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The annual reproductive cycle of the freshwater mussel *Dreissena polymorpha* Pallas in lakes

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Summary. The annual development of the gonads of *Dreissena polymorpha* was studied at three sampling sites in two lakes over 3 and 1½ years, respectively. A resting stage occurred after the last spawning in summer/autumn. Oogenesis (accompanied by multiplying segmentation of the oogonia and early growth processes of its oocytes) restarted in specimens at least 1 year old at low temperatures (below 10° C) during winter and early spring. At one location (Fühlinger See) the onset of the spawning season was correlated with an increase of water temperatures above 12° C. At 2 m depth, two main spawning periods in May and August were normally recognized, the first at temperatures of 12–16° C, the second at 16–21° C. It was clearly demonstrated for the first time in *Dreissena polymorpha* that the oocytes became mature in successive cohorts within one gonad. A female mussel may spawn several times during the reproductive season. At 9 m depth, the onset of spawning also started at about 12° C; this occurred in late summer, with two spawning periods within 1 month at a temperature range of 12–16° C. At another location (Heider Bergsee) the size of the gonads and the oocytes was reduced during April of both years studied, when food supply was low simultaneously with rapidly rising water temperatures in this shallow lake. There was no spawning period during spring. The major spawning period was delayed until July (temperatures 19–22° C). This shows (1) the synchronizing influence of low winter temperatures on the annual reproductive cycle and (2) a temperature threshold of at least 12° C for the start of the spawning processes. The results are discussed with regard to the geographical limits of further spread of *Dreissena polymorpha*.

Key words: Reproductive cycle – Gonad development – Oocyte size frequency distributions – Temperature threshold – *Dreissena polymorpha*

The freshwater mussel *Dreissena polymorpha* has become one of the most dominant species in many European waters, since it started spreading from the Caspian area at the beginning of the last century (Stańczykowska 1977). This success can be attributed to the passive transfer of adult mussels attached by their byssus threads to a wide range of substrates including ships, combined with the formation of pelagic larvae – common in marine invertebrates (Thorson 1946), but unusual for freshwater bivalves. In 1985, *Dreissena* even reached the Great Lakes of North America, entering the area via ballast water discharge (Hebert et al. 1989), and is possibly now starting to spread rapidly over the nearctic continent.

Although much is known about the distribution of *Dreissena*, detailed data on reproduction are rare.

1. Earlier studies have mainly concerned temporal occurrence, abundance and the development of the larvae in the plankton (e.g. Breitig 1965; Einsle 1973; Walz 1973; Kornobis 1977; Lewandowski 1982; Lewandowski and Ejsmont-Karabin 1983; Siller 1983; Stańczykowska et al. 1983). Discussing laboratory studies on the fertilization of *Dreissena* eggs (Sprung 1987), Sprung (1989) predicted that the onset of the spawning period during May must coincide with the water temperature rising above 12° C.

2. Laboratory experiments on inducing gamete release revealed that *Dreissena* is in principle able to spawn four times per season with a minimum delay of more than 7 days between two successful inductions (Walz 1978).

3. All authors studying the histology of the gonads of *Dreissena* agreed that there was an annual gametogenetic cycle with a high level of synchronization between the members of a population (Wlastov and Katchanova 1959; Antheunisse 1963; Galperina and Lvova-Katchanova 1972; Walz 1973, 1978; Borcherding 1986).

However, only few details were known of the different phases of the cycle, its correlation with seasonal and local conditions, the reason for the occurrence of several larval peaks per year, and the reproductive effort of young and old specimens. The main problem was to quantify the size of the gonads, because it was not possible to completely separate the gonads from the remaining tissues of

the visceral sack (Walz 1978; Borcherding 1986; see also Morvan and Ansell 1988 for *Tapes rhomboides*). Therefore a new method of quantitative and qualitative analysis of *Dreissena* gonads was applied in this study. Some new data we presented on gonad development and spawning period through the year, and the external factors controlling the timing of the reproductive cycle.

Material and methods

Study sites and sampling

Dreissena polymorpha was collected from three study sites in two lakes near Cologne by SCUBA diving. The first lake, Fühlinger See, is a 84-ha recreation area close to the river Rhine. The two sample sites in this lake (max. water depth 13 m) were at 2–3 m depth (FS–2m) and at 8–9 m depth (FS–9m), respectively, i.e. above and below the summer temperature discontinuity layer. The third study site was at 1–2 m depth in the Heider Bergsee (HS–2m), a shallow 47-ha lake (max. depth 6 m). The lakes differed in the amount of seston, with distinctly higher food concentrations for suspension feeders in the Fühlinger See, especially at FS–2m (for further description of both lakes see Sprung 1989).

Histological procedures

As it is not possible to separate quantitatively the gonads from the digestive tract in *Dreissena polymorpha* (Walz 1978; Borcherding 1986), histological techniques were used to determine the size of the gonads in order to analyse the gametogenetic cycle. Immediately after sampling, the visceral sack of the mussels (containing gonads, stomach, gut, digestive gland, byssus gland, foot, parts of the adductor muscle) was separated from the remaining tissues (gills, mantle, heart, kidneys, parts of the adductor muscle), fixed in Bouin-Allen's fixative, dehydrated in an ascending alcohol series and methylbenzoate and embedded in Paraplast Plus (melting point 56° C). Using a Leitz sliding microtome (Typ 1300) each visceral sack was completely cut in transverse sections (10 µm), thereby recording the lenght of the visceral sack (distance from the first to the last section). During this process about 15 serial sections were taken along the whole visceral sack, stained with Mayer's Haemalaun (MERCK) and eosin (Adam and Czihak 1964), and mounted in Canada balsam.

Tissue volume measurements

According to Delesse's principle (1848; as cited by Henning and Mayer-Arendt 1963), the ratio of areas of the individual components equals the ratio of their composition by volume. Thus, the estimation of volume of the various tissues is possible by an integration of tissue-area measurements over the whole visceral sack (Borcherding 1986, 1990; Morvan and Ansell 1988). A picture of each section, illuminated with a light-box, was taken with a CCD-Video camera (with a macro-objective). The areas of the gonads and the visceral sack were measured using an image analysing system (SIS GmbH, Münster). In order to calculate the volume, the mean tissue areas of each mussel were multiplied by the corresponding length of the visceral sack (see above). In addition, a gonad index (GI) was calculated as gonad volume (GV)/ visceral sack volume, percent.

