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# Duration of meiosis in barley

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SUMMARY

The duration of meiosis and its stages has been estimated in *Hordeum vulgare* variety Sultan grown at 20·0 °C with continuous illumination. The results are in good agreement with a previous estimate of the duration of meiosis and some of its stages in barley. The duration in barley does not differ greatly from that predicted from the previously calculated regression of DNA amount on meiotic duration in diploid plant species. As in other cereal species, meiosis is comparatively short.

## 1. INTRODUCTION

Lindgren, Eriksson & Ekberg (1969) attempted to estimate the relative duration of the stages of meiosis from pachytene onwards in pollen mother cells (*p.m.c.*'s) of Bonus barley by calculating the percentage of *p.m.c.*'s at various stages in anthers from different spikelet positions. They concluded that the total duration of the stages from pachytene to tetrads is not more than 3 days, and the stages of shortest duration last for less than 1 h. Bennett, Chapman & Riley (1971) have measured the duration of meiosis in diploid rye, hexaploid wheat and octoploid *Triticale* and shown that in these species the meiotic duration is short compared with other plant species such as *Lilium longiflorum*, *Trillium erectum* (Ito & Stern, 1967), and *Agapanthus umbellatus* (Lima-de-Faria, 1965). Bennett (1971) has shown that the duration of meiosis in diploid plant species is positively correlated with DNA amount per cell. In the present work the duration of meiosis has been estimated in barley. The significance of the work stems from the importance of barley as a crop plant and the consequent need to understand its pattern of development fully. In addition, the opportunity was provided to test the correlation between the content of DNA per nucleus and the duration of meiosis.

The DNA amount per cell in diploid barley, measured in root-tip squashes processed by the method of McLeish & Sunderland (1961), is $20{\cdot}3 \times 10^{-12}$ g and from the regression of DNA amount on meiotic duration in diploid species (Bennett, 1971), meiosis may be expected to last for about 48 h.

## 2. MATERIALS AND METHODS

The experiments were performed using *Hordeum vulgare* L. variety Sultan ($2n = 2x = 14$), a 2-rowed spring barley. Seeds were germinated on moist filter paper and potted up after 2 days. The plants were grown in a glasshouse without

artificial lighting until about 1 week before the leading tillers reached meiosis, at which time they were transferred to a growth chamber maintained at 20·0 °C ± 1 °C with continuous illumination.

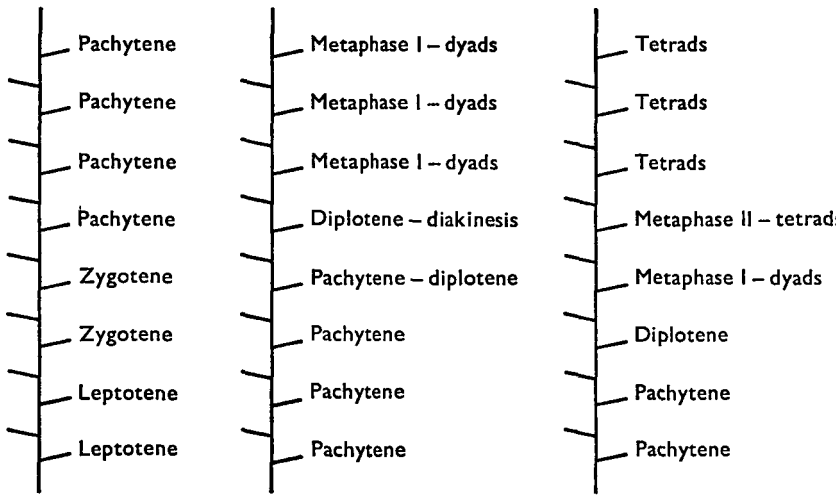


Fig. 1. The meiotic stages in sampled florets of three typical spikes at sampling time.

Barley spikes and anthers are too small for the application of methods which involve sampling anthers or parts of anthers within a floret. Consequently, the duration of meiosis was measured on spikes *in situ* by a method which depends on the ordered sequence of spikelet development along each spike and on the near synchrony of anther development in each floret (Bennett *et al.* 1971). Each spike was stripped of its enclosing leaf sheaths and at a recorded time ('sampling time') 4–8 spikelets were removed from about the middle of one side of the spike and instantly fixed in 1 : 3 acetic alcohol. A small polythene bag moistened inside with distilled water was then fastened over the spike and the plant was immediately returned to the growth chamber. Anthers from the fixed florets were Feulgen stained and their meiotic stages were determined in squash preparations. After a known period of time (4–22 h), the whole spike was fixed at a second recorded time ('fixing time'). All operations were done carefully and swiftly to minimize damage to, or desiccation of, the spikes. Fixed anthers from florets whose positions were intermediate between those of the florets already sampled were dissected out from the florets on the unsampled side of the spike. The anthers were then stained by the Feulgen procedure and squashed. The experimental treatment did not affect the appearance of the meiotic stages.

