361	0.373–0.516				
April	0.180–0.435	0.335±0.055	0.280–0.390	0.400–0.557				
May	0.156–0.440	0.311±0.061	0.250–0.372	0.360–0.530	0.350–0.830	0.562±0.134	0.428–0.696	0.600–0.990
June	0.165–0.470	0.290±0.057	0.233–0.347	0.330–0.490	0.220–0.900	0.555±0.138	0.417–0.693	0.596–0.990
July	0.170–0.410	0.270±0.049	0.220–0.320	0.314–0.457	0.320–0.860	0.604±0.120	0.484–0.724	0.691–1.034
Aug.	0.200–0.350	0.248±0.036	0.212–0.284	0.303–0.406	0.360–0.760	0.543±0.118	0.425–0.661	0.607–0.944
Sep.	0.150–0.370	0.286±0.048	0.238–0.334	0.340–0.477				
Oct.	0.200–0.415	0.289±0.055	0.234–0.344	0.344–0.491				
Nov.	0.200–0.410	0.292±0.058	0.234–0.350	0.334–0.500				
Dec.	0.210–0.440	0.317±0.062	0.255–0.379	0.364–0.541				

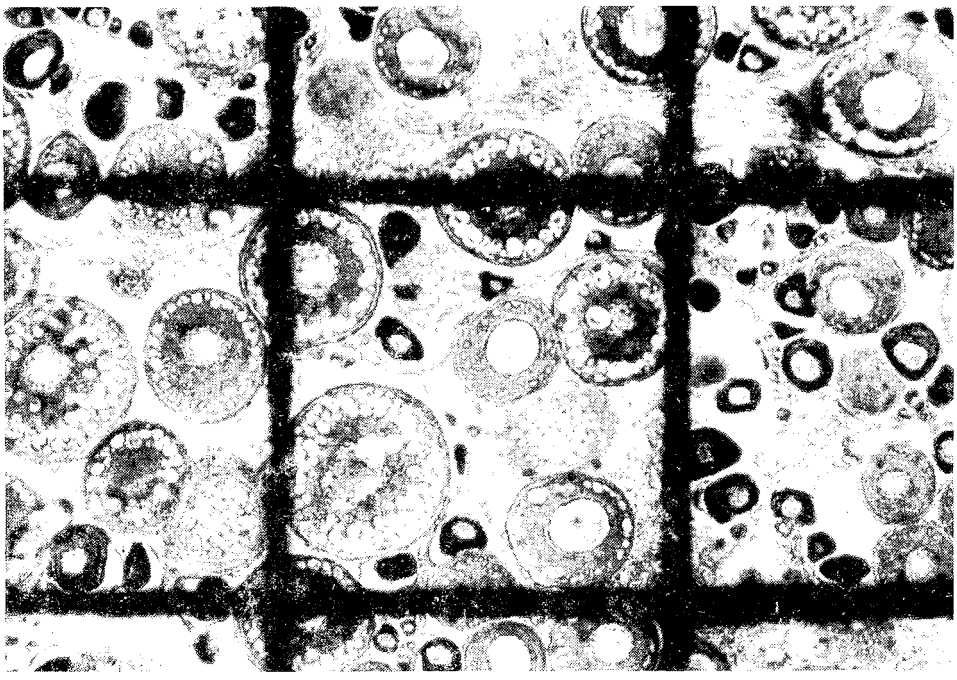

Photo 1. The grid used in quantitative study (x 60)

2. Number of atretic oocytes and oogonia throughout the year, were estimate using a 5-degree scale.

3. Atretic oocytes totally in stages α and β, and totally in stage γ and in course of follicular envelope resorption. These oocytes were counted only during the reproductive season (June-August). Average number of atretic oocytes in a given sample of tench females was calculated dividing their total number by fish number in this sample.

Oocyte diameter was also measured in the histological preparates (Tab. 2, column a). An ocular with 8 x magnification was used together with the objective, magnification of the latter differing depending on the measured cell: oogonia A – objective magnification 40 x, oocytes C and D – 20 x, oocytes E, – 10 x. Due to irregular shape of oocytes B, average diameter was calculated. An objective magnification of 40 x was used, and the oocytes were measured along longitudinal and transversal axis.

It is impossible to use directly the data from histological examinations to study the fish fecundity. This is due to the fact that cells preserved in formalin and AFA shrink, the extent of this being different for different cells. It was found experimentally (Pimpicka 1986) that oocytes measured in histological sections (preserved in AFA) were about 30% smaller than those preserved in formalin. The latter are usually used for fecundity estimates. Consequently, diameter of cells preserved in formalin

(Tab. 2, column c), corresponding to diameter of particular oocytes in the histological sections, was calculated as follows:

$$b : 70 = c : 100$$

where: b – diameter of the oocyte measured in a histological section,
c – the looked for diameter of the oocyte preserved in formalin.

Due to considerable differences in sizes of cells in particular stages, minimal and maximal values were used in the calculations (Tab. 2) column a), with a range of meam diameter variability ($\bar{x}$, Tab. 2). values of b were obtained adding and substracting standard deviation (SD) from the mean ($\bar{x}$) diameter (Tab. 2).

RESULTS

Histological picture and size of reproductive cells visible in the cross-sections of tench ovaries in the annual cycle

The following stages of reproductive cells were observed in the histological sections throughout the year:

a. Oogonia (Fot. 2) – sex cells with large, round nucleus and narrow belt of surrounding plasma resembling the nuclear matter. Size of these cells ranged from 0.003 to 0.022 mm. They were especially numerous immediately after spawning.

