

Botany. — *Results of the temperature during flower-formation for early Hyacinths. (l'Innocence and la Victoire.)* By H. F. WATERSCHOOT. (Communication N^o. 26 of the Laboratory for Plant-physiological Research, Wageningen, Holland.) (Communicated by Prof. A. H. BLAAUW.)

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§ 1. *Introduction, material and method.*

By BLAAUW the influence has been examined of various temperatures during flower-formation on the whole hyacinth. For this purpose the *late* variety Queen of the Blues was taken (BLAAUW, 1924).

Next however it was important to know how other varieties would behave. For this purpose we now examined the varieties l'Innocence and la Victoire *flowering early* in culture. In this case the bulbs were exposed only to those five temperatures for which most optima were found in Queen of the Blues, while we took care that sufficient room was left for possible deviating optima in these early hyacinths. Besides the results of these temperatures were only traced for the most important organs. This examination however is more extensive in so far, that the bulbs to be planted were not only exposed to an after-treatment in 13° C. after a preliminary treatment of 8 weeks in 23°, 25.5° and 28° C., but after a preliminary treatment of 3,5 and ca 8 weeks in 17°, 20°, 23°, 25.5° and 28° C.

To get an insight in the foundation and object of this experiment, for drawings, more detailed description and for comparison, see BLAAUW 1924.

At the beginning of July 1926 there were destined for this examination 210 + 640 bulbs of l'Innocence and 640 bulbs of la Victoire. Both varieties were so-called biennial of scooped material (= 3 year old) and selected for a circumference of 12—13 cms. The Queen of the Blues previously examined was a year younger and of a circumference of 7.5—9 cms. This should be specially borne in mind in judging flower-cluster-formation and number of flowers.

Of the 210 bulbs l'Innocence 10 bulbs were fixed on July 5, 1926 in alcohol 96 %, in order to trace the condition of the vegetation-point on the day on which the experiment was started. 200 bulbs were put in the temperatures 17°, 20°, 23°, 25.5° and 28°, viz. 40 in each temperature. Of these 40 bulbs groups of 10 bulbs were left in these temperatures for 3, 5, ca 8 and 12 weeks and next fixed, in order to trace in which

temperature after a definite period the development had progressed farthest.

The 640 bulbs of l'Innocence and la Victoire were treated as has been explained with respect to the above 200 l'Innocence, but they were not fixed: after 3,5 and ca 8 weeks they were transferred from the above temperatures to 13° and 17° and planted after 12 weeks simultaneously with the groups of 20 which had been kept in 17° to 28° throughout the 12 weeks. For each of the 32 different combinations produced in this way 20 bulbs were taken. Since however the 20 bulbs l'Innocence which after 8 weeks should have gone from 17° to 13°, were kept in 17° by mistake, this combination is left out; accordingly in the subjoined tables we find two groups with the treatment 17° for 12 weeks mentioned for l'Innocence.

§ 2. Results of the temperature-treatment for the leaves.

Number of foliage-leaves formed.

1. *At the beginning of the experiment.* The number of young foliage-leaflets on July 5, 1926 was 54 for the 10 bulbs examined. Of these 10 bulbs 7 had 5 leaflets, 2 bulbs 6 leaflets (in one of these bulbs one leaflet was not entirely split off) and 1 bulb 7 leaflets (one of which was not entirely split off).

2. *After the fixations.* After 12 weeks 200 bulbs had been treated in 20 various modes (see tab. 1) and fixed. The number of leaves in these 200 bulbs averagely amounted to 54.7 per 10, when those which are not entirely split off are also counted. Therefore some increase is again to be noted, just as in Queen of the Blues, but also with respect to l'Innocence we mean to be justified in drawing the conclusion that this slight *increase is of an accidental nature* and not due to the exposures to these temperatures.

TABLE 1. *l'Innocence.*

Number of young foliage-leaflets per 10 bulbs, when they have been in 17°—28° C., from 3 to 12 weeks.

	After 3 weeks	5 weeks	7½ weeks	12 weeks
17°	55	56	53	54
20°	53	53	54	54
23°	54	56	53	56
25.5°	54	53	57	58
28°	54	55	56	56

Is not this sufficiently clear from the fact that 54 is found in 25.5° after 3 weeks, 53 after 5 weeks, after 7½ weeks 57 and after 12 weeks 58. This

would have to signify an increase, but this is impossible, for after 5 weeks the leaf-formation has long been finished and the flower-cluster-formation is in full swing.

