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Observations on plant development (VIII). The development of *Rumex obtusifolium*

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In natural conditions *Rumex obtusifolium* flowers from June till August. Observations were carried out on plants sown at various dates, namely: Experiment I — on July 13, 1959, Experiment II — on August 25, 1959, Experiment III — on March 1, 1960, Experiment IV — on September 6, 1961, Experiment V — on April 15, 1962.

Experiments I and II. The plants grew throughout the experiment in a glasshouse under continuous daylight (L_{24}) and 8-hour day (S_8); after 15 months some plants were transferred from L_{24} to S_8 and vice versa. Slight differences in the habitus were observed between the S_8 and L_{24} combinations. The leaves of the S_8 combinations were smaller and darker, with undulated blades and short petioles. The seasonal variability of the growth activity was hardly perceptible.

Experiment III. The plants were sown in a vegetation house. A part of the plants (controls) were transferred to the glasshouse on September 1 (age — 166 days). The other plants remained out-of-doors and were successively transferred to the glasshouse in the course of winter (age of plants first transferred — 221 days). A part of the plants bloomed (Table 1). The plants which remained vegetative did not bloom to the end of 1962 (observations interrupted).

Experiment IV. After sprouting a part of the plants was transferred to the glasshouse under L_{24} and S_8, whereas the others remained out-of-doors and were successively transferred to the glasshouse during winter. None of the plants bloomed, thus it may be supposed that they underwent a juvenile inactive phase.

On the other hand, the vernalized plants exhibited a markedly more intensive vegetative development than the controls. The differentiation and growth of leaves was rapid and the blades were visibly larger. The quantity of leaves was counted on the control plants every week from November 11 to May 12.

The total number of leaves was in combination L_{24} — 6.3 and in S_8 — 6.6 on November 11, whereas on May 12, 6.9 and 7.7, respectively. The dying and differentiation of new leaves in L_{24} were more or less uniform in the course of the entire period (increment +5.2, decrement −4.6).

Table 1

Date of observation	Control	Date of bringing into glasshouse														
		25.X	15.XI		2.XII		22.XII		9.I		26.I		18.II		3.III	
		b	a*	b**	a	b	a	b	a	b	a	b	a	b	a	b
10.I	0	0	56	1	39	2										
4.II	0	0	81	2	64	4	44	4	26	4						
2.III	0	0					70	5	52	5	41	2				
12.IV	0	0									82	4		0		0
n = 5 Total up to June 2	0	0		2		4		5		5		4		0		0

*a - No. of days after bringing into glasshouse.

**b - No. of flowering plants.

In S_8 the total mean increment was +5.2 and decrement −4.1. A certain tendency to a decrease of the number of leaves (more intensive drying and stagnation of differentiation) was observed in December, whereas a more rapid differentiation occurred in February.

Experiment V. Combinations: differentiation of light conditions from May 11 (2 leaves): varying natural daylight (N), 16-hrs long day = 8 hrs of natural daylight + 8 hrs, in a growth chamber and in winter 16 hrs. in a growth chamber (L_{16}), short natural daylight in winter in growth chamber (S_8).

On October 30, a part of the plants was subjected to natural vernalization for the following periods: (I) till November 30, (31 days), (II) till January 4 (66 days), (III) till February 9 (102 days), (IV) the entire winter out-of-doors.