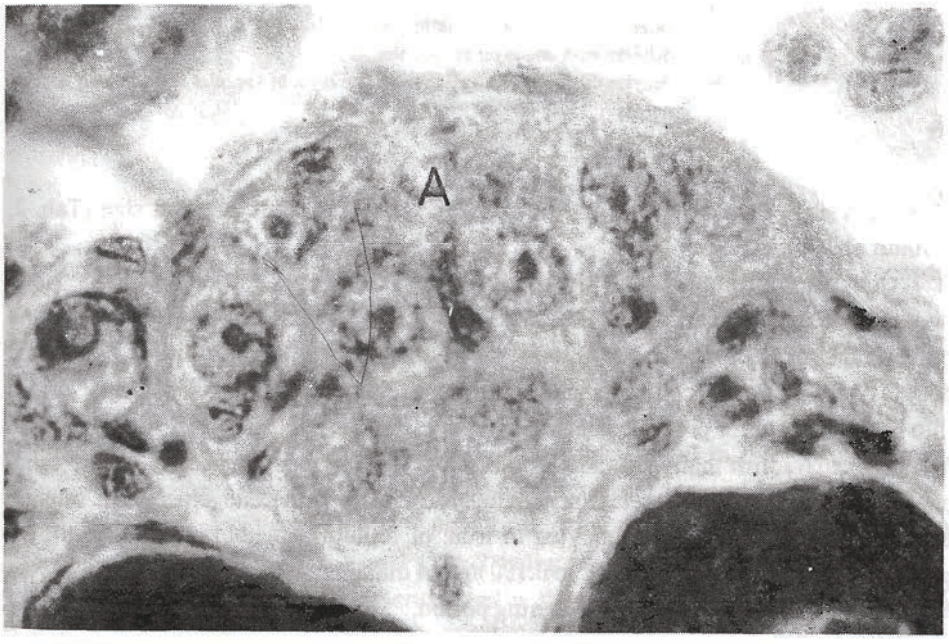

Photo 2. Oogonia in tench gonad (x 1000)
A – Oogonia

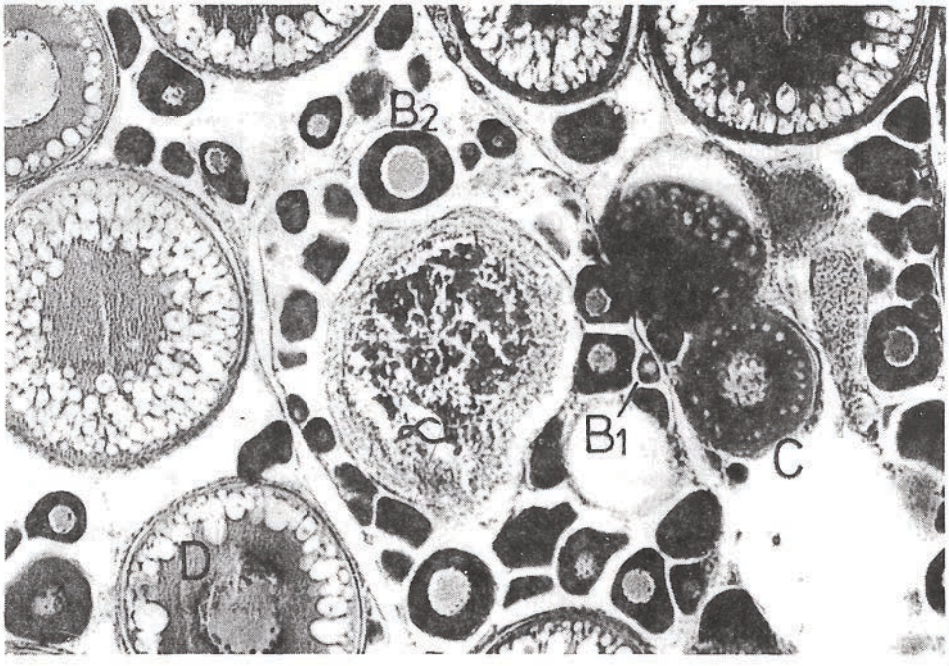

Photo 3. Cross section of tench female gonads at maturity stage III (x 100)
B_1 – Small oocytes of protoplasmatic growth
B_2 – Large oocytes of protplasmatic growth
C – Oocytes with one layer of vacuoles
D – Oocytes during vacuolization from one layer of vacuoles till the end of the process
α – First stage of atretic oocytes

b. Oocytes of the I order, differentiated with respect to structure and size (Tab. 2, column a). These were:

1. Oocytes of protoplasmatic (small) growth B (Fot. 3, 4). These could have been divided into oocytes B_1 (Fot. 3, 4) possessing a narrow belt of cytoplasm during the early stage of protoplasmatic growth, and slightly brighter cell nucelus. These oocytes grew systematically in size, increasing the amount of cytoplasm, and passed into bigger oocytes of protoplasmatic growth B_2 (Fot. 3, 4). Nuclei of B_2 oocytes were large, circular, with many nucleoli attached to the nucleus membrane. Their cytoplasm stained blue with haematoxylin. By the end of previtellogenesis, an outside envelope formed around the oocytes in from of follicular membrane. Oocytes B of protoplasmatic growth were 0.012 to 0.190 mm in diameter (Tab 2, column a). Oocytes B were biggest in April, prior to spawning period.

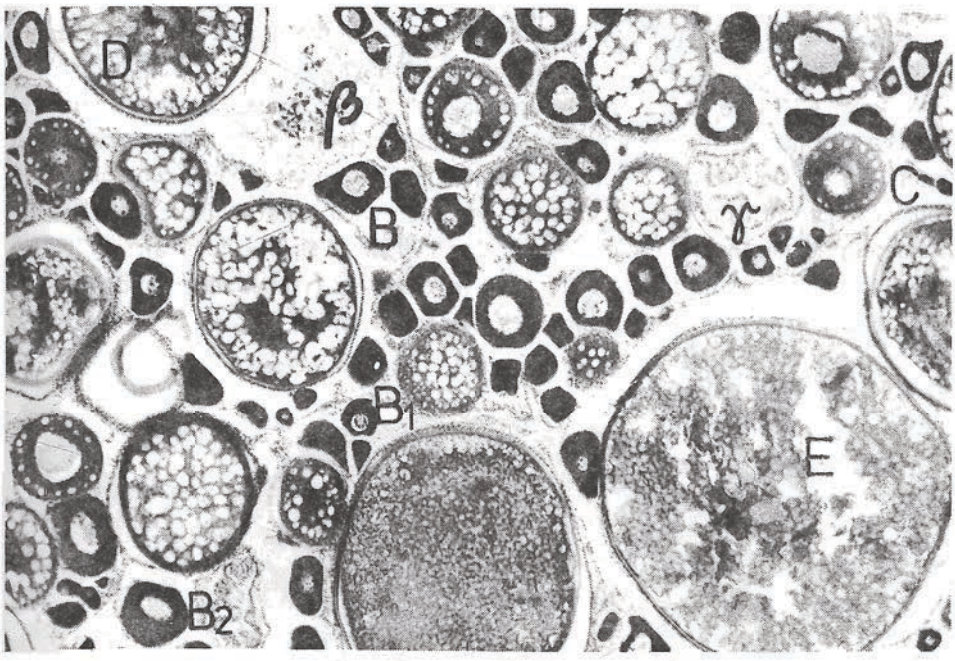

Photo 4. Cross section of tench female gonads at maturity stage VI/III-IV (x60)

B_1 – Small oocytes of protoplasmatic growth
B_2 – Large oocytes of protoplasmatic growth
C – Oocytes with one layer of vacuoles
D – Oocytes during vacuolization from one layer of vacuoles till the and of the prosess
E – Oocytes in vitellogenesis
β – Second stage of atretic oocytes
γ – Third stage of atretic oocytes and the EFE empty follicles envelopes at the first stage of atresia

2. Oocytes of trophoplasmatic (large) growth . These were differentiated according to the accumulation of trophic substances into.