Number of shooting leaves in spring.

In § 1 it has been said, that 640 bulbs of l'Innocence and of la Victoire were planted out after the various exposures. The tables 2 and 2a give their number of leaves, which in spring, when they had been pushed from the bulbs, could be counted. The recorded numbers have been counted on groups of 20 bulbs and converted per 10.

From these tables it appears, that (just as in Queen of the Blues) not all leaves grow out in lower temperatures.

TABLE 2. *l'Innocence*.
Number of assimilating leaves in the spring of 1927 per 10 bulbs.

Preliminary exposure	3 weeks		5 weeks		7½ weeks		12 weeks
	17°	34.0	—	35.5	—	—	—
20°	39.5	48.0	47.5	52.5	51.1	50.0	54.0
23°	51.0	52.5	53.2	53.7	53.5	54.5	54.5
25.5°	46.5	54.2	53.0	54.5	55.5	53.5	53.0
28°	52.0	55.0	54.0	56.5	54.5	56.5	55.5
After-exposure to:	13°	17°	13°	17°	13°	17°	—

TABLE 2a. *la Victoire*, as table 2.

Preliminary exposure	3 weeks		5 weeks		7½ weeks		12 weeks
	17°	56.5	—	61.6	—	61.5	—
20°	59.5	60.0	61.5	62.6	61.0	62.0	62.5
23°	61.5	59.0	62.0	59.5	61.1	60.5	60.5
25.5°	60.5	61.0	60.0	60.5	61.5	62.3	62.0
28°	62.0	62.0	61.5	61.5	62.5	62.5	64.0
After-exposure to:	13°	17°	13°	17°	13°	17°	—

In order to make all leaves grow out 13° is nearly always less favourable than 17°. Further it may be observed that the higher the temperature of

the preliminary exposure, the shorter it need last to make all the leaves shoot. In l'Innocence 20° is necessary for 12 weeks, 25.5° and 28° but for 3 weeks. l'Innocence however can have a lower temperature than Queen of the Blues. All or nearly all the leaves already grow out in l'Innocence in 20° for 12 weeks, whereas in Queen of the Blues this does not occur before 25.5° for 8 weeks.

La Victoire (table 2a) deviates still more from Queen of the Blues; here there are no great differences to be indicated in the chosen temperatures. The complete number 64, which probably averagely amounts to 61 or 62, is here (with 64) already attained in 17° for at most 12 weeks, whereas the number 56.5 for the lowest temperature, viz. 3 weeks in 17° and next in 13°, compared with the two other varieties, deviates but little from the complete number. So in this respect la Victoire can stand 13° much better than l'Innocence.

Length of the foliage-leaves.

1. Table 3 gives the length of the young outer foliage-leaf measured at the peeled bulbs of the *fixed material*. The optima are printed fat. For Queen of the Blues and l'Innocence they are found in nearly the same treatment. The principal differences are, that for l'Innocence an optimum

TABLE 3. *l'Innocence*.

Average lengths in mms of the outer foliage-leaves, still in embryonic condition ($n=10$).

	Beginning July 5, '26	After 3 weeks	5 weeks	7½ weeks	12 weeks
17°	3.69	4.72	6.45	10.30	20.83
20°	3.69	5.24	6.86	9.79	18.97
23°	3.69	5.23	7.02	10.82	18.12
25.5°	3.69	5.47	6.51	9.47	15.51
28°	3.69	5.19	6.31	9.09	13.30

lies at ± 8 weeks 23°, whilst for Queen of the Blues this lies at ± 8 weeks 20° and further that for Queen of the Blues the leaves in 3 weeks 23°, 25.5° and 28° are equally long. Accordingly there is a striking correspondence. Here it is again corroborated, that according as the organ-enlargement (especially in September) is acting a more important part, as compared with the organ-formation, the optimum is shifted to a lower temperature.

2. Of the *planted* bulbs the tables 4 and 4a give the average length of the outer leaf above the ground at the point of time, that the leaves of the greater part of the bulbs already showed above ground.

The measuring was rather inaccurate, especially on account of the rough surface of the soil, so that only the great differences are of value.

TABLE 4. *l'Innocence*.