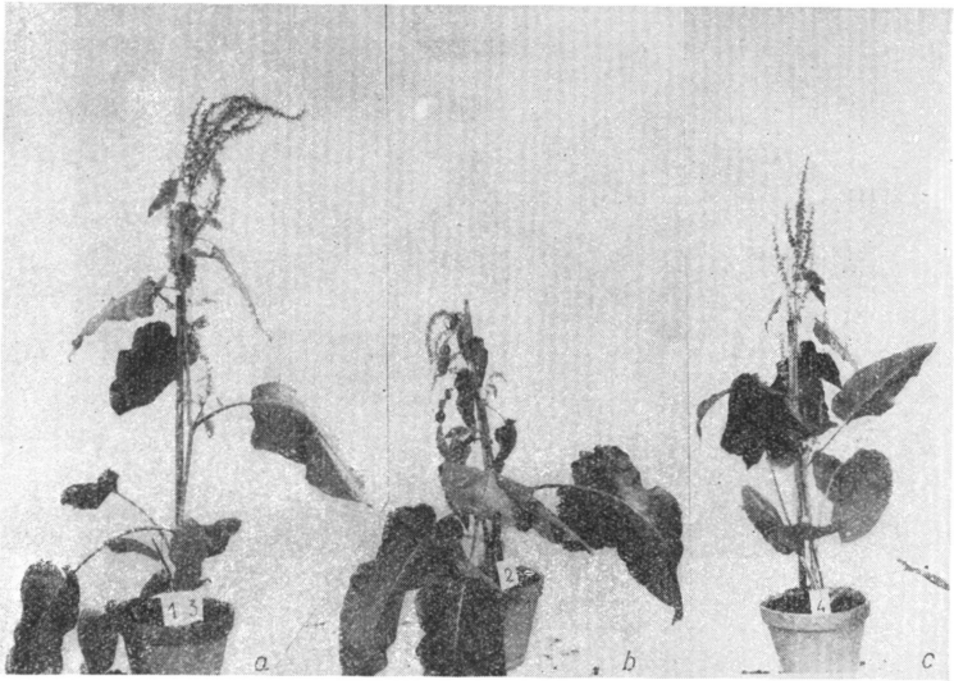
After vernalization the plants grew under L_{16} and S_8, thus the following combinations were obtained for each period of vernalization: N/vern./L_{16}, L_{16}/vern./L_{16}, S/vern./L_{16} and N/vern./S_8, L_{16}/vern./S_8, S_8/vern./S_8. In combination IV, in which the plants grew throughout the winter out-of-doors, they remained under natural varying daylight.

For vernalization the plants were placed under cucumber frames; none of the plants died, the main apex was undamaged and even the youngest leaves survived.

After transferring to the glasshouse the plants immediately began to develop.

Vegetative development.

In the entire course of the experiment observations were performed on the differentiation and dying of consecutive leaves (numbered). The differences in habitus and leaf size were also determined.



Fot. M. Spóz

Fig. 1. Series V. 22.I.63. Vernalization I. *a* — comb. $N/v/L_{16}$; *b* — $S_8/v/L_{16}$; *c* — $L_{16}/v/L_{16}$.

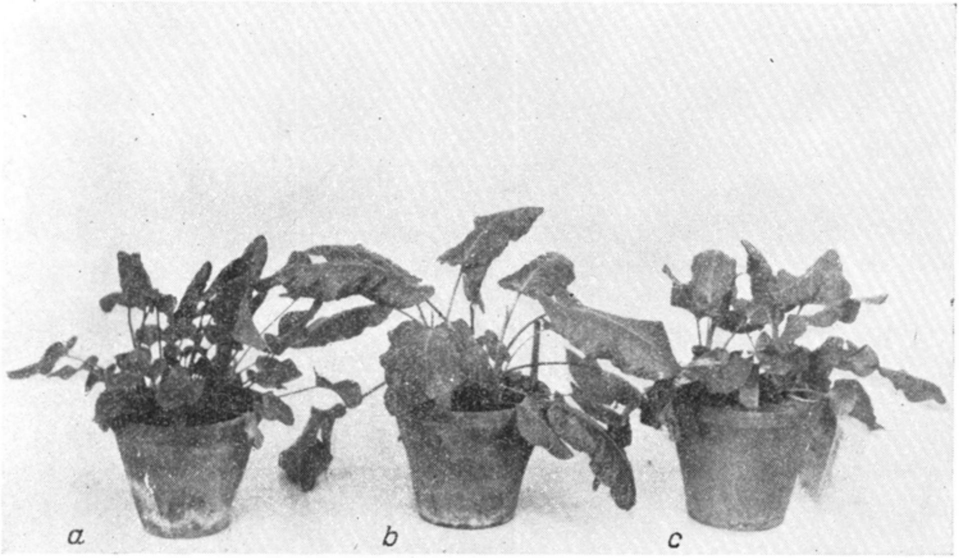


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Fig. 2. Series V. 8.II.1963. Vernalization I. *a* — $N/v/L_{16}$ (13); *b* — $L_{16}/v/L_{16}$ (2); *c* — $S_8/v/L_{16}$ (4).