– Oocytes C, begining with the cells in which the first vacuoles appeared at the cytoplasm perimeter, and ending with those which contained complete row of vacuoles. They were observed in the histological picture throughout the year (Fot. 3, 4). Nuclei of these cells were circular or elipsoid, with nucleoli in the peripheral zone. Zona radiata commenced to form between oocyte membrane and follicular envelope. Diameter of these cells was 0.095 – 0, 285 mm; the cells were observed in the histological picture throughout the year (Tab 2, column a).

– Oocytes D: since the moment when a second row of vacuoli formed in the cytoplasm till the whole cytoplasm was filled with vacuoli. The outline of these cells was not well visible; only nuclei were easily seen. Similary as oocytes C, also oocytes

D, were observed in the histological picture throughout the year (Fot. 3, 4). Diameter of these oocytes was 0.150 – 0.470 mm (Tab. 2, column a).

– Oocytes E were present in the histological picture since May till August i.e. prior to and during spawning season. This was connected with batch spawning of tench (Fot. 4). Oocytes E were filled with yolk grains and fat drops. Accumulation of yolk commenced from the cell nucleus and proceeded towards micropyle (Fot. 5). Diameter of these oocytes ranged from 0.22 to 0.90 mm (Tab. 2, column a).

c. Atretic oocytes in different stages of development (vacuolization and vitellogenesis, Fot. 3, 4) and follicular membranes after reproduction (Fot. 4).

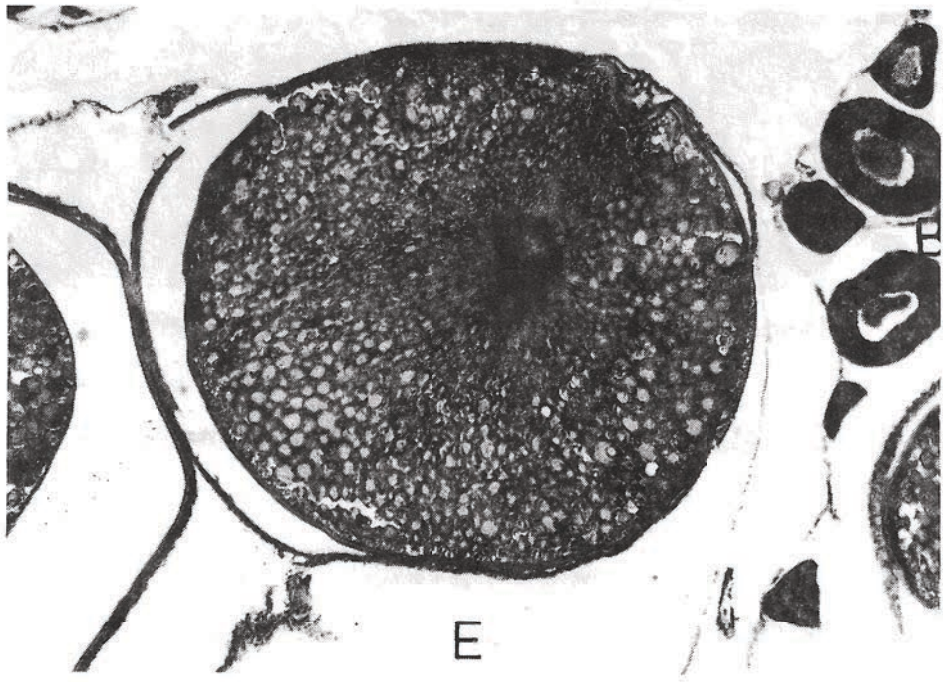

Photo 5. Oocyte E with micropyle (x 400)
E—Oocytes in vitellogenesis

Diameter of oocytes B, C, D, E (Tab. 2, column a) suggests that oocyte size is characteristic for particular stages of cell development and relatively constant throughout the annual cycle. This means that passing of an oocyte into another stage is determined not only by changes taking place in the nucleus, cytoplasm, and follicles, but also by the oocyte size (diameter). In view of this, it seems possible (with high probability) to classify the oocytes isolated from an ovary to the development stages (B, C, D, E) in order to estimate tench fecundity. Recalculations showed

that diameter of oocytes preserved in formalin was: for oocytes B from 0,043 to 0.169 mm, oocytes C from 0.200 to 0.321, D from 0.303 to 0.557, and E over 0.596 mm (Tab. 2, colimn c).

Mean values calculated for oocytes preserved in formalin suggest that oocytes of the diameter from 0.2 mm onwards should be taken into account in estimating the fecundity. This way, oocytes C which commenced trophoplasmatic growth are also included. Although few of these oocytes were present in the histological picture (this stage is very brief), their omitting might lead to underestimation of the fish fecundity.

Percentage of the oocytes of trophoplasmatic (C, D, E) and protoplastamic (B) growth in the ovaries of tench females in the annual cycle

Examination of the histological picture of tench ovaries revealed that in Lake Drwęckie the ovaries were in stage III of development since September till April, i.e. for eight months (Tab. 1). A number of processess took place in this stage, which changed the pools and structure of the reproductive cells. However, it was not yet possible to state which oocytes would attain maturity for the coming reproductive season. The following processes were observed in this stage: multiplication of oogonia (Fig. 1) and their passing as B_1 cells into prophase I of meiotic division, growth of the cells protoplasmatic growth B_2, passing of the latter into the large growth stage, in which cells C were formed, and these later became cells D. The latter underwent further vacuolization and increased in diameter. In all years of the studies, vitellogenesis began in May (temperature > 10°C, Fig. 2). The pool of oocytes of trophoplasmatic growth (potential fecundity) was established in May and June. These oocytes attained successively full maturity and were spawned in consecutive batches (physiological fecundity). Some mature cells were resorbed, decreasing potential (absolute) female fecundity (Fig. 1).

From May till the end of the reproductive season, stages of ovary development were very differentiated (Tab. 1), and so were the pools of trophoplasmatic oocytes in the ovaries of particular tench females (Tab. 1, Fig. 1, 3).

This differentiation suggests that oocytes developed asynchronously, and so did the ovaries of tench females in natural spawning populations in big lakes (Pimpicka 1989).