During a preliminary study period (September 1984 January 1986), the mean (±95% confidence interval = c.i.) GV and GI values of up to 10 animals with a shell length of about 24 mm were measured. Subsequently 20 mussels of different shell lengths were chosen, just as they were found at the sampling site. For these samples, all individual GV and GI values and the shell length (SL) were fitted to the allometric equation $y = a \cdot SL^b$ (Reiss 1989), where y is the predicted GV or GI respectively; a and b are fitted parameters. In a second step, the values for GV or GI were transposed to a standard animal with a shell length similar to the 'mean shell length' at the corresponding sampling site (FS–2m and FS–9m: 24 mm; HS–2m: 20 mm; for a review of methods see Grant and Tyler 1983). The 95% confidence intervals were calculated according to Sachs (1984).

Oocyte analysis

For measurements of the oocytes with the image analysing system, the CCD-Video camera was linked to a Leitz light microscope with 100× magnification. Assuming that every oocyte area equals a circle (for details see Borcherding 1990) the diameters of 120–200 oocytes per female with a clearly visible nucleus (to ensure the central section through the oocyte) was measured. If a fraction of ripe oocytes was identified within the gonads a mean oocyte diameter for all values >40 µm was computed. Ripe oocytes were clearly round showing no attachment sites to the follicle wall, and in addition there was a distinct peak of large oocytes in the oocyte size-frequency distributions (see also Galperina and Lvova-Katchanova 1972).

The proportion of the gonad tissue consisting of oocytes (i.e. the gamete volume fraction, GVF, Bayne et al. 1978) was estimated with the image analysing system by means of a grey-scale analysis of 10 histological gonad pictures per animal (for details see also Borcherding 1990). The numbers of oocytes for every size class (width 4 µm) and for the whole mussel were calculated according to the following equations:

$$ON_{(I)} = \frac{4/3 \cdot \pi \cdot r_{(I)}^3 \cdot n_{(I)}}{\sum_{I=1}^{k} (4/3 \cdot \pi \cdot r_{(I)}^3 \cdot n_{(I)})} \cdot (GV \cdot GVF/100) \quad (1)$$

$$ON = \sum_{I=1}^{k} (ON_{(I)}) \quad (2)$$

where

$ON_{(I)}$ = the number of oocytes per size class I
ON = number of oocytes per animal
k = the number of size classes
$r_{(I)}$ = the mean oocyte radius of size class I
$n_{(I)}$ = the number of measured oocytes per size class I
GVF = gamete volume fraction within the gonad tissue
GV = gonad volume

The mean number of oocytes for one sample (g) was either the average of all mussels studied or the value for a standard animal with a particular shell length calculated from the fitted allometric equation (see above).

Results

Fühlinger See 2-m depth (FS–2m)

At the end of a spawning period each year, usually at the beginning of September, the gonads of *Dreissena polymorpha* had a disorganized follicle structure, so that it was usually impossible to identify the sex due to the lack of oocytes or sperms. This phase was called "resting stage", although the tissue may not be completely inactive. It is indicated by minimum values of the gonad volume and the gonad index (Fig. 1). The gonad volume