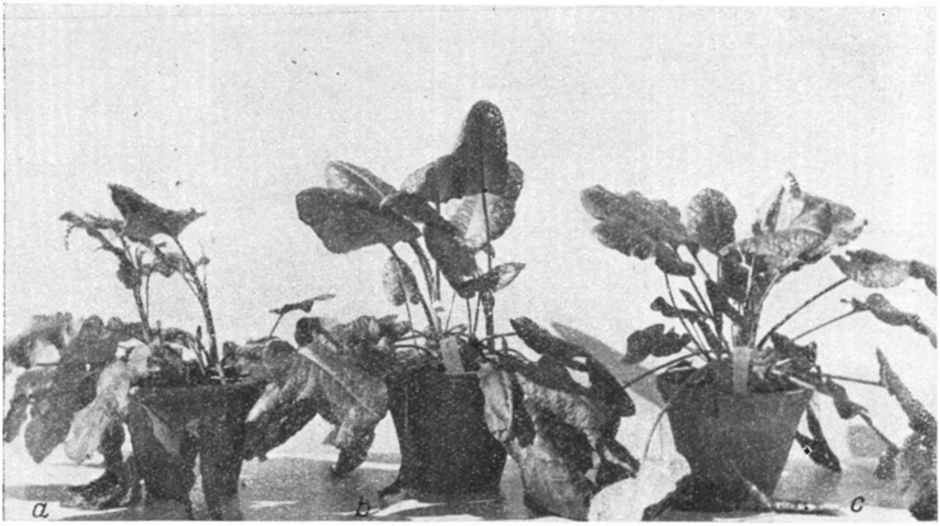

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Fig. 3. Series V. 22.I.63. Control plants. *a* — N; *b* — S_8; *c* — L_{16}.

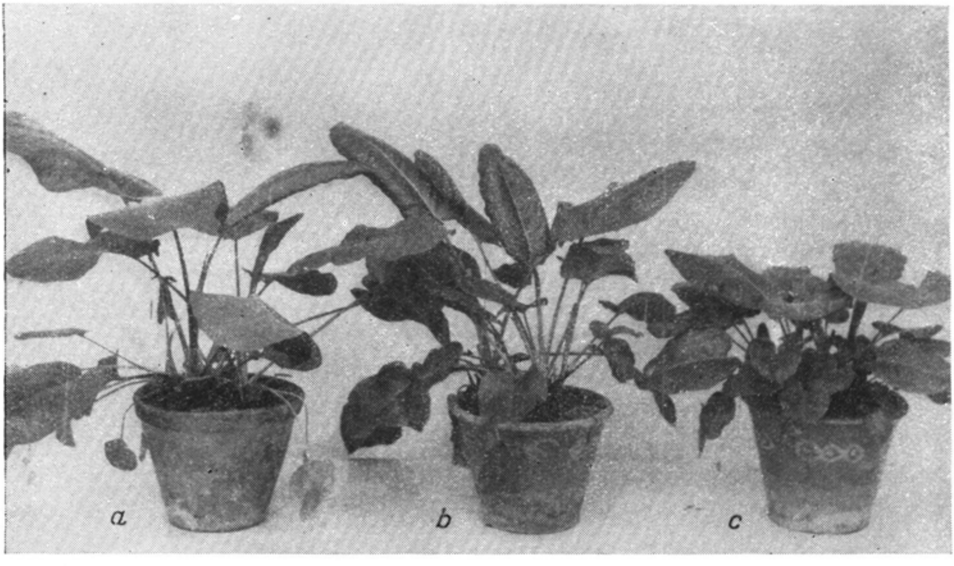

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Fig. 4. Series V. 10.VII.63. Control plants. *a* — N; *b* — S_8; *c* — L_{16}.