Average lengths of the outer foliage-leaves above ground in mms per bulb after removal of the cover (peat-litter) on January 28, 1927 (× = under the surface or on the same level).

Preliminary exposure	3 weeks		5 weeks		7½ weeks		12 weeks
	17°	15.5	—	17.3	—	—	—
20°	16.0	×	13.3	1.3	4.3	6.0	2.8
23°	27.8	20.8	26.0	7.3	10.5	4.0	2.0
25.5°	59.0	28.8	25.5	21.3	6.0	4.5	2.3
28°	36.3	25.0	31.0	26.8	22.5	23.5	6.0
After-exposure to:	13°	17°	13°	17°	13°	17°	—

TABLE 4a. *la Victoire*, as table 4.

Preliminary exposure	3 weeks		5 weeks		7½ weeks		12 weeks
	17°	4.0	—	3.8	—	1.5	—
20°	10.3	2.8	12.8	×	2.3	×	×
23°	31.3	1.3	13.0	1.8	6.3	×	×
25.5°	23.8	1.5	19.0	0.8	6.5	5.3	×
28°	31.8	2.8	19.3	5.3	7.8	5.0	×
After-exposure to:	13°	17°	13°	17°	13°	17°	—

From the tables 4 and 4a this conclusion may be drawn: for the stretching of the leaves up to this time the after-treatment in 13° is better than in 17°. Further it may be stated, that *l'Innocence* 3 weeks 25.5° and next 13° surpasses the other treatments favourably. Here too it appears, that the optimum which is found after 12 weeks 17° with the leaflets in embryonic condition (table 3) is shifted when reckoned over a longer period (during the stretching).

For *la Victoire* (table 4a) we find a corresponding optimum in January in 3 weeks 23°, 25.5° and 28° with after-treatment in 13°.

Number of sheath-leaves in *l'Innocence*.

On July 5 each of the 10 bulbs examined had 2 sheath-leaves. In the bulbs examined after exposure (fixed material) there occurred in each

TABLE 5. *l'Innocence*.

Number of sheath-leaves per 10 bulbs. (The number at the beginning of the experiment was 20 per 10 bulbs.)

	After 3 weeks	5 weeks	7 $\frac{1}{2}$ weeks	12 weeks
17°	19	17	19	16
20°	17	19	19	18
23°	19	19	19	16
25.5°	17	19	16	18
28°	19	17	17	17

group of 10 at least 1 bulb with one sheath-leaf (table 5). Of the 200 bulbs 155 bulbs had two sheath-leaves, 44 bulbs one sheath-leaf and 1 bulb three sheath-leaves (this latter in 3 weeks 17°). That a certain exposure can influence this figure cannot be concluded from this.

§ 3. *The direct effect of 17°—28° during 3 to 12 weeks on the development of the floral whorls in l'Innocence.*

About the 210 fixed bulbs the results as to number of foliage-leaves and sheath-leaves and length of the outmost foliage-leaf have already been communicated in § 2. In the present § the other observations on these bulbs are mentioned, viz. in what stage of development the vegetation-point that is to yield the flower-cluster for the following spring, was found in consequence of the 20 temperature-exposures.

In order to enable us to represent the progress of the formation of the floral whorls BLAAUW (1920 and 1924) adopted 10 stages of development. Stage I is still a simple vegetation-point, in stage X this has grown into a cluster, the lowest flowers of which are quite complete. In the variety *l'Innocence* the stages can in the main be distinguished in the same way, as in the previously examined *Queen of the Blues*. What differences there are, will be discussed afterwards.

The stages of development in question have been drawn from nature and lithographed by Mr. VAN TONGEREN. The objects, stained with a strong aqueous solution of iodine and iodide of potassium were examined (also during the drawing) through a binocular microscope. The magnification has been given with the illustrations (45—50 \times). What has been drawn, was partly selected from the material of this research, partly from other *l'Innocence*-material of equal age. In determining the stages the lowest flowers were examined as these are the first to develop. The number of flowers, their size and shape do not act a part in this. Neither

does the origin and development of the new vegetation-point, that (normally) will pass on to flower-formation the next summer.