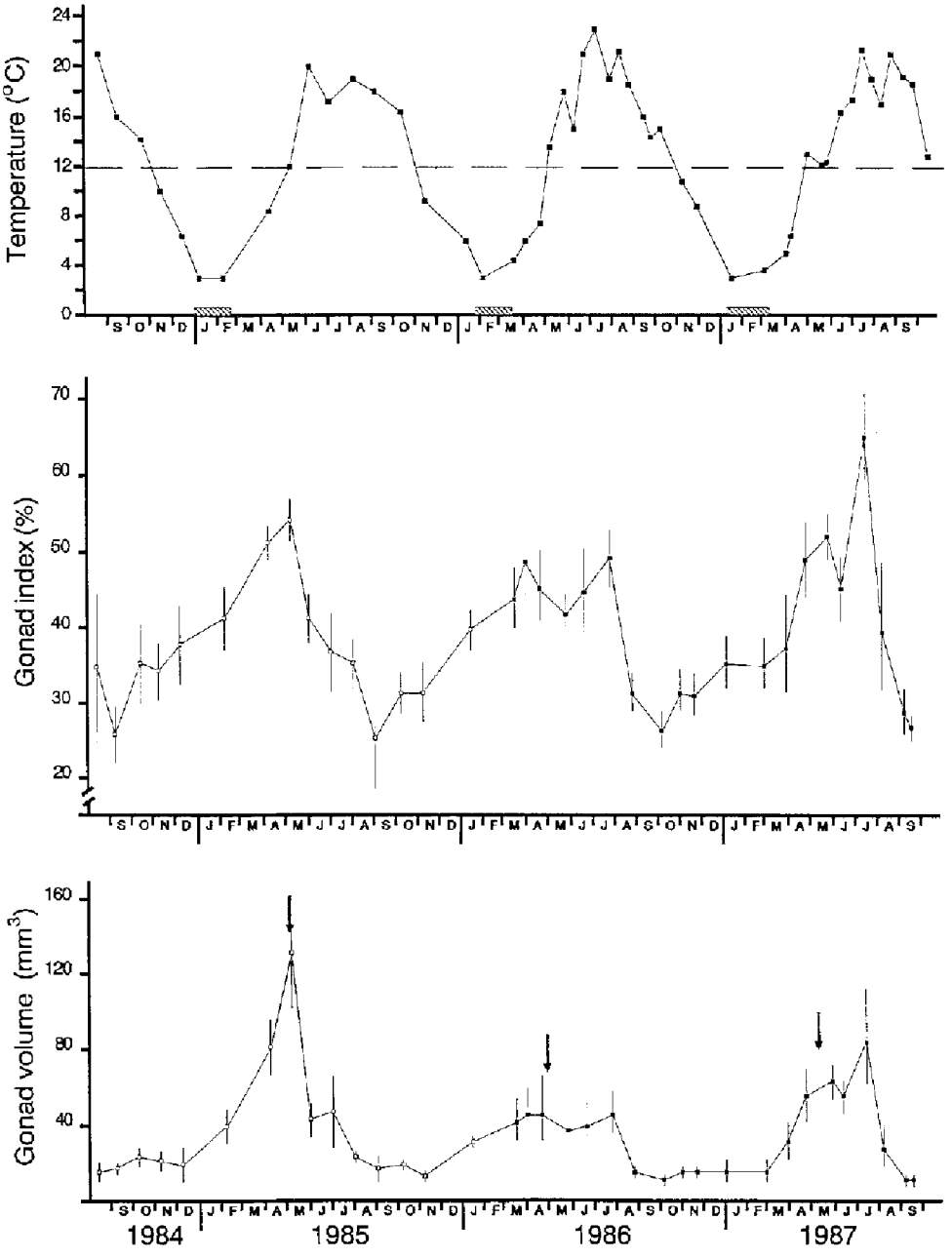

Fig. 1. *Dreissena polymorpha* (24 mm SL) at location FS–2m. Seasonal course of gonad volume, gonad index (*vertical lines*, 95% confidence intervals), and temperature at 2 m depth. *Open symbols:* mean values of specimens of about 24 mm shell length (SL); *black symbols:* values calculated from regressions (see Methods). *Hatched bars:* the period of ice cover. *Arrows:* the time when temperature rose above 12° C

remained low at least until the end of November (in 1986/87 until as late as the end of February). Normally gametogenesis restarted during the winter when the temperatures declined below ≈ 10° C, and was indicated by increasing gonad volume. Histological sections from the final growth phase of the oocytes and the following maturation and spawning in 1985 are shown in Fig. 2, and will be explained in detail together with the results of the oocyte analysis (Fig. 3).

The increase of gonad volume during February and April resulted from both an increase in the number of oocytes (e.g. FS–2m 1985: 780,000 to 1,070,000: Fig. 3) and oocyte growth (mean diameter increase 34.8 µm to 38.9 µm: Fig. 3; histological photographs see Fig. 2a, b). During this early period, frequency distributions of the oocyte sizes were clearly still unimodal. However, the increase in gonad volume from April to May was due only to the growth of the larger fraction of the oocyte population, while the number of oocytes remained nearly constant. In addition, the histological sections (Fig. 2c) showed for the first time mature oocytes which are distinct due to their round shape and the lack of attachment sites to the follicle wall. However, during this final phase of oocyte maturation, low growth of the oocytes could still be occurring.

The beginning of the spawning season was indicated by the first decrease of the gonad volume. This obviously took place in all years studied when the temperatures increased above 12° C (Fig. 1). During the first spawning period, for instance in 1985, a mean of nearly 400,000 oocytes per female (24 mm SL) was spawned. Immature oocytes were left behind (elongated oocytes with distinct

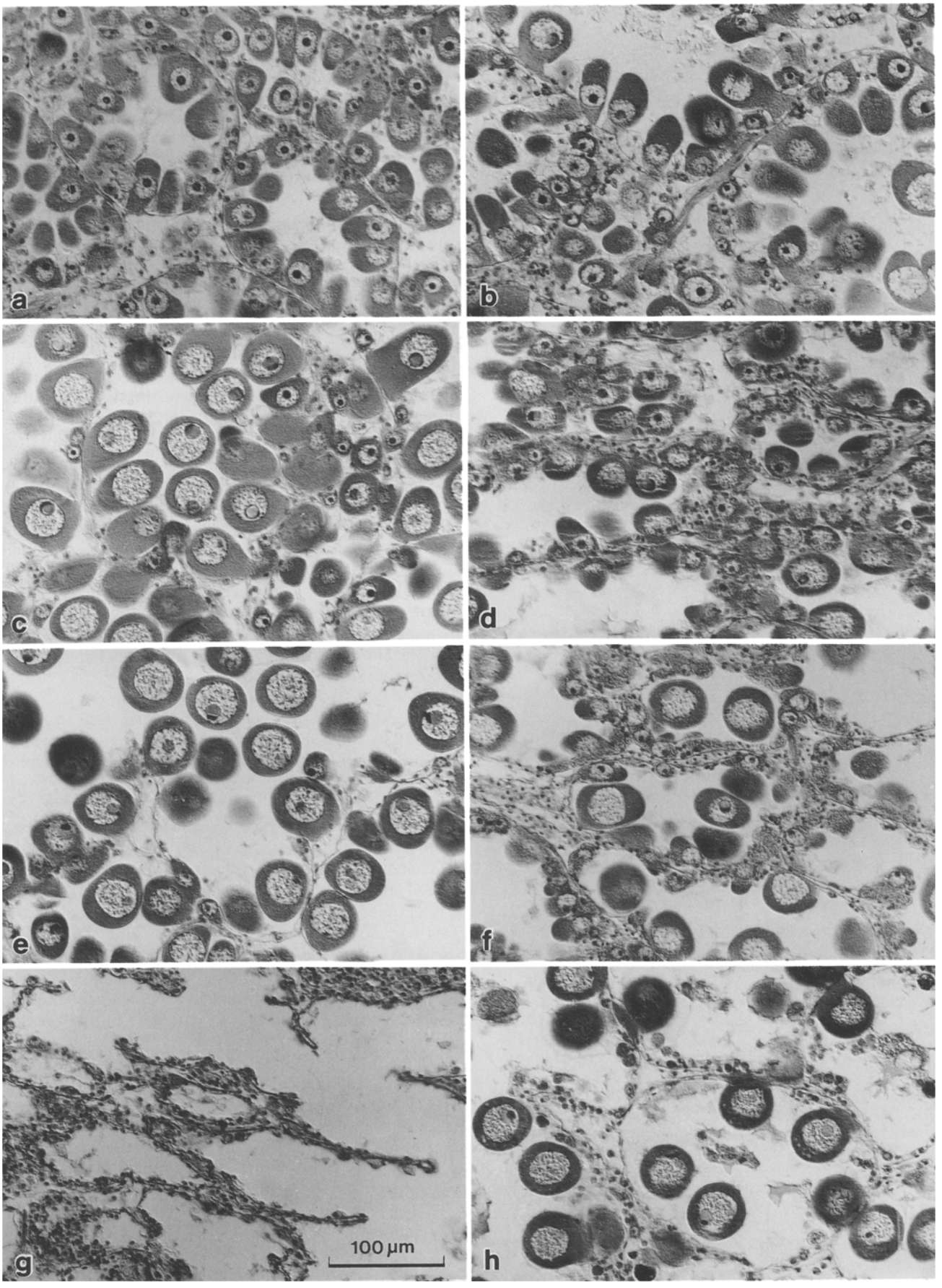

Fig. 2. *Dreissena polymorpha* at FS–2m in 1985. Histological sections of females (also shown in Fig. 3) collection **a** 7 Feb 1985, **b** 11 Apr 1985, **c** 9 May 1985, **d** 4 Jun 1985, **e** 2 July 1985, **f** 5 Aug 1985, **g** 4 Sep 1985 (specimen with empty gonads), **h** 4 Sep 1985 (specimen with some ripe oocytes remaining)