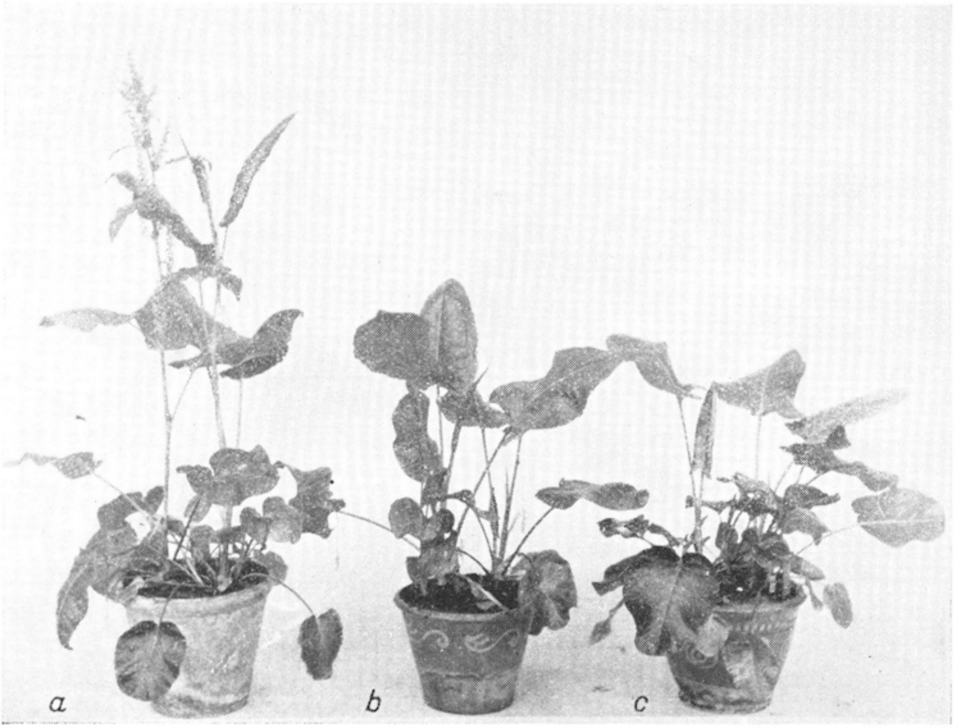

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Fig. 5. Series V. 22.I.63. Vernalization I. *a* — N/v/S_8; *b* — S_8/v/S_8; *c* — L_{16}/v/S_8.

1. Seasonal variation of growth activity was generally weak, relatively most marked in N combination.

This variation was manifested in the first place by an increased intensity of drying of "summer" leaves in autumn.

2. The total amount of leaves was invariably lower in N plants than in L_{16} and S_8. On the other hand differences in the number of leaves and in the rate of their differentiation occurring between non-vernalized L_{16} and S_8 plants were slight.

3. The comparison of the number of green leaves as well as their total amount and rapidity of their differentiation proved that its rate increases after vernalization. This acceleration is most intensive in plants shortly vernalized (vernalization I). The total number of green leaves of plants of "vernalization I" equalled as early as in June the control plants if, before vernalization, they grew under L_{16} and S_8.

In all experiments, the lowest increment of leaves occurred in combinations in which the plants grew under varying daylight (N) before vernalization.

Table 2

Combination	Beginning max. florescence				No. of plants		
	date of flowering	age of plants	no. of days after vernalization	during (days)	total	flowering	%
Controls	(not vernalized)						
N	7.II-29.VII	283-455	-	172	14	8	60
L_{16}	-	-	-	-	10	0	0
S_8	-	-	-	-	10	0	0
I vernalization - (30.X - 30.XI) 31 days							
$N/v/L_{16}$	18.I-15.III	263-329	49-105	66	10	9	90
$S_8/v/L_{16}$	24.I-15.III	269-329	55-105	60	10	6	60
$L_{16}/v/L_{16}$	8.II-15.III	284-329	70-105	45	10	2	20
$N/v/S_8$	13.II-25.IV	289-370	75-146	81	10	2	20
$S_8/v/S_8$	5.VIII	472	248	-	10	1	10
$L_{16}/v/S_8$	-	-	-	-	10	0	0
II vernalization (30.X - 4.I) 66 days							
$N/v/L_{16}$	3.V-23.V	378-398	119-139	20	10	2	20
$S_8/v/L_{16}$	-	-	-	-	10	0	0
$L_{16}/v/L_{16}$	-	-	-	-	10	0	0
	29.III transferred to vegetation house (n=6)						
N/v/N	18.IV-27.V	363-402	104-143	39	6	2	33
$S_8/v/N$	18.IV	363	104	-	6	1	17
$L_{16}/v/N$	-	-	-	-	6	0	0
$N/v/S_8$	-	-	-	-	10	0	0
$S_8/v/S_8$	-	-	-	-	10	0	0
$L_{16}/v/S_8$	-	-	-	-	10	0	0
III vernalization (30.X - 9.II) 102 days the same combinations - none of the plants flowered							
IV vernalization (all winter out-of-doors- then under N)							
N/v/N	22.VI-15.VII	423-445	-	22	8	8	100
$S_8/v/N$	12.VI-5.VIII	412-466	-	54	11	10	90
$L_{16}/v/N$	30.VI-5.VIII	430-466	-	36	10	7	70