Analyses of the annual cycle of tench ovaries showed that samples used to estimate the fish fecundity should be collected by the end of May or at the begining of June, possibly just prior to spawning of the first egg-batch (Pimpicka 1989).

Data presented in Table 1 a, b, c and Fig. 1 show that percentage of oocytes CDE of trophoplasmatic growth differed in particular months as well as in particular years, from 4.9 to 31.9% of all oocytes. These differences were probably due to the processes

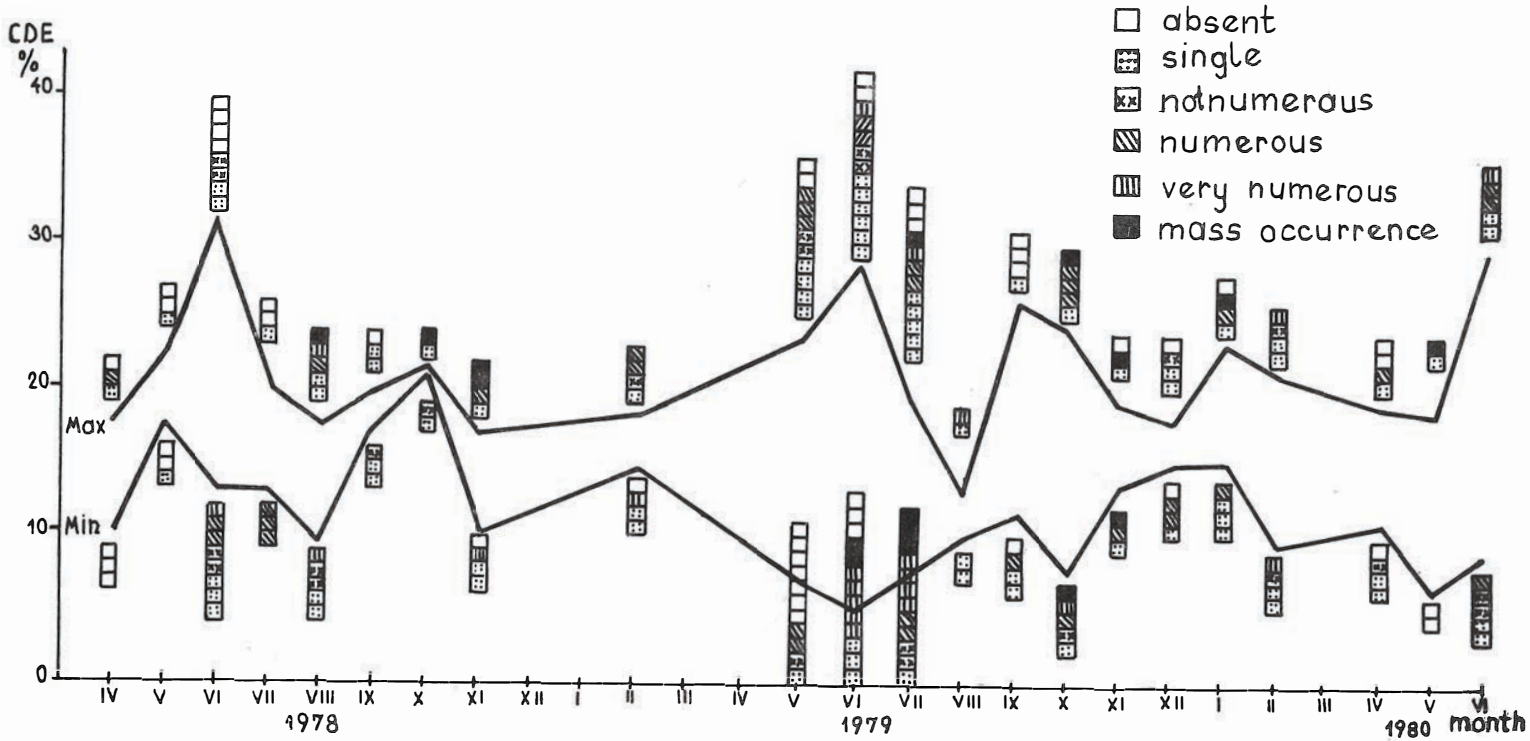

Fig. 1. Changes in percentage of trophoplasmatic growth oocytes (%, C, D, E) in tench gonads in 1978, 1979 and 1980 and occurence of oogonia (upper bar) and atretic oocyte (lower bar)