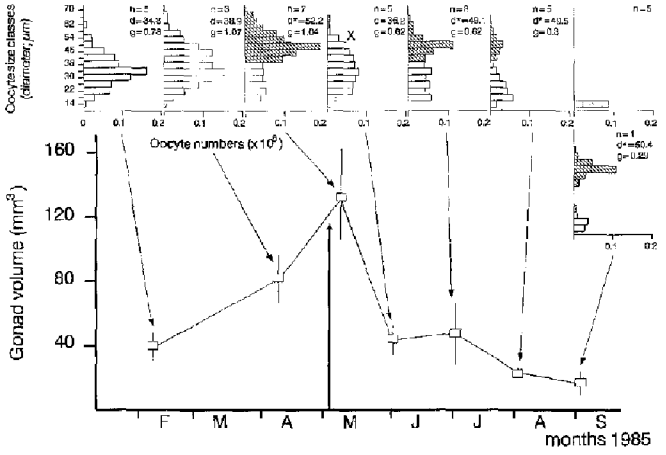

Fig. 3. *Dreissena polymorpha* (24 mm SL) at FS–2m in 1985. Gonad volume (1985 section of Fig. 1) and corresponding oocyte sizes (*y* axis in 4-µm size classes) plotted as frequency distributions (*x* axis: oocyte numbers $\times 10^6$). *n:* number of mussels; *d:* mean diameter of all oocytes; *d**: mean diameter of the oocytes of the ripe fraction *(striped bars)*; *g:* total numbers of oocytes per mussel ($\times 10^6$). *(X)* in June plot: as in April, the large oocytes were frequently classified as immature. If no exact oocyte analysis was possible, the estimated oocyte values are presented with *stippled bars*. The *arrow* in the gonad volume plot indicates the time when the temperature rose above 12° C

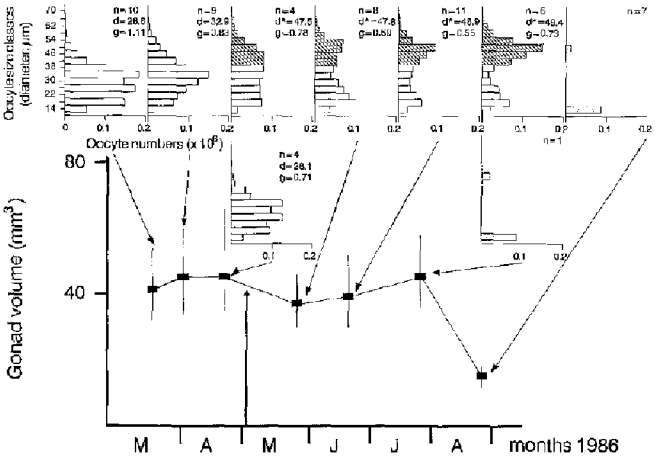

Fig. 4. *Dreissena polymorpha* (24 mm SL) at FS- 2m in 1986. Gonad volume (1986 section of Fig. 1) and corresponding oocyte sizes plotted as frequency distributions. Symbols as in Fig. 3

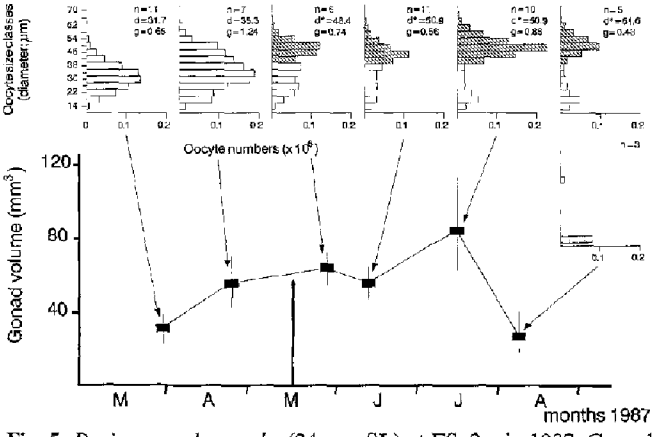

Fig. 5. *Dreissena polymorpha* (24 mm SL) at FS–2m in 1987. Gonad volume (1987 section of Fig. 1) and corresponding oocyte sizes plotted as frequency distributions. Symbols as in Fig. 3

attachment sites to the follicle wall, Fig. 2d, Fig. 3 June [X]). In contrast to the bimodal frequency distribution including both mature and immature oocytes in May, the histogram for early June was again unimodal (Fig. 3, June). Since the process was sampled only every 4 weeks, it was not possible to show whether this first spawning period of the FS–2m mussels covered several spawning events during these weeks, or whether there was one main synchronized spawning period lasting a few days or hours only.

Following this first decrease in gonad volume, the oocyte number remained unchanged from June to July (Fig. 3). However, the next oocyte cohort of about 300,000 oocytes became mature and was shed by early August. After this second main spawning period, a third and final oocyte cohort became mature during the following 4 weeks. Because only one of the six female mussels studied in September still contained ripe oocytes (Figs. 2h, 3), it may be assumed that this last mature oocyte cohort was normally spawned by early September. Evidently, spawning events during this late part of the spawning season were not completely synchronized in mussels living close together. The histological sections of the empty gonads of most of the September specimens showed the breakdown of the follicle structure (Fig. 2g) and marked thereby the beginning of the next annual resting stage. While the first spawning period occurred in the temperature range of 12–15° C, the second spawning, after the summer peak in mature oocytes, corresponded with temperatures from 16–21° C.