In the plates denotes : VP, vegetation-point ; L, foliage-leaf ; LL, scar foliage-leaf ; L*, rudimentary foliage-leaf ; NVP, new vegetation-point ; BR, bract ; BLP, flower-primordium ; NPH 1, first leaf of the new phyllome-series ; S, bractlet ; T I and T II, tepals of the outer and the inner whorl ; M I and M II stamens of the outer and the inner whorl ; VD, carpel. When of the NVP and of the first sheath-leaf of the NVP the swelling is not yet visible (not any or extremely little external differentiation), but the spot where these organs will arise, is already visible (of the NVP through strong staining by the aqueous solution of iodine and iodide of potassium), this was designated (NVP) and (NPH 1).

The subsequent stages in l'Innocence are :

- I. The vegetation-point VP is still splitting off leaves (L) and is still low (fig. 1).
- II. The vegetation-point has finished splitting off leaves and is raised, while no differentiation is visible but a weak indication (NVP) or a slight swelling NVP of the new vegetation-point (figs. 2, 3 and 4).
- III. Besides the new vegetation-point which gets clearer now some crescentshaped prominences BR are visible (figs. 5 and 6).
- IV. In the lower primordia we now see the bract (as a roundish prominence) and the primordium of the proper flower BLP (likewise as a roundish prominence) separated from each other by a furrow (figs. 7 and 8).
- V. The three outer tepals T I are to be distinguished as three independent primordia (fig. 9).
- VI. The three inner tepals T II id. (figs. 10 and 11).
- VII. The three outer stamens M I id.
- VIII. The three inner stamens M II id. (fig. 12).
- IX. The three carpels VD id. (fig. 13).
- X. The three carpels are raised, while the margins of each carpel are turned in (fig. 14).

Besides the above description the plates and stages need the following explanation of a more secondary nature :

Figures 1 and 2 have been drawn in the same magnification and also in the same position, so that the height of the two vegetation-points (fig. 1 and fig. 2) are comparable in the drawings. Figs. 3 and 4 give the growing-point seen from above.

Figure 4 has been drawn from the same object as fig. 3, but turned about 180 degrees.

In fig. 3 the last fully formed foliage-leaf has been slightly lifted to show how this leaf covers the vegetation-point. This innermost foliage-leaf

namely frequently presses upon the growing-point or the developing flower-cluster so heavily, that a distinct ridge or dent is visible on it. This therefore has no actual signification with respect to the formation of organs; this should be borne in mind, because this may give rise to errors in judging the object. In figures 2, 3, 4, 5 and 10 the dent thus originated has been marked with a \times .

The leaflet last formed (L^* in figures 2 and 5) has often not even been finished and is arrested as a rudimentary organ in the shape of a scale. This rudimentary leaflet may be followed as far as stage X and further, but it is no more visible in the illustrations after stage III.

In stage IV— (which approaches stage IV very closely) we see (fig. 7) the first indication of a sheath-leaf in the new vegetation-point; in the figure (NPH 1). In the following fully drawn clusters the further development may be traced.

In stage IV (fig. 8) in the lowest flowers we see the first differentiation of a bractlet, indicated with S, the further development of which may likewise be traced in some flowers in figures 9 and 10. See on this bractlet: BLAAUW 1920, Summary § 3.

In stage VI the entire flower-cluster has been given (fig. 10), but the first (lowest) flower, indicated BLP 1 of this same flower-cluster has been opened and drawn in the right hand top corner of the plate (fig. 11), in order to indicate the feature of stage VI, i.e. the differentiated condition of the inner tepals. Of the following stages but one of the flowers has been drawn after being opened. This latter is necessary, because after stage V the tepals begin to overlap the further inward parts of the flower. This opening sometimes causes the turned down tepals to be torn loose at the inside of the base, which is shown by the scars in the drawings.

The differences with Queen of the Blues with regard to the stages.

Ad stage II. In Queen of the Blues stage II was considered the condition in which the vegetation-point is raised, whilst no further differentiation is visible. In l'Innocence it was observed, that the new vegetation-point may already be visible in the axil of the last leaflet (by cell-divisions at that place or by some swelling) fully split off (binocular; staining with iodine and iodide of potassium), when the vegetation-point has finished splitting off leaves and before there is any indication of a bract.

Characteristic of stage II is only the rise of the vegetation-point, not the appearance of the new vegetation-point, because also in judging the later stages we have left this out of account.

Ad stages III and IV. In Queen of the Blues stage III has been called the condition, in which the 1st or the 1st and 2nd flower-primordia are visible as weak swellings. Besides the new growing-point at the base is not mentioned before this.