Fig. 1 shows typical patterns of meiotic development for florets along the sampled side in three spikes. They resemble the corresponding patterns in field grown Foma and Bonus barleys reported by Ekberg & Eriksson (1965). At sampling time, several adjacent spikelets along the sampled side of a spike were often at the same stage of meiosis and this allowed accurate determination of the meiotic stages

at sampling time in the unsampled intermediate spikelets on the opposite side. Hence, the development completed by these latter spikelets in the period between sampling and fixed times could be measured.

The central anther in a floret often lagged slightly behind the others in development (cf. Ekberg & Eriksson, 1965) and the tips of some anthers were 2 h or sometimes more ahead of the bases in meiotic development. Consequently, in calculating the duration of meiosis we considered only the two most advanced anthers in each floret and reckoned the developmental progress made between sampling and fixing times as spanning either both youngest or both oldest stages in each of the florets compared at sampling and fixing times. This minimized the error due to asynchrony within florets and anthers. This error means that our estimates of the durations of individual short stages of meiosis are approximate. This, however, does not apply to our timing of the longer stages of first prophase, the total duration of meiosis or the tetrad stage.

## 3. RESULTS

A summary of results is given in Table 1 and Fig. 2 shows a representative sample of the most informative results.

Table 1. *The durations of meiosis and its stages and the tetrad stage in Sultan barley*

| Stage | Duration (h) | Stage | Duration (h) |
|---|---|---|---|
| Leptotene–telophase II inclusive | 39·4 | Anaphase I | 0·5 |
| | | Telophase I | 0·5 |
| Leptotene | 12·0 | Dyad stage | 2.0 |
| Zygotene | 9·0 | Metaphase II | 1·2 |
| Pachytene | 8·8 | Anaphase II | 0·5 |
| Diplotene | 2·2 | Telophase II | 0·5 |
| Diakinesis | 0·6 | Tetrad stage | 8·0 |
| Metaphase I | 1·6 | | |

From the patterns of distribution of meiotic stages along spikes and the durations of the stages, one can calculate that the developmental interval between successive florets along one side of the sampled region of the spike is not more than 3 h and is less in the most advanced part just above the middle. The corresponding figure for wheat and rye is about 6 h or less (M. D. Bennett, unpublished).

While the *p.m.c.*'s were at mid-pachytene, a synchronous division of all the tapetal cells occurred and each cell became binucleate. Often, the whole tapetum was in division at the same time, but a gradient of mitotic stages along the anther was seen. Thus an anther tip might be at telophase while the middle was at anaphase and the base at an even earlier stage. A similar pattern of development along the length of the anther occurred among the *p.m.c.*'s.

Each tetrad persisted for about 8 h and then broke up to give four pollen grains.