Variations in the spawning season from year to year. (1) In 1986 and 1987, the spring peak in gonad volume was lower than in 1985 (Fig. 1). During a rapid increase of water temperatures in early spring 1986 (Fig. 1, top), only half of the mussels exhibited maturing oocytes before the end of April. The other half showed a slight, but significant reduction of the oocyte size (and no mature stages) compared with mussels from early April (Fig. 4). In 1987, the mature oocytes were observed at the end of May (Fig. 5). However, at this date the number of oocytes had already decreased distinctly since the end of April. Because temperatures rose above 12° C during mid May, it may be assumed that spawning events occurred just before the sampling date. This may also account for the less distinct peak in gonad volume recorded in that year.

(2) In 1986 and 1987, the summer peak of gonad volume in July appeared more distinct than in 1985 (Fig. 1). In both years, the number of oocytes might have increased slightly during June to July, although statistically not significantly (Figs. 4, 5). During this time (end of May–July) however, the mean oocyte diameter of the ripe oocyte cohort also increased significantly (1986 $P < 0.05$; 1987 $P < 0.01$; t-test).

A surprising observation deserves mention here. In 1986 in the Fühlinger See the first settling of young mussels was recorded at the end of June. Only 6–7 weeks later, at temperatures of about 20° C, these young mussels had already reached a shell length of about 7–8 mm and they contained ripe oocytes within their still small gonads, as indicated by histological slides. This demon-

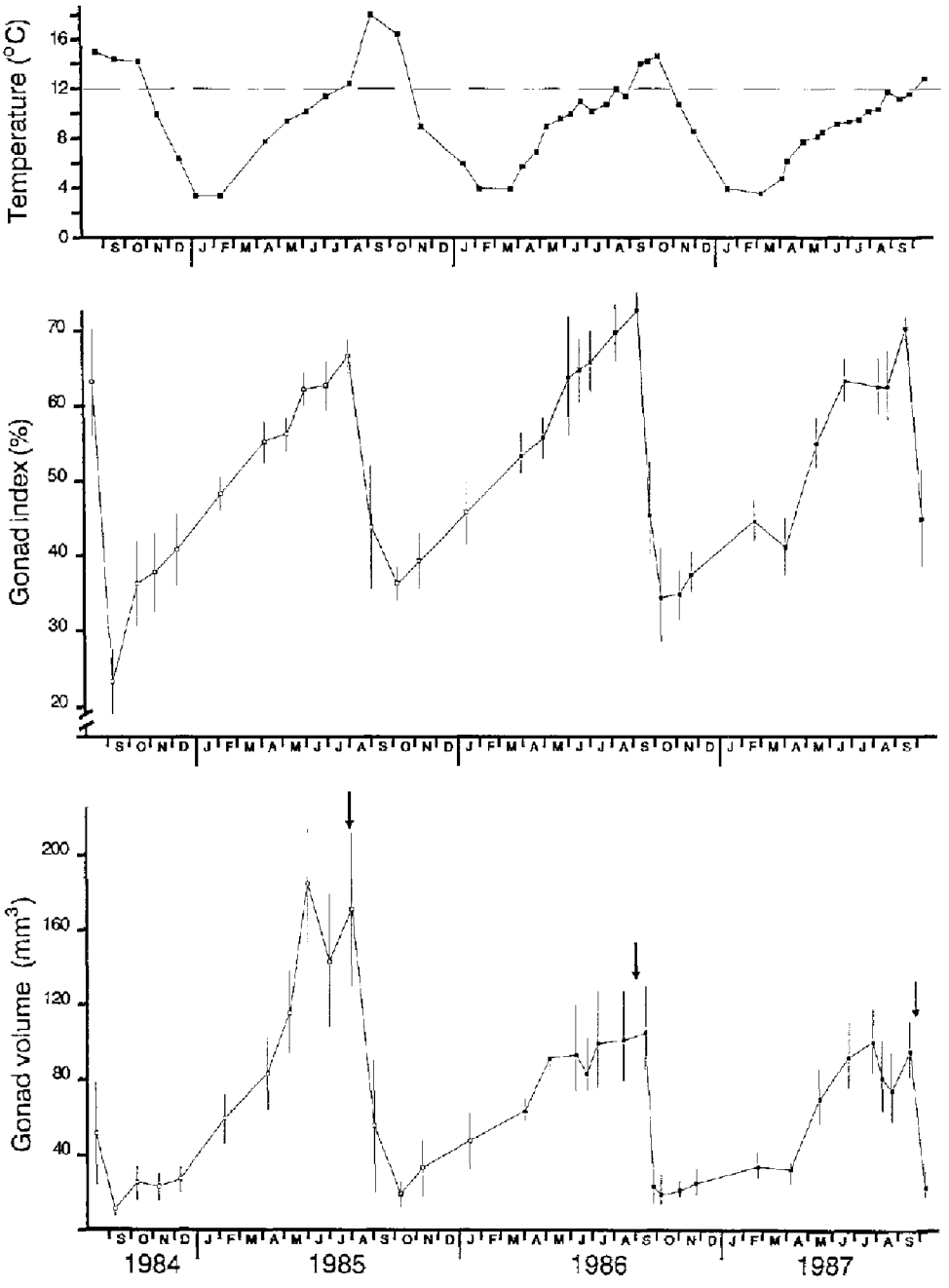

Fig. 6. *Dreissena polymorpha* (24 mm SL) at FS 9m. Seasonal course of gonad volume, gonad index (both with 95% confidence intervals) and temperature at 9 m depth. Symbols as in Fig. 1

strates that the gametogenesis can be stimulated in young mussels as small as about 6 mm, at water temperatures far above 10° C. Thus, the temperature range of the onset of gametogenesis and spawning is different in these young mussels and the older ones.

Fühlinger See 9 m depth (FS–9m)

The annual cycle of gonad development and spawning (Fig. 6) was even more precise in the mussels from the deeper part of the lake (FS–9m) than in the shallow-water mussels (FS–2m). The sampling point was below the summer temperature discontinuity layer, i.e. during summer the mussels lived in a colder environment than those from FS–2m (compare Figs. 1 and 6). If temperature influences both gonad development and the first release of the gametes, a time delay would be expected compared with FS–2m. This is obvious in the data.