In l'Innocence we first observed a crescent-shaped roundish prominence of the bract (= stage III), while the part above this which is to give rise to the proper flower-primordium does not show any prominence. Not until after this there appears above the bract a prominence of the proper flower. At first this is lower than the bract (= III+ or IV—), but it will soon be higher (= IV). Further the bract lags considerably behind in growth.

In limiting the stages we can also mention as a criterion for stage III the rise of a single crescent-shaped prominence. Then it is left undecided for the present, whether this prominence is the bract or a primordium which will yet be differentiated into a bract and a flower.

Ad stage VII. For Queen of the Blues (and also for l'Innocence) stage VII is reached, when the three outer stamens are visible as independent organs. But by that time in the examined material of l'Innocence there was always something to be seen of the inner whorl; for that reason the stage is called VII+ or VIII—. Now it is possible, that not all the stages and transitions from one stage to another could be observed in this material, the number of different treatments being limited here. Yet the impression remains that the stages VII and VIII, when they do not fairly coincide, succeed very quickly, i.e. in this way, that the inner whorl commences forming, when the outer is not yet finished.

The progress of the flower-cluster-formation.

This has been synoptically represented in a "curve of dots" (fig. 15). (See BLAAUW 1924 and LUYTEN, JOUSTRA, BLAAUW 1925). Each dot indicates a bulb and is placed with the stage of development (ordinate), in which the vegetation-point or the lowest flower were, while the treatment is indicated on the abscis. A +stage was put in the upper and a —stage in the lower half of a square. In the advanced stages IX the anthers could be measured under the binocular; their length was ca 0.2 mm. As smallest measure in stage X ca. 2.5 mms. was found. A sub-division of stage X or rather the development after stage IX has been rendered according to the length of the anthers. It is necessary therefore to take into account, that on the ordinate above and below the heavy line values have been plotted which were obtained in different ways.

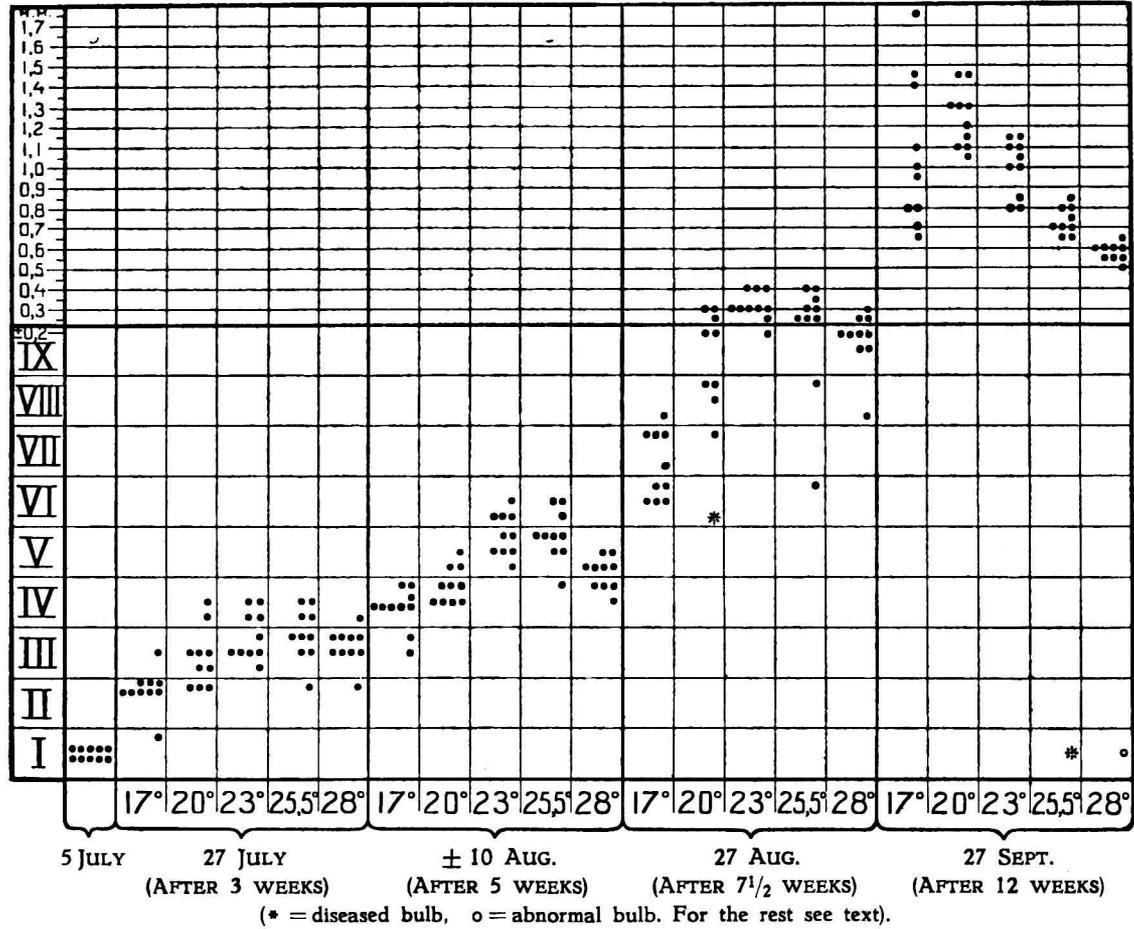
Here a discussion follows of the Fig. 15 in question :