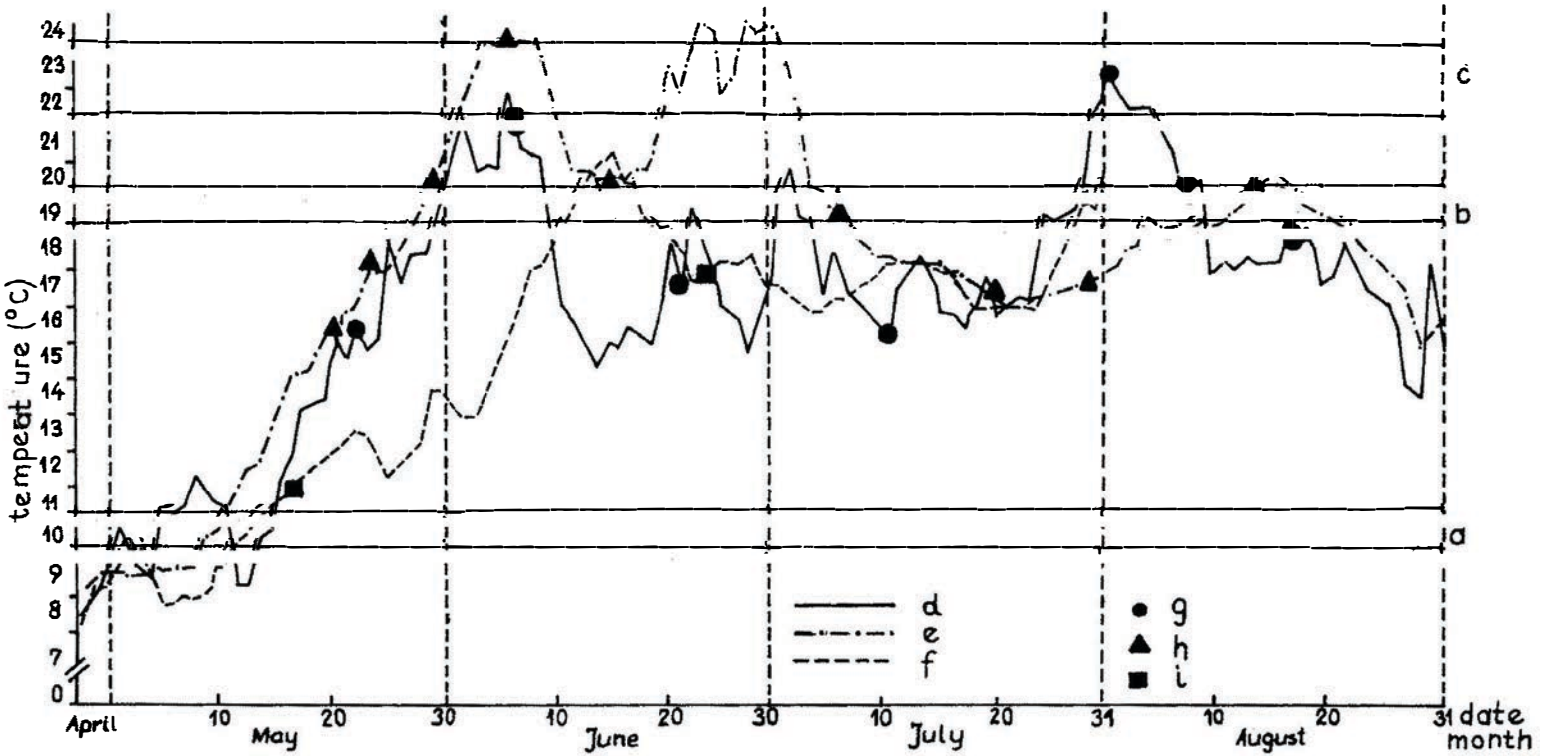

Fig. 2. Temperature of water during of tench reproduction period (d – 1978, e – 1979, f – 1980) and dates of fish sampling (g, h, i); a – temperature of stimulated vitellogenesis, b – temperature evoking spawning, c – optimal temperature of tench reproduction

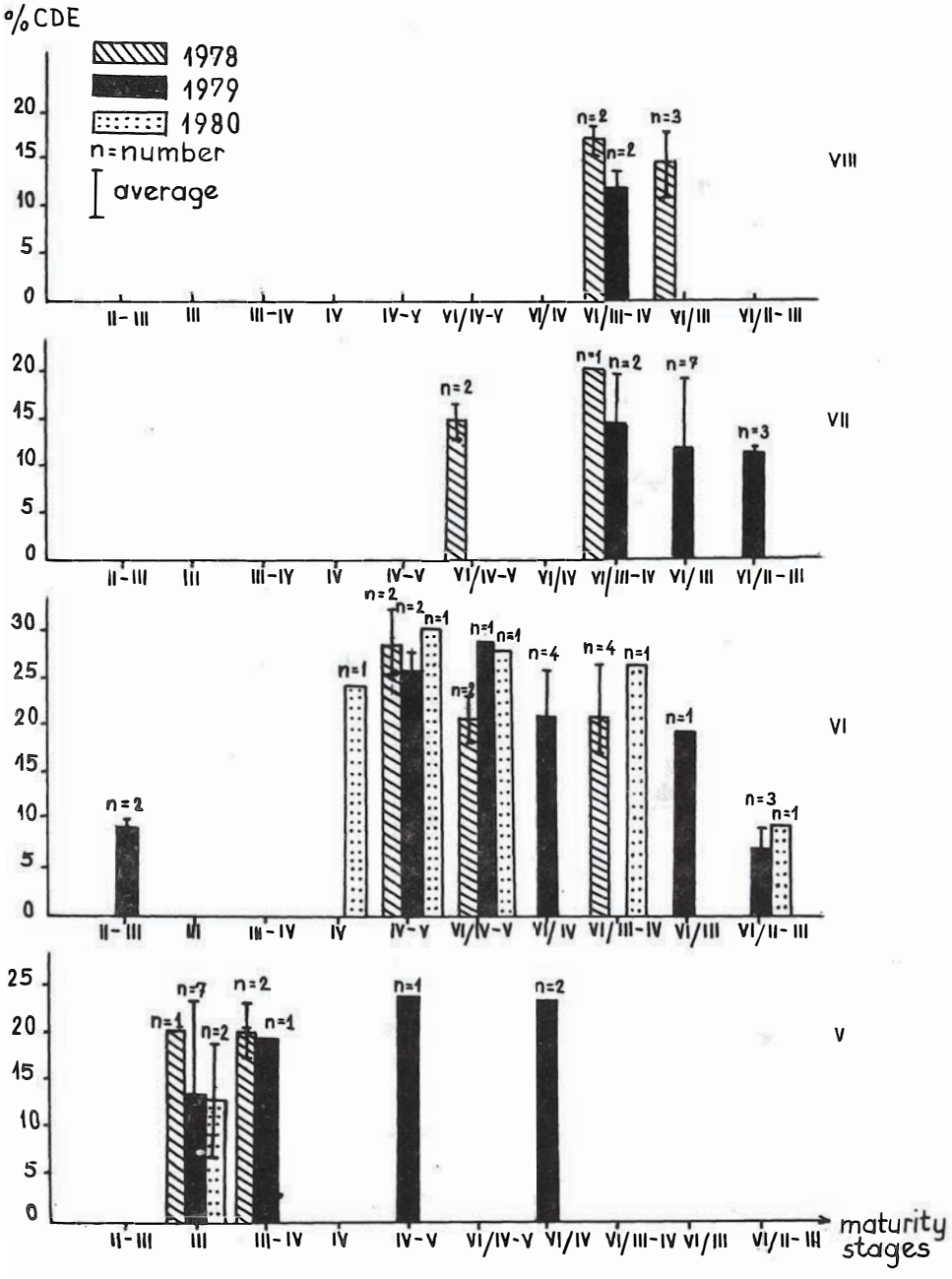

Fig. 3. Percentage of trophoplasmatic growth oocytes (%, C, D, E) at different stages of gonad maturity during tench reproduction period

of multiplication, maturation and resorption of the reproductive cells in the ovaries, but possibly also to other factors, such as thermal conditions in particular years (Fig. 2), different sample numerosity, varying size of the females (Tab. 1).

In the reproductive season of 1978 (Tab. 1a, Fig. 1, 3), percentage of trophoplasmatic oocytes gradually increased in May and June, from 12.9 to 31.9%. The latter value was the highest recorded during the studies. In this year, water temperature increased steadily since the second decade of May till the begining of June (Fig. 2). In July and August, when consecutive egg batches were spawned, percentage of the oocytes of trophoplasmatic growth was lower: 9.6 – 19.9% (Tab. 1a, Fig. 1, 3). At the same time, considerable resorption of the developing oocytes D and E was observed. It was caused by temperature variations in June, July and August (Fig. 2, 4).