In 1985 (Fig. 7), there was only slight variation in the number of oocytes between April and the beginning of August. The small intermediate minimum of July was within the accuracy of the estimation for oocyte numbers. Therefore, the increase of the gonad volume between April and June may be referred solely to the maturing of the first oocyte cohort. The mean oocyte diameter of this cohort increased significantly between May and August ($P < 0.01$). The first spawning period at this water depth was seen in August–September, i.e. at a time after the temperature rise above the 12° C level comparable to FS–2m (compare Fig. 6). Until the beginning of September, 4 of the 5 mussels analysed spawned about

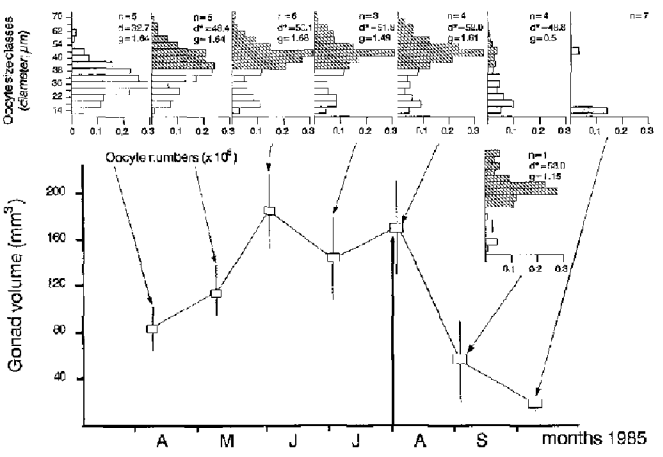

Fig. 7. *Dreissena polymorpha* (24 mm SL) at FS–9m in 1985. Gonad volume (1985 section of Fig. 6) and corresponding oocyte sizes plotted as frequency distributions. Symbols as in Fig. 3

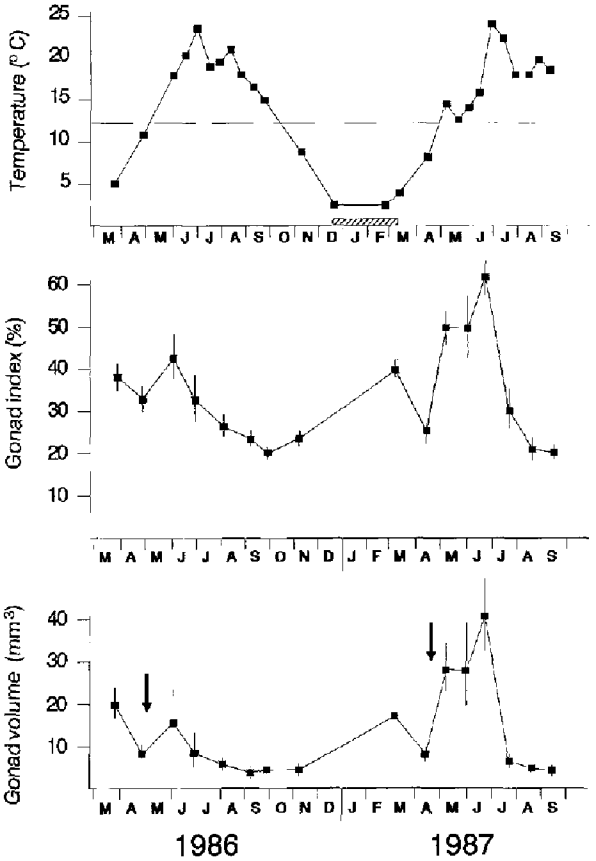

Fig. 8. *Dreissena polymorpha* (20 mm SL) at HS–2m. Seasonal course of gonad volume, gonad index (both with 95% confidence intervals) and temperatures at 2 m depth. *Arrows:* the time when the temperature rose above 12° C

1,100,000 oocytes each, while 1 spawned only about 500,000 oocytes (Fig. 7). The spawning period was over by early October, when only empty gonads with a breakdown of the follicle structure were found. These observations taken together suggest that spawning occurred in batches.

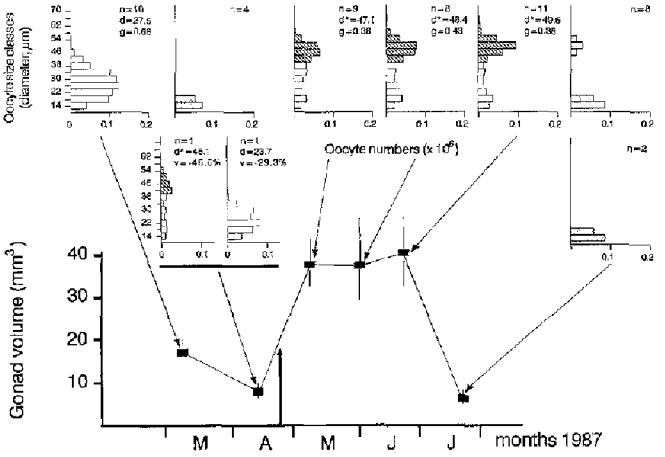

Fig. 9. *Dreissena polymorpha* (20 mm SL) at HS–2m in 1987. Gonad volume (1987 section of Fig. 6) and corresponding oocyte sizes plotted as frequency distributions. v in two of the April plots: change of the total number of oocytes compared with the preceding sampling date. Symbols as in Fig. 3

The temperatures during the spawning periods at FS–9m ranged between 12° and 16° C. By the histological examination of the November specimens in 1985, it was established that gametogenesis had already restarted in October without a clear resting stage, at temperatures about 10° C. However, in winter 1986/87 any further increase of the gonad volume and the development of small oocytes was retarded until March, in the same way as at FS–2m. This may indicate that there is at least one other factor acting besides temperature (e.g. food supply).

Heider Bergsee (HS–2m)

Temperature conditions at HS–2m and FS–2m were very similar (cf. Figs. 8 and 1). The main difference between the lakes was a lower food availability for the animals of HS–2m (Sprung 1989), especially during spring and summer. If food conditions as well as temperature influence strongly the course of the reproductive cycle, differences in gonad development of the mussels of HS–2m, compared with those of FS–2m, should be expected.