1. *On July 5* at the beginning of the experiment all of the 10 examined bulbs were in stage I. It was however the end of the period of splitting off leaves as appears from tables 1 and 2.

2. *After 3 weeks* 25.5° has progressed farthest, though 23° and 28° are falling behind but little.

3. *After 5 weeks* the flower-cluster is further developed than in whatever temperature after 3 weeks. 23° and 25.5° are farthest. It was

Fig. 15. The progressing flower-cluster-formation.



remarkable in this case, that in 25.5° in two bulbs the new vegetation-point at the foot of the cluster had already passed on to flower-formation. In both bulbs this had already attained stage IV.

4. *After 7½ weeks* 23° and 25.5° are farthest and besides, especially in 23° very uniformly developed. The development of the 10 bulbs in 20° is particularly unequal. One of the bulbs in 20° was diseased, but reached stage VI after all (indicated with * in fig. 15).

5. *After 12 weeks* 17° is farthest, but (just as in Queen of the Blues) most unequal. In this case anthers were measured of a length of 0.65 to 1.75 mms. Of a failure of the flower-cluster, as in Queen of the Blues, there is no question in l'Innocence in 17°. But the Queen of the Blues examined at the time was a year younger with a circumference of 7.5—9 cms and accordingly already therefore possessing slighter flowering-capacities. Averagely most advanced is 20°. Most uniform in development are the bulbs in 28° (anthers of a length of 0.5 to 0.65 mm) and in 25.5° (from 0.65 to 0.85 mm.); 25.5° is the farthest of these. In 25.5° there was a diseased bulb, which had not progressed at all in its development. In 28° there was a similar bulb likewise entirely lagging behind in development, which was abnormal in a different way. Here too in 25.5° the new vegetation-point in one of the bulbs had proceeded to flower-formation (stage V+). This was not the bulb that had not developed at all.

§ 4. *The total effect on the flower-cluster in l'Innocence and la Victoire.*

In order to trace the total effect of the various exposures the bulbs were planted out after 12 weeks (on Sept. 27, 1926) in a cistern with fixed groundwaterlevel of 60 cms. (described in BLAAUW 1922). The total effect on number and length of the leaves has already been rendered in § 2 (together with the direct effect); in this § the total effect on the flower-clusters follows. See the tables 6 and 6a. At the top (between the double and the heavy horizontal lines) the exposure has been given. Next the number of planted bulbs follows. 20 bulbs were taken per treatment, but in some cases, where the figure 19 is found, one has dropped out. Next there is added to the dates the number of flower-clusters, which started flowering on these dates. Then the number of flowering and non-flowering bulbs and the average number of flowers per cluster; at the foot of these the mean error of this average. The signification of the * we find at the close of this §. Mind, that in table 6 two groups are found with an exposure of 12 weeks 17°.

Beginning of flowering.

On March 17, 1927 the first flowers of la Victoire and on March 19 those of l'Innocence came into bloom. The number of clusters which came

TABLE 6. Flowering of *IInnocence* (see § 4).