4. The plants differed by habitus as well as by leaf sizes and petiole length (Fig. 1).

The leaves of the N plants were smallest, stiff, with undulated edges, whereas those of S_8 plants were largest and smooth, and those of L_{16} plants — intermediate.

The shorter vernalization period (vernalization I) markedly intensified the growth of leaves.

Thus the leaves may be ranged in the following order according to leaf size: N$<$L$_{16}$$<L_{16}$ vern.$<$S$_8$$<S_8$ vernal. Longer vernalization exerts a weaker influence and the leaves in vernalization III were even smaller than those of the controls. The leaves of control plants (particularly in L$_{16}$) decreased in size with the plants' age if it remained in the vegetative phase. An analogous decrease of leaf size was observed also on plants of vernalization II, and, particularly, of III which also did not flower. On the other hand, the plants left for the winter out-of-doors bloomed and their leaves were markedly larger.

Generative development.

The observations on the generative development are summarized in Table 2. The columns "Beginning → max florescence" give the date and the age of the first plant coming into flower as well as the date and age of the plants in the given combination at the moment of blooming of the maximum number of plants.

The experiment was interrupted on September 1.

It should be added that the habitus of the flowering plants exhibited some differences (Fig. 2).

In L$_{16}$/vern./L$_{16}$ plants as well as in those which grew under S$_8$ after vernalization I, the flower stems were shorter and the inflorescence consisted of a smaller number of flowers, analogously as in N plants. The phenomenon of flower stem shortening and decrease of the number of flowers was most pronounced in the blooming plants of vernalization II.

DISCUSSION

Both lower temperatures and daylength before and after vernalization exercise an influence on the generative development.

1. Considering the number of flowering plants and the abundance of florescence, the maximum reaction was observed in N/vern. IV/L, next came N/vern. I/L and S/vern. IV/L, i.e. under conditions in which either the plants grew under L → S or S and then were left out-of-doors throughout the winter and started to grow in spring under varying gradually rising temperatures, or at first grew under L → S, then were vernalized in late autumn, transferred in the beginning of winter to the glasshouse, and grew at higher temperatures under long day.

2. As regards the influence of daylength after vernalization, *Rumex obtusifolium*, behaves like a long-day plant. Short day has an inhibitory effect on it.

Only few plants growing under short day in interaction with vernalization, and under varying daylight L → S before vernalization, came into flower.

3. None of the non-vernalized plants growing under continuous

daylight or constant day-to-night proportion (L_{16} and S_8) passed to the generative phase or elongated into shoots.

This inhibitory influence of constant day-to-night proportion on generative processes occurred also in interaction with vernalization.

4. Under natural daylight (N) varying in length from L to S to L, a part of the plants of experiment V bloomed, though the individual time of florescence varied widely, and the character of florescence, i.e. the shorter stalk and small number of flowers in the inflorescence pointed to "non-optimum" conditions for the generative process. In experiment III the plants did not flower under varying daylight. The different behaviour of plants in both experiments may be attributed to the different light conditions in the winter and spring of 1961 and 1963. Characteristic for the winter and early spring of 1963 were many sunny days, thus much light. The experiments in 1961 were interrupted in the beginning of June, so, maybe, some plants would have bloomed, if the observations had been continued. At any rate, under the daylight varying from L to S to L, at least a part of the plants came into flower.