In both years studied, the gonad volume and the gonad index were surprisingly higher in March than in April. The size and the number of March oocytes were quite similar to those found at FS–2m. At both locations these oocytes were clearly immature at this time of the year. Thus, the decrease in gonad volume at Heider Bergsee in April certainly cannot be explained by an early spawning period at temperatures below 12° C. It was due to an unexpected reduction of both oocyte numbers and oocyte sizes of all females, as established by histological analysis (Fig. 9). For instance, the estimated values of v at Fig 9 (i.e. the change of the total numbers of oocytes in comparison to the preceding sample) mean that the theoretical calculated value of the total numbers of oocytes for the standard animal of 20 mm SL (g) was re-

duced from 0.68×10^6 (March) to $0.33–0.48 \times 10^6$ (Fig. 9). This reduction period, which was also found at HS–2m in 1986 (for GV and GI see Fig. 8; oocyte measurements not shown), may be comparable to the reduction period observed at FS–2m in April 1986 (Fig. 4). However, it seems to be more distinct here. After this reduction period, all HS animals were again well synchronized in their gonad development. The mean diameter of the remaining oocytes increased significantly ($P < 0.01$) during the following weeks, before these oocytes were spawned, at temperatures around 20° C, about mid-July. There was no evidence for a second or third cohort. Presumably the remaining small oocytes were resorbed. Consequently, the spawning season at HS–2m was already over at the beginning of August, and resting stage started much earlier than at FS–2m.

Discussion

The reproductive cycle

The reproductive cycles of various iteroparous molluscs may be either annual or independent of the seasons (see reviews by Giese 1959; Sastry 1979; Mackie 1984). The reproductive cycle of *Dreissena polymorpha* has always been attributed to the annual type (Antheunisse 1963; Galperina and Lvova-Katchanova 1972; Walz 1978). This was confirmed by this study. The high level of synchronization between the specimens is well demonstrated by the small confidence intervals of the gonad parameters for most sampling dates. However, there has previously been no adaequate method to quantify the size of the gonads (e.g. Galperina and Lvova-Katchanova 1972 – classification of gonad developmental stages; Walz 1978 – average oocyte sizes; Antheunisse 1963 – relative oocyte size-frequency distributions).

Walz (1978) demonstrated that *Dreissena* in Lake Constance can spawn several times per season. This has also been shown for other species, e.g. *Mercenaria mercenaria* (Davis and Chanley 1956; Ansell 1967). However, the temporal scheme of gonadal development giving rise to this pattern was not known. In theory, there are three possibilities:

1. After all oocytes of the spawning season have been generated and have developed synchronously to maturity, they are spawned in several portions.
2. Successive oocyte cohorts are generated, ripen and are spawned.
3. All oocytes are generated by the beginning of the spawning season, and then successive cohorts mature and are spawned.

The quantitative gonad analysis combines information on the gonad volume (e.g. Morvan and Ansell 1988), the gamete volume fraction (e.g. Bayne et al. 1978) and the oocyte size-frequency distributions (e.g. Brown 1984) to calculate the absolute numbers of oocytes of every size class in a standard mussel of a defined size at any time of the spawning season. The results of this study verified the third possibility. Although additional proliferation of new germ cells [i.e. the combination of (2) and (3)] cannot be totally excluded, there was no case where the number of oocytes increased significantly after the onset of the spawning season (e.g. Figs. 3, 7). Nevertheless, the overlapping phases of maturation and spawning can be explained by distinct oocyte cohorts which riped and were spawned gradually. This is the first time that such a phenomenon has been demonstrated for a species containing high numbers of oocytes.

Similar results were derived for *Modiolus modiolus* (Brown 1984) and *Tapes rhomboides* (Morvan and Ansell 1988). In both cases, the diagrams of the gonad index (based on gonad developmental stages – Brown 1984; based on gonad volume – Morvan and Ansell 1988) were similar to those shown for *Dreissena* in this study (e.g. Fig. 1). The application of the method used in this study may thus lead to comparable schemes of oocyte development for species with similar gonad size/index curves (e.g. *Mytilus edulis* – Gabbott and Bayne 1973; Pipe 1985; *Chlamys islandica* – Sundet and Lee 1984; *Argopecten [= Aequipecten] irradians* – Sastry 1966).

Temperature and spawning season

The timing and duration of the individual phases of the annual reproductive cycle of different species are highly variable (see reviews by Giese 1959; Sastry 1979; Mackie 1984). Generally, temporal synchronization and the duration of the phases are likely to be controlled by the interaction between endogenous and exogenous factors (Giese 1959). Where variations of the annual reproductive cycle have been found between different populations of one species, they have been attributed to small-scale local or large-scale geographical conditions (e.g. *Mytilus edulis* Seed 1975; *Modiolus modiolus* Brown 1984). Temperature and food supply have been identified as the main factors (Sastry 1979).

In this study, the onset of the spawning season for *Dreissena polymorpha* was always associated with temperatures of about 12° C (the delay of the onset of the spawning season at HS–2m will be discussed later). The existence of a temperature threshold has been accepted for many species (e.g. 10–12° C for *Mytilus edulis* and *Mya arenaria;* 15–16° C for *Ostrea edulis, Pecten irradians* and *Teredo navalis;* 20° C for *Crassostrea virginica;* 24–25° C for *Venus mercenaria* and *Mytilus recursus;* see review by Mackie 1984). Thorson (1950) pointed out that the temperatures of the spawning season were always within the lethal limits for the development of eggs and larvae. The results of Sprung (1987) give additional evidence for the suggested temperature threshold, because he demonstrated that temperatures above 10° C are required for the successful fertilization of *Dreissena* eggs. This threshold is also confirmed by the first appearance of larvae in the plankton (Sprung 1989). Nevertheless, temperature thresholds may show some variation, e.g. between physiological races (e.g. *Ostrea edulis*, Mackie 1984).