In 1979, percentage of the oocytes of trophoplasmatic growth showed different trend. This was probably connected with different thermal regimes in the two years (Fig. 2). In 1979, temperature increased steadily already since the first decade of May, and spawning commenced earlier than in 1978, already by the end of May. In three females collected on 30 may, percentage of the oocytes CDE was very high, over 23% (Fig. 3, Tab. 1b). In June, percentage of these oocytes differed considerably between the females, from 4.9% to 28.6% (Fig. 1, 3, Tab. 1b). Most probably, the first egg batch was spawned at different time. The stage of gonad development differed accordingly (Tab. 1b, Fig. 3). In addition to this, oocytes D and E were intensively resorbed due to rapid variations of water temperature (Fig. 2). In July, fish samples were collected after a drop of water temperature which caused oocyte resorption (Fig. 1, 4), and resulted in considerable differences as to the pool of the oocytes of trophoplasmatic growth in particular females. This percentage ranged from 6.8 to 19.3% (Tab. 1b, Fig. 1, 3). In August, percentage of CDE was 10.1 – 12.9% i.e. lower than during the rest period (Tab. 1b, Fig. 1, 3), suggesting that the females had completed the reproduction.

In May 1980, water temperature was exceptionally low (Fig. 2), and vitellogenesis commenced only in the second decade. As a result, percentage of trophoplasmatic oocytes was very low: 6.4 – 18.5%. In June, percentage, of CDE oocytes reached 9.0 – 30.2%, in accordance with gonad development in particular females (Tab. 1c, Fig. 1, 3).

This period of tench reproduction was characterized by the most pronounced differences in percentage of the trophoplasmatic oocytes (Tab. 1a, b, c, Fig. 1, 3). This was caused by different gonad development in particular females (Tab. 1a, b, c, Fig. 3). Calculations (Tab. 1 a-c) presented in Fig. 3 showed that the lowest percentage of the trophoplasmatic oocytes was observed in females with ovaries in II-III and VI/II-III stage of development; it was only 6.9-10.3%. The highest percentage was observed in ovaries in stage IV-V and VI/IV-V of development, in June in all

years of the studies (Tab. 1 a-c, Fig. 3). This suggests that fecundity for the current reproductive season was determined in June.

Stage VI/IV-V of gonad development was determined basing on the presence of empty follicles in the histological picture as well as of indicatory oocytes. E. This stage does not necessarily mean that the female has already spawned the first egg batch. It can be observed also in females in which part of mature oocytes E has been liberated from follicular envelopes during sampling. Thus, similar percentage of CDE oocytes for females in stage IV-V and VI/IV-V, VI/IV of development, caught at the same time (Tab. 1a, b, c, Fig. 3). In consecutive months, July and August, subsequent egg batches were spawned, or else (if there was no such possibility) resorbed. Consequently, percentage of the oocytes of trophoplasmatic growth decreased in the ovaries in stages VI/IV-V, VI/III-IV, VI/III of development (Tab. 1, Fig. 3). These trends showed that the pool of the trophoplasmatic oocytes (potential fecundity formed in May and June) was used to spawn consecutive egg batches, so that by the end of the reproductive season this pool became similar to that during the rest period, or even lower. This suggests that the oocyte pool was not supplemented in any significant way the oocytes B of protoplasmatic growth. Large numbers of resorbed oocytes D, and especially E (Fig. 4), were due to temperature variations (Fig. 2) and certainly limited the amount of spawned eggs. Due to this, absolute (potential) fecundity calculated before the spawning period was much higher than physiological fecundity. Intensity of vitellogenesis was also affected by water temperature. Thus, the pool of CDE oocytes (determining fecundity in the given season) also depended on temperature (Pimpicka 1989).

In September and October, following a long spawning season, percentage of the trophoplasmatic oocytes increased again (oocytes C and D, Tab. 1a, b, Fig. 1). Tench feeds intensively in these months, this being a probable cause of the oocyte development and subsequent vacuolization. In consecutive months, in autumn and winter, percentage of the oocytes B of protoplasmatic growth increased (Tab. 1a, b, c). These were probably supplied from the pool of oogonia A which were very numerous at that time (Fig. 1). At the same time, resorption of the oocytes of trophoplasmatic growth was very intensive (Fig. 1).

Quantitative analyses revealed that number of the oocytes of protophasmatic growth in tench ovaries was much higher throughout the whole year than number of the oocytes of trophoplasmatic growth (Tab. 1a, b, c). Percentage of oocytes B was 68.1 – 95.1% of all oocytes, and the ratio B : CDE was from 2.1 : 1 to as high as 19.4 : 1. This means that percetage of protoplasmatic oocytes in the ovaries was from 2.1 to 19.4–fold higher than of trophoplasmatic oocytes all year round. Such a large reserve of the oocytes B of protoplasmatic growth suggests that mature cells in the given reproductive season are formed from the oocytes of protoplasmatic growth only.

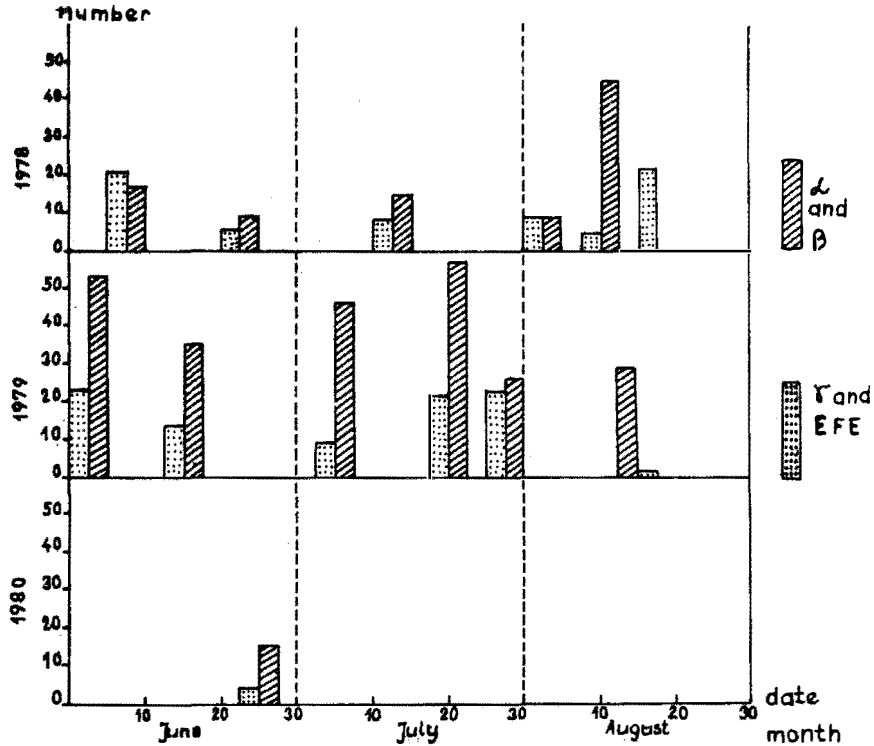

Fig. 4. Number of atretic oocytes at different stages of development in tench gonads during months: June, July and August in 1978, 1979, 1980. α– first stage of stretic oocytes, β – second stage of atretic oocytes, γ – third stage of atretic oocytes and EFE empty follicle envelopes at the firest stage of atresia