Thus it may be assumed that, from the standpoint of accumulation of development activators, these conditions lay on the border line of "suboptimum" and "inhibitory", with deviations in both directions depending on light intensity.

5. A marked influence of the light conditions before vernalization on its effectiveness was observed.

In vernalization I the long daylight before vernalization exercised an inhibitory action, whereas the constant short daylight decreased the effectiveness of vernalization.

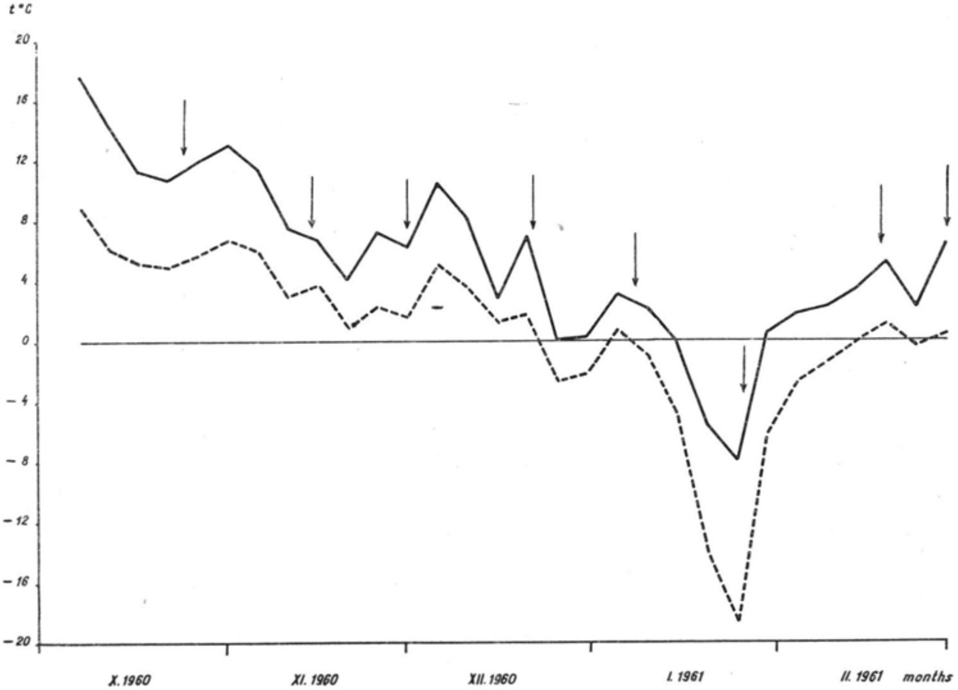
In vernalization IV in which the plants were left throughout the winter out-of-doors, this influence was weaker though it was also visible particularly in the L/vern./L combination.

6. In respect to the influence of vernalization:

a) the results of the experiment of 1961/1962 (Expt. IV) point to the existence of a juvenile nonactive phase in respect to the vernalization stimulus,

b) in the experiment of 1960 (Expt. III) the known phenomenon of dependence of the vernalization effectiveness on a certain "minimum" of time of low temperature action was observed,

c) in both experiments of 1960/1961 (Expt. III) and 1962/1963 (Expt. V) the plants were transferred to the glasshouse at various dates, and in both cases the authors observed the same fact of "effectiveness" of vernalization at certain dates. The plants either did not bloom or they flowered in a very low percentage and moreover, as it has been mentioned in the description of the influence of vernalization on the vegetative development, the certain influence of the "chilling" process, intensifying leaf growth and differentiation, did not occur.



Graph. 1. Exp. III. Mean air temperature for five day periods "minimum and maximum". The arrows indicate the consecutive dates of plant transfer from chilling into the hothouse.

The differences in the "inactive" dates in both experiments may be attributed to thermal differences of both years, nevertheless the fact of the occurrence of such "blocking" dates is interesting.