Food availability and spawning season

The significant reduction of the gonad size in animals at HS–2m in spring (temperatures still below 12° C, Fig. 8) cannot be correlated with an early spawning event between mid-March and mid-April. A similar phenomenon occurred in several specimens at FS–2m in April 1986, when the oocyte size classes were shifted to smaller stages. What were the reasons for this reduction? On the one hand, food availability in the lakes was at the low winter level, as indicated by estimated amounts ingested (Borcherding 1990). On the other hand, temperature rose relatively fast during these weeks (HS–2m 1986 – 0.17° C/day; HS–2m 1987 – 0.19° C/d; FS–2m 1986 – 0.19° C/d); for comparison only 0.10° C/d at FS–2m in 1985, and 0.13° C/d in 1987 (Figs. 1 and 8). While the energy input of *Dreissena* remained low at HS–2m in both years and at FS–2m in 1986, the respiration rate nearly tripled as the temperatures rose (Sprung in prep.). It is likely that the mussels had a negative energy balance during this time, probably forcing them to respire part of the highly-energetic gonad tissue to ensure vital physiological processes. Walz (1978) denied the possibility of gonad resorption in *Dreissena*, but quite recently these processes have been described in detail (Bielefeld 1991; Sprung and Borcherding 1991). However, these results suggest a mechanism whereby insufficient food supply can delay the first spawning event (possibly only in combination with increasing temperatures).

Energetics may also explain the variability in the length of the spawning season at the different locations. Without doubt, high numbers of oocytes need more energy than lower ones. There are two possible ways to produce high numbers of oocytes under moderate energy conditions. On the one hand, the spawning season might be prolonged by increasing the number of cohorts of maturing oocytes. This was observed at FS–2m – with up to four spawning periods per season – where in all three years oocyte numbers per animal were high and the spawning season was long compared with HS–2m (with only up to two spawning periods). Moreover, with higher summer temperatures the maturation periods of the successive oocyte cohorts may be shorter when food supply is ample, as it probably was at FS–2m in 1985. On the other hand, another pattern was seen in the colder, deeper part of the lake, where the maturation period was extremely prolonged, until late summer. Then, a higher proportion of the total oocytes matured and were released during the first spawning event (about 70% at FS–9m compared with 40% at FS–2m), resulting in lower oocyte numbers for following spawning events. Thus, a shorter spawning season in total might be expected.

Resting stage

The resting stage in the *Dreissena* populations studied started between August and the end of October, probably initiated by the end of the spawning season. This is known from many species (e.g. *Mytilus edulis*, Chipperfield 1953; *Ostrea edulis*, Loosanoff 1962) and has also been demonstrated for *Dreissena polymorpha* (Antheunisse 1963; Walz 1978). The timing of the end of the spawning season varied between populations, but normally the next gametogenesis started at the same time, so that the length of the resting stage also varied (e.g. compare Fig. 1 and 6), something also observed in other species (e.g. *Mytilus edulis*, Seed 1975; Sprung 1983).

Gametogenesis

The nature of the exogenous factor(s) controlling the onset of the gametogenesis is regarded as species-specific (Sastry 1979), temperature has usually identified as the main factor [e.g. decreasing temperatures in autumn for *Mytilus edulis*, Seed (1975); minimum temperatures during winter for *Astarte sulcata*, Saleuddin (1975); increasing temperatures in spring for *Crassostrea virginica*, Loosanoff (1965)]. To what extent the onset of gametogenesis of *Dreissena* depends on temperature is not clear from the present results. Gametogenesis started, as a rule, in late autumn as temperatures fell (below 8–10° C), but there was also the important exception of the young mussels of the Fühlinger See in 1986, where gametogenesis may have started at higher temperatures. How far this ability is restricted to these young mussels needs further clarification.

This study mainly demonstrated a distinct gonad growth in *Dreissena* during winter at temperatures about 2–4° C. These results contrast with those of Antheunisse (1963) for *Dreissena* from the river Amstel near Amsterdam and Walz (1978) for *Dreissena* from Lake Constance, who reported that gonad growth was inhibited during the low temperatures in winter. In both these studies, an interaction with low food supply is possible, which might also account for the retarded increase in gonad volume for *Dreissena* in the Fühlinger See in 1986/87. In neither of these studies, did the author know the size of the gonads, and thus their conclusions are not free from doubt. Nevertheless, some species showing inhibited gonad growth due to low temperatures (e.g. *Argopecten [= Aequipecten] irradians*, Sastry 1963). Generally the gonads grow during the low temperatures in winter, although sometimes more slowly (e.g. *Mytilus edulis*, Sunila 1981; *Chlamys islandica*, Sundet and Lee 1984). As described for many other bivalves (for review see Sastry 1979), the development of the gonads for *Dreissena polymorpha* accelerated with increasing temperatures in spring, leading to ripe oocytes within 2 months.

General conclusions

What conclusions may we draw concerning the further spread of *Dreissena polymorpha*? First of all, temperature seemed to be an important factor limiting the colonization of new waters. The onset of the spawning season – and consequently perhaps the timing of the whole reproductive cycle – depends on the temperature rise above a 12° C threshold. Thus, if temperatures remain above

12° C throughout the year, oocyte maturation and spawning may become desynchronized within a population. Thus, an important prerequisite for fertilization success in this reproduction type would be lost. Hence it is probable that *Dreissena* will not successfully colonize warm-monomictic lakes with minimum littoral winter temperatures above 12° C.

Spread to cold-monomictic lakes may also be limited by the 12° C threshold. According to the present results, no spawning would occur and, in addition, even the fertilization of the eggs would not be possible (Sprung 1987). These predictions agree with the actual European distribution: *Dreissena* is only found between 40° and 60° N (Odebrecht 1957; Breitig 1965; Stańczykowska 1977), where (referred to a height above sea-level of up to 800 m) neither of the above lake types are expected to occur (Wetzel 1983).

In addition, the amplitude of annual temperature variation may influence the reproductive success of *Dreissena*. Low amplitudes may result in desynchronization of gonad development and spawning, because small and long-lasting temperature changes might be not sufficient for temporal control (e.g. deep-sea species, Mackie 1984). On the other hand, high amplitudes might also be unfavorable if rates of respiration and ingestion are not balanced during periods of rapidly changing temperature (see above, and Bayne Newell 1983). This hypothesis might explain the absence of *Dreissena* populations from such biotopes as small shallow ponds, and could be tested there.

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