DISCUSSION

In fish of multi-batch spawning it is very difficult to separate the rteproductive cells that are to be spawned in the given season from those representing the so-called reserve. Hence, the main problem in studies of these fish consists of finding out a reliable criterium for distinguishing the developing and the reserve oocytes (Bagenal 1978).

Absolute fecundity of tench is usually expressed as the number of oocytes of trophoplasmatic growth (Kaj et al. 1964, Skóra 1964, Moroz 1968, Brylińska et al. 1979, Pimpicka 1981, 1986). These authors assumed that only the oocytes which were in the phase of trophoplasmatic growth before the first egg batch has been spawned were likely to be spawned in the same season. Other oocytes present in the ovaries were of protoplasmatic growth and constituted a reserve for the next and the subsequent years. However, estimates of fecundity were based on oocytes of different size, depending on the author.

My own studies and recalculations showed that size of the oocytes of trophoplasmatic growth was as follows: C from 0.200 mm to 0.321 mm, D from 0.303 to 0.557 mm, E over 0.596 mm (Tab. 2, column c). The results of diameter measurements (Tab. 2, column c) revealed that in calculating the absolute fecundity, oocytes of dimeter from 0.2 mm should be taken into account. Vibrickas (1968) also included oocytes from 0.2 mm in diameter, whereas Brylińska et al. (1979), Pimpicka (1981, 1986), Moroz (1968), Mcrawska (1981, 1982, 1984) took only oocytes bigger than 0.3 mm i.e. they excluded part of oocytes C. Monič (1953), Čeban (1975) and Zubenko (1975) took into account only the oocytes bigger than 0.4 mm, thereby excluding not only oocytes C but also part oocytes D. Consequently, absolute fecundity calculated by these authors was underestimated.

In sexually mature fish of batch-spawning, development of the reproductive cells (from oogonia to mature eggs) takes place continuously. Oven (1976), Sakun and Buckaja (1968) are of the opinion that the process of protoplasmatic oocyte growth is longer than one sexual cycle. According to these authors, it can last even for 3 years, its length being different in different fish species. In view of this, development of the reproductive cells for the given spawning season commences from oocytes B, and not from oogonia, i.e. from the oocytes which have already completed their protoplasmatic growth.

In the histological picture of tench ovaried from Lake Drwęckie, percentage of the oocytes of protoplasmatic growth was from 2.1 to 19.4 times higher than of the oocytes of trophoplasmatic growth all year round. It amounted to 68.1 – 95.1% (Tab. 1a, b, c).

This large reserve of protoplasmatic oocytes confirms the results by Oven (1976) and Sakun, Buckaja (1968).

Decrasing percentage of the oocytes of trophoplasmatic growth in course of the spawning season (Fig. 3) suggests that absolute fecundity estimated at the begining of this season is not supplemented from the pool of the oocytes of protoplasmatic growth. The same opinion was presented by Oven (1976) and Crossland (1977) for other fish species of batch-spawning. Only the fish inabiting thermally polluted waters do not conform to this rule. Oven (1976) cited Peters (1957) and Egami (1959) as well as his own results on some fish from the Black Sea, while Morawska (1981, 1982) presented the results of studies on tench cultured in heated ponds. These authors found that absolute fecundity formed during spawning might change basing on the reserve of tche oocytes of protoplasmatic growth Additional egg batches (over three) were formed with the oocytes which had been classified as a reserve before the spawning season.

The ratio between oocytes of protoplasmatic and trophoplasmatic growth was studied and discussed for many fish species. Mestorff (1959) was the first to state that in *Merlangus merlangus* (L), the ratio of immature to mature oocytes was as 4 :1. He estimated the reserve of cells of protoplasmatic growth before and after spawning, and concluded that protoplasmatic growth lasted in this fish for 3 years. Göting (1961) studied 10 Teleostei species and stated that number of the oocytes of protoplasmatic growth was 5 times higher than of trophoplasmatic ones. He also suggested that percentage of the reserve oocytes increased with fish age. Dunn and Tyler (1969) that percentage of immature oocytes was 66.4, and of mature 33.6, the latter being either spawned or resorbed. Yamamoto and Yamazaki (1961) examined oocyte numbers and development in *Carassius auratus* and found that absolute fecundity depended upon the pool of the oocytes of protoplasmatic growth (reserve) which attained this stage a year before the fish reproduction. These authors used their own scale of development and divided the oocytes into three groups; two of these probably developed and were spawned in the given season, the third remained in the ovaries and represented a „base" for the next reproductive season. Yamamoto and Yoshicka (1964) found that ovaries of *Oryzias latipes* (a fish of batch-spawning) contained 82.6% of the oocytes of protoplasmatic growth immediately after the spawning season. Crossland (1977) found in *Chrysophrys auratus* that oocytes of protoplasmatic growth dominated throughout the annual cycle, constituting from 47.6 to 97.0%. This author is of the opinion that development of the reproductive cells from oogonia to mature eggs lasted 3-4 years. Kopiejewska (1983, 1989) studied annual cycle of bream (one-portin spawning) and found that reserve oocytes constituted from 78.9 to 96.6% of all oocytes in the ovary. Monič (1953) described annual cycle of sexually mature tench females and mentioned that reserve oocytes were 4-5 times more numerous than those spawned during the reproductive period. This author suggested that in West Siberia, oocyte development lasted for 3 years.

The above studies suggest that the reserve oocytes of protoplasmatic growth in different fish of batch-spawning were much more numerous than those which formed absolute fecundity in the given season. My studies confirm this suggestion.