An active, i.e. effective as regards vernalization, influence was observed when:

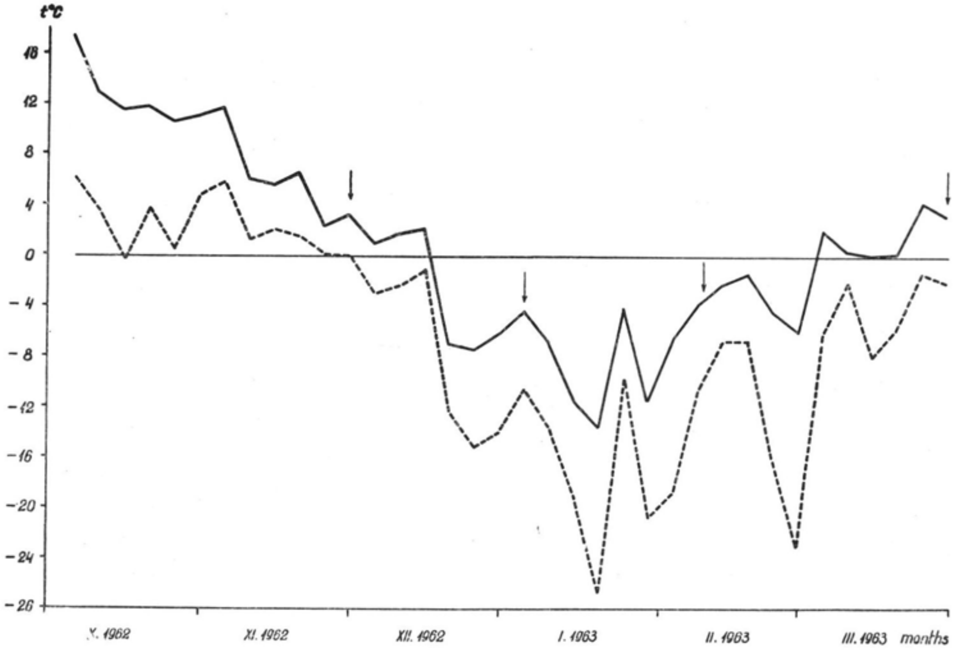
a) either the plants were left in field conditions throughout the winter and in spring were exposed to gradually increasing and less varying temperatures,

b) or the vernalization period lasted relatively a shorter time, i.e. during autumn or autumn and the first half of winter, and then the plants were transferred to higher temperatures in the glasshouse.

The intermediate dates were inactive.

The different reaction of *R. obtusifolium* to stable and varying light conditions as well as the interaction between florescence and vernalization, in dependence on the daylength before vernalization, are analogous to the observations on other plants, though few in number.

Recently, plants have been described which bloom only if they grow under daylight varying from L to S (*Castrum nocturnum* and several other species — Lockhard 1961). There exist particularly interesting, though hitherto scarse, species blooming e.g. under 16-hr daylight, and



Graph. 2. Exp. V. Mean air temperature for five day periods "minimum and maximum". The arrows indicate the consecutive dates of plant transfer from chilling into the hothouse.

not coming into flower under short and continuous daylight (Mathon and Stroun 1961).

On the other hand, the positive influence of S preceeding L was also observed (e.g. in clover — Thomas 1962, Ryńska 1962). *Campanula medium* (Wellensiek 1960) bloomed without vernalization under S→L conditions, whereas under S only in interaction with vernalization. The accelerating influence of S preceeding L was also reported for wheat (McKiney and Sando, cit. after Lockhardt 1961) and rye (Purvis 1961). In his investigations on the influence of daylight on the development of *Dactylis glomerata,* Calder (1963) recently discovered that the plant does not bloom under continuous daylight without vernalization, whereas it flowers under daylight varying from S to L. The naturally varying daylight is particularly effective.

Short day preceeding vernalization influences its effectiveness in many grass species (Napp-Zinn 1961 and literature quoted there). A similarly favorable influence was observed under conditions of lower light intensity preceeding the higher intensity after vernalization (Napp-Zinn 1960 in *Arabidopsis*). Maximum florescence in kohlrabi was observed in the case when the plants grew under long and then short daylight of lower intensity before vernalization. This trend of variation corresponds with the natural variability of light conditions under which biennal plants grow (Junges 1958).

This phenomenon has a different aspect when the problem of "active" and "inactive" dates of vernalization is taken into account.