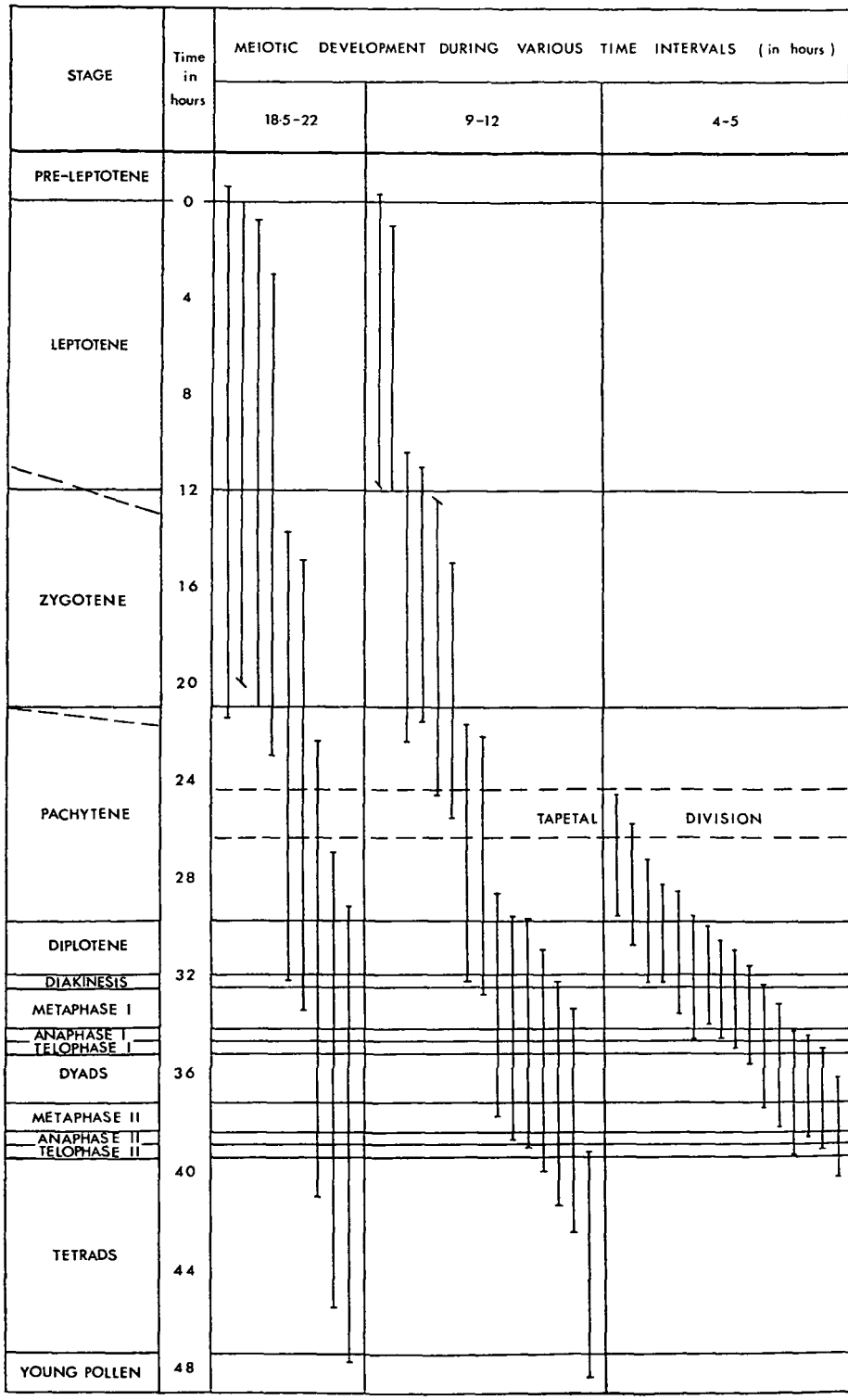


Fig. 2. Meiotic development completed during various time periods by Sultan anthers at 20 °C.

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## 4. DISCUSSION

The present results show that Lindgren *et al.* (1969) were correct in concluding that the meiotic stages from pachytene to tetrads in barley occupy 3 days or less and that the shortest stages last less than 1 h. In fact the duration of meiosis is only slightly more than $1\frac{1}{2}$ days and the duration of the shortest stage is half an hour or less. A comparison of the present results with estimates of the relative durations of the stages from diplotene to telophase II obtained by Lindgren *et al.* is given in Table 2 and shows that there is a good measure of agreement between them.

Table 2. *The relative lengths of the meiotic stages from diplotene to telophase II in the present work and taken from Lindgren* et al. *(1969)*

| | % of total duration of diplotene to telophase II, inclusive | |
|---|---|---|
| Meiotic stage | Present work | Lindgren *et al.* (1969) |
| Diplotene | 22·9 | 26·6 |
| Diakinesis | 6·3 | 8·7 |
| Metaphase I | 16·7 | 19·1 |
| Anaphase I plus telophase I | 10·4 | 8·4 |
| Dyad stage | 20·8 | 16·1 |
| Metaphase II | 12·5 | 9·0 |
| Anaphase II plus telophase II | 10·4 | 12·1 |

The observed duration of meiosis in diploid barley, 39·4 h, is close to that expected from the regression of mean DNA content per cell on meiotic duration calculated by Bennett (1971). The meiotic duration obtained is the duration expected for a diploid species having a mean DNA content per cell of 18·2 picograms (pg) while Sultan has 20·3 pg. The deviation of expected from observed DNA content is $2{\cdot}1 \pm 12{\cdot}0$ pg and is not significant. This result increases confidence in the previously calculated relationship between DNA amount and meiotic time.

Bennett (1971) has shown that at 20 °C, the various stages of meiosis occupy similar proportions of the duration of the entire division despite large interspecific differences in the duration of meiosis. At 19 °C in barley, the proportions of the overall length of meiosis occupied by the constituent stages are compatible with this observation, although zygotene and pachtene are a little longer than might have been expected. The departures from the expected duration of these two stages are not excessive and while the stages of meiosis probably have similar relative durations in most higher plants, some variation is to be expected. Since the error in the estimates of the duration of zygotene and pachytene is not known, we cannot say whether this departure from expectation is real.

The synchronous division of tapetal cells in Sultan occurred at mid-pachytene in the present instance but at diakinesis in some field grown material. It seems, therefore, that in barley there is no absolute correlation between the timing of this division and the development stage of the *p.m.c.*'s. A further study of the relation-

ship between these divisions may help our understanding of the influence of environmental factors on the rate of development of anther tissues. Sultan barley differs from other cereal species grown in the same environment in that the division which gives rise to binucleate tapetal cells occurs during mid-pachytene in barley but at the start of leptotene in wheat and the end of leptotene in rye (Bennett *et al.* 1971).

A knowledge of the timing of events immediately preceding meiosis, during meiosis and in microsporogenesis is of obvious practical and fundamental interest. Clearly such information is relevant to seed production and may be useful in planning the field crossing programmes that will be necessary for the production of $F_1$ hybrid varieties.

Detailed knowledge of the duration of individual meiotic stages in a single environment in one organism permits the precise experimental treatment of those stages by irradiation, temperature shock or chemical means. Knowledge of the duration of meiosis in a wide range of organisms has already led to increased understanding of the mechanism of meiosis and of the factors which determine its duration (Bennett, 1971).

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