Reserve of reproductive cells is determined by oogonial divisions. I have observed oogonia in tench ovaries all year round; their number increased after the spawning period (Fig. 1). The same was observed by Kazanskij (1949) in tench and by Crossland (1977) in *Chrysophrys auratus*. Hence, it may be stated that the pool of oocytes B of protoplasmatic growth, which constitute a reserve of reproductive cells in tench ovaries, was supplemented with developing oogonia.

Also the pool of oocytes of trophoplasmatic growth is constantly changing (Fig. 1). Not all of them are spawned. Some undergo resorption and this phenomenon is responsible for the difference between absolute fecundity (calculated before the first egg batch has been spawned) and physiological fecundity of tench females. I have observed that atretic oocytes of various stages were present in tench ovaries during the whole annual cycle (Fig. 1). The same was observed by Kazanskij (1949). Resorption processes in fish with batch spawning do not inhibit the development of other oocytes, and may occur simultaneously with maturation and ovulation of the consecutive egg batches (kazanskij 1949, Šichšabekov 1974, 1985, Oven 1976, Morawska 1982). I have also observed that oocyte resorption was most intensive during spawning period. Intensity of this process depended on temperature. Oocyte resorption was very intensive (Fig 1, 4) when water temperature changed rapidly (Fig. 2).

Thermal regime determined also the intensity of vitellogenesis (Pimpicka 1986, 1989). Consequently, it influenced formation of CDE oocytes of trophoplasmatic growth and, thus, also the fish fecundity in the given season.

CONCLUSIONS

1. Diameter of the oocytes of trophoplasmatic growth (preserved in formalin), which should be taken into account in estimating absolute fecundity, was as follows: C from 0.200 to 0.321 mm, D from 0.303 to 0.557 mm, and E from 0.596 mm.

2. In the annual cycle, the reserve of oocytes of protoplasmatic growth in the histological picture of tench ovaries was from 2.1 to 19.4 times higher than number of the oocytes of trophoplasmatic growth.

3. Large reserve of the oocytes B of protoplasmatic growth suggestis that their pool is sufficient to from fecundity in the given reproductive season in a natural water body, i.e. that it is not supplemented by oogonial development.

4. In the histological picture, the pool of the oocytes of trophoplasmatic growth represented from 4.1 to 31.9% of all oocytes present in the ovary in the annual cycle.

5. Percentage of CDE oocytes in tench ovaries decreased since the begining of the reproductive season, suggesting that absolute fecundity esimated prior to this period (on the basis of CDE pool) was not significantly supplemented from the pool of oocytes B.

6. Resorption of the oocytes took place throughout the annual cycle. It was most intensive during spawning. Due to this, absolute fecundity calculated prior to spawning was always higher than physiological fecundity i.e. than the real number of spawned eggs.

7. Oogonia were observed in the histological picture all year round. They were most numerous immediately after the reproductive season. This suggests that the pool of the oocytes of protoplasmatic growth was supplemented in this period by the developing oogonia.

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FORMOWANIE SIĘ PŁODNOŚCI SAMIC LINA – *TINCA TINCA* (L.) W JEZIORZE DRWĘCKIM

STRESZCZENIE

Materiał do badań stanowiły jajniki 108 samic lina pobrane w okresie od kwietnia 1978 r. do czerwca 1980 r. (tab. 1a, b, c). Gonady utrwalono w płynie AFA, skrawki histologiczne barwiono hematoksyliną Delafielda z eozyną. W 30 kwadratacj siatki (0,1 mm x 0.1 mm), nałożonej na preparat liczono owocyty protoplazmatycznego (B) i trofoplazmatycznego (C, D, E) wzrostu a także przy pomocy pięciostopniowej skali określono szacunkowo liczebność owocytów atrezyjnych oraz owogonii. Wykonano pomiary owocytów i otrzymane średnice mikroskopowe komórek B, C, D, E przeliczono na średnice makroskopowe tych komórek (średnice makroskopowe są to średnice owocytów utrwalonych w formalinie które są brane do obliczeń płodności absolutnej).

Przeprowadzone pomiary i przeliczenia pozwoliły na stwierdzenie, iż średnice owocytów utrwalonych w formalinie są następujące: protoplazmatycznego wzrostu B od 0,043 mm do 0,169 mm; trofoplazmatycznego wzrostu C od 0,200 do 0,321 mm, D od 0,303 do 0,557 oraz E od 0,596 mm (tab. 2, kolumna c). Obliczenia te wskazują, że do określenia płodności powinny być wliczone owocyty od średnicy 0,2 mm; żeby ująć wszystkie komórki trofoplazmatycznego wzrostu.

Przeprowadzone badania ilościowe wykazały, że w jajnikach lina w cyklu rocznym udział owocytów protoplazmatycznego wzrostu wynosił 68,1 do 95,1% (tab. 1a, b, c) wszystkich liczonych owocytów i był 2,1 aż do 19,4-krotnie wyższy od owocytów trofoplazmatycznego wzrostu, a stosunek B:CDE wynosił od 2,1:1 aż do 19,4:1. Taki wysoki zapas komórek protoplazmatycznego wzrostu wskazuje, że formowana w danym roku płodność potencjalna (absolutna) nie potrzebuje korzystać ze znajdującego się w jajnikach zapasu owogonii.

Zasób owocytów trofoplazmatycznego wzrostu w ciągu roku wynosił 4,9 do 31,9% (tab. 1a, b, c). Najwyższy udział procentowy owocytów trofoplazmatycznego wzrostu notowano we wszystkich latach badań przed okresem rozrodu (w miesiącu maju i czerwcu) (tab. 1a, b, c; rys. 1,3) oraz po rozrodzie (we wrześniu i październiku) (tab. 1a, b, rys. 1). W okresie rozrodu obserwowano największe różnice zasobu procentowego owocytów trofoplazmatycznego wzrostu u poszczególnych samic. Powodowane to było różnymi terminami składania pierwszej i następnych porcji jaj. Na różnice te miała również wpływ silna resorpcja dojrzewających owocytów występująca wskutek gwałtownych spadków temperatur wody w czerwcu 1978 i 1979 r., w lipcu oraz sierpniu 1978 r. (rys. 4). Te silne procesy resorpcyjne z pewnością obniżyły płodność fizjologiczną badanych samic lina czyli liczebność jaj złożonych na tarliskach.

Zmniejszający się udział procentowy owocytów trofoplazmatycznego wzrostu w jajnikach samic lina od rozpoczęcia tarła do zakończenia sezonu rozrodczego (rys. 3) sugeruje, iż oszacowana na początku danego sezonu rozrodczego płodność absolutna nie jest w znaczący sposób uzupełniana z zapasu owocytów protoplazmatycznego wzrostu.

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