A series of experiments, in which the influence of the time of vernalization was compared, indicated that its effectiveness rises with the increase of time of vernalization, however, within certain limits (Hänsel 1953). Very long vernalization of rye "if it exceeds three or four months seems to become less effective" (Purvis — cit. after Chouard 1960). In Junges' (1959) experiments with biennal vegetables the optimum duration of vernalization was 8—10 weeks, whereas longer periods diminished the percentage of blooming plants.

In the above mentioned experiments of Wellensiek (1960) *Campanula medium* bloomed under S only after very long lasting vernalization. Chouard (1960) reports a very interesting observation, in *Geum*, in which the main apex transforms to the flowering stem, and this perennial plant "becomes" monocarpic only when subjected to 30—50 weeks vernalization.

All these data give no grounds for the explanation of the phenomenon observed in *Rumex obtusifolium*.

Since the plants with shorter terms and earlier dates of vernalization bloomed, it would seem that the process of their vernalization must have been terminated. Therefore, the facts observed cannot be interpreted as resulting from devernalization after transfer of the plants to the glasshouse. This may only occur when the "vernalizing" process is at the stage of synthesis of intermediate labile development activators, and may be reversed.

Temperatures below a certain minimum are known to be ineffective as regards vernalization. If, however the process of vernalization starts at a temperature above zero, then, in various wheat varieties, a possibility of completion of the vernalization at temperatures below zero has been demonstrated (Zarubajło and Kiśliuk 1951). The same authors observed an inhibitory effect of considerably lower temperatures (—8°). These observations concerned, however, "an uncompleted vernalization process", therefore phenomena other than those observed in the present study.

Analysis of the course of temperatures during vernalization in Expt. III indicates that, in the period up to January 26, i.e. almost to the end of the period of lowest temperatures, it gave a positive effect, whereas the later vernalization periods during a distinct rise of temperature were "inactive".

Consequently one might suppose that the main apex froze in the interval between the "active" and "inactive" periods. Owing to this, the plants transferred to the glasshouse later dates would have started to grow from differentiating axillary apexes which developed later and could not be vernalized. This could not have been the case, since the plants transferred to the glasshouse started to grow immediately by differentiation of the main appex.

The following fact should be moreover taken into account: a part of the plants of vernalization II (ineffective) were transferred on March 29 to a cool vegetative house, and some of them bloomed.

Finally, some differences occurred between the plants of vernalization I and IV. The quantity of plants which bloomed in the latter was higher, notwithstanding the daylength before vernalization, and the florescence was markedly more intensive.

Thus the phenomenon observed requires further investigations. At present we may only assume, as working hypothesis, that "certain" inhibitors blocking the vernalization effect are produced under the influence of certain temperature conditions with prolonged vernalization and subsequent abrupt transfer to the glasshouse, i.e. to higher temperatures (18—22° C). The reduction of these inhibitors would occur only under conditions of gradual temperature changes occurring in nature in early spring and spring.

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Obserwacje nad rozwojem roślin
VIII. Rozwój Rumex obtusifolium

Streszczenie

Obserwacje prowadzono na kilku seriach w ciągu paru lat porównując rozwój roślin na różnej długości dnia oraz w interakcji z jaryzacją.

1. Rośliny bez jaryzacji — zakwitły w pewnym procencie jedynie na dniu naturalnym, o kierunku zmienności L → S → L, natomiast nie zakwitały na dniu o stałym stosunku dnia do nocy zarówno długim (16 godz.), jak i krótkim (8 godz.).

2. Maksimum procesów kwitnienia przebiega na dniu o zmiennej długości, o kierunku L → S → L, w interakcji z jaryzacją.

3. Dzień długi lub ciągły przed jaryzacją — hamuje lub zmniejsza efektywność jaryzacji, z tym, że to hamujące oddziaływanie ulega znacznemu osłabieniu po długim okresie jaryzacji.

4. W obu latach doświadczenia zaobserwowano zahamowanie kwitnienia przy wniesieniu roślin do szklarni w różnych okresach w ciągu zimy, mimo ukończonej poprzednio jaryzacji.

W pracy dyskutowane są różne możliwości tłumaczenia reakcji roślin, zarówno gdy chodzi o zakwitanie na dniu zmiennym bez jaryzacji, jak i o „blokujący" wpływ zimowych terminów jaryzacyjnych.