For.....	3 weeks					5 weeks					7½ weeks					12 weeks																
in	17°	20°		23°		25.5°		28°		17°	20°		23°		25.5°		28°		17°	20°	23°	25.5°	28°									
next in.....	13°	13°	17°	13°	17°	13°	17°	13°	17°	13°	17°	13°	17°	13°	17°	13°	17°	13°	17°	—	—	—	—	—								
Number of planted bulbs	20	20	20	20	20	19	20	20	19	20	20	20	20	20	20	20	20	20	20	20	20	20	20	20								
Beginning of flowering on:	19 Mrch				1	1			7 ₍₈₎	7 ₍₁₁₎	6	3			1																	
	21 ..		1	13	8	10	9	10	16	3	9	12	9	13	19	14	15			2	10	3	9	5	16	6						
	23 ..	5	5	4	6	10	2	8	8	3	3	5	3		10		1		2	10	8	8	16	10	15	3	11	1				
	25 ..	2	1	12		2		1			3	3	10		1				1	4	6	8	1			1	3	4	9	6		
	27 ..			2															6	1	2		1					9	7	11	5	
	29 ..			1								6							5		2							3	4	2	12	10
	31 ..																			3										1	1	4
	2 April																													1		2
4 ..			1																													2
Number of flowering bulbs (= n)	7	7	20	19	20	13	19	18	19	9	17	19	19	20	20	20	19	20	20	19	20	19	20	20	20	20	18	20	20	20	20	20
Number of non-flowering bulbs	13	13		1		7		2	1	10	3	1											1								2	
Number of flowers per cluster (= M)	9.3*	8.6*		9.9*	10.7*	8.2*		10.6*	11.4*	10.6*	11.6*	11.2*		10.0*	10.4*	11.1*	11.3	10.9*										9.5*	10.1*	10.9*	11.2*	11.1*
		10.1		9.7(*)	9.1	10.5				10.4*	10.8*	11.5	9.7		10.0*	11.0	11.9	10.1														
m = ±	0.9	0.9	0.3	0.5	0.5	0.4	0.5	0.4	0.4	0.6	0.4	0.4	0.5	0.5	0.4	0.4	0.2	0.3	0.5	0.6	0.5	0.4	0.3	0.3	0.6	0.5	0.5	0.4	0.4	0.3	0.4	0.4

TABLE 6a. Flowering of *la Victoire* (see § 4).

For.....	3 weeks					5 weeks					7½ weeks					12 weeks					
in	17°	20°	23°	25.5°	28°	17°	20°	23°	25.5°	28°	17°	20°	23°	25.5°	28°	17°	20°	23°	25.5°	28°	
next in.....	13°	13° 17°	13° 17°	13° 17°	13° 17°	13°	13° 17°	13° 17°	13° 17°	13° 17°	13°	13° 17°	13° 17°	13° 17°	13° 17°	—	—	—	—	—	
Number of planted bulbs	20	19 20	20 19	20 20	20 20	19	19 19	20 20	20 19	20 20	20	20 20	19 19	20 20	20 20	20	20 19	20 20	20 20	20 20	
Beginning of flowering on:	17 Mrch		2	3				1	6	1					1						
	19 ..	3	7	5	2		3	13	14	14 6			11	3	6						
	21 ..	3 5	8 5	9 7	11 5	4 11		6 12	15	5 11	3 13		5 2	17 16	12 13						
	23 ..	1 7 2	2 6	3 10	5 11	3		6	4	2	9 5 4		3 11	4	1 5						
	25 ..	1 2 7	5	3	2	6 4 7		2		1	5 2 11		6		2						
	27 ..	3 1 4	3		2 2	5 1 11					2	5							3 2		
	29 ..	6	7				1												13 16 10 12 16		
	31 ..	1																	4 2 6 6 4		
2 April																			2 1		
4 ..																			1 1		
Number of flowering bulbs (= n)	15	18 20	19 19	20 20	20 20	18	19 19	20 20	20 19	20 20	19	20 20	19 19	20 20	20 20	20	20 19	20 20	20 20	20 20	
Number of non-flowering bulbs	5	1	1			1					1										
Number of flowers per cluster = M	14.9*	19.3*	19.4*	18.3*	13.9*	21.3*	23.4*	22.3*	20.5*	19.5	24.9(*)	27.0	21.8	20.0	19.4	23.9	25.4	24.1	20.8	19.5	
		24.6	23.8	20.4	19.3		24.4	23.7	20.3	18.1		24.2	22.1	22.0	17.8						
m = ±	1.5	1.1 0.9	1.1 1.2	0.5 1.1	0.8 0.5	1.5	2.1 0.7	0.8 1.0	0.6 0.6	1.4 0.7	0.7	1.3 0.9	0.7 0.7	0.6 0.9	0.6 0.5	1.1	1.8	1.1	0.7	0.9	

into bloom on these days (and further every other day) have been given in the tables.

l'Innocence flowers earliest in an exposure of 5 weeks to 25.5°, 23° and 28° with an after-treatment to 13°. In 23° and 25.5° the figures 8 and 11 are placed between brackets. We doubted namely in these cases whether we should note 7 or 8 clusters for each in 23° and 11 clusters in 25.5°, but at last we decided on 7 for each. Yet it may appear from this, that 25.5° is a little in advance.

La Victoire has a conspicuous start in an exposure of 5 weeks to 25.5° with an after-exposure to 13°.

Finally it is evident that for a rapid flowering the after-exposure to 13° is more favourable than to 17°.

When we compare the celerrimum for *flower-formation* in fig. 15 with the rate of *coming into bloom* in table 6, we see parallel "curves". After 3 and 5 weeks and 7½ weeks 23° and 25.5° and also 28° are the first to flower in each case, while the flower-cluster-formation is likewise most advanced in these exposures. After an exposure of 12 weeks to the same temperature the flowers from 17° are the first to bloom, from 28° the last. The rate of flower-formation in these temperatures is parallel. Throughout the winter this celerrimum was maintained.

Accordingly a celerrimal effect is obtained by exposure first for some weeks to a high temperature (25.5°, 23° to 28°), next to a low temp. (17°, still better: 13°). If however in the later weeks the temperature is kept high, this retards so much, that permanently 17° and 20° are more rapid than permanently 23°—28°.

It is a striking fact, that this effect in these early varieties is achieved in the same temperatures as in the late variety *Queen of the Blues*. Especially with a view to treatment for early flowering it was important to control this point once more in well-known early varieties.

Number of flowering and non-flowering bulbs.

It has appeared from fig. 15, that in *l'Innocence* every growing-point has proceeded to flower-formation in the chosen temperatures. From table 6 it is to be seen, that it may happen, that the flower does not develop in the lower temperatures, especially in an after-treatment to 13°, preceded by too short a preliminary exposure to but 20° or 17°. After 5 weeks to and above 23° nearly every flower-cluster succeeds, both in the after-exposure to 13° and to 17°.

In *la Victoire* (table 6a) fewer bulbs have fallen behind in the chosen temperatures, from which it may be concluded, that this variety can endure a lower temperature.

The *Queen of the Blues* previously examined was a year younger and already for that reason the number of flowering bulbs is smaller (see § 6).

Number of flowers per cluster.

The number of flower-clusters (= n) varies from 7 to 20 in the two varieties; it usually amounts to 19 or 20 (tabs. 6 and 6a). The mean error of the average number of flowers per cluster was calculated according to JOHANNSEN with the formulas:

$$m = \pm \frac{\sigma}{\sqrt{n}} \text{ and } \sigma = \pm \sqrt{\frac{\sum p a^2}{n} - b^2}.$$

For l'Innocence the mean error is fairly uniformly low. In 3 weeks 17° and 20° and next in 13° it is greatest (number of flower-clusters = 7). For la Victoire the mean errors differ more and are greater on account of the deviations of the variants of the mean value being greater and more irregular, owing to the great number of flowers that occurs on the clusters in this variety.

In what temperature must the preliminary exposure take place and after what time? Taking the mean error ($M \pm 3 m$) into consideration, we can but point out differences between the greatest and the smallest numbers in l'Innocence. The ever recurring optimum at ca 25.5° after 3, 5, ca 8 and 12 weeks is however striking. Even clearer is a similar optimum in la Victoire, but here it is found in 20° (after 3 and 5 weeks also in 23°). In 23°, 25.5° and 28° the numbers get smaller and smaller. The preliminary treatment must last longer in the two varieties than 3 weeks, though this is not so necessary, when the above-mentioned optima are applied.

If we consider whether the after-treatment to 17° or to 13° is necessary, we must not give preference to either of them in l'Innocence. For only where the slightest number of flowers occurs (3 w. 28° + 9 w. 13°) there is, taking the mean error into account, a real difference to be stated between the treatment in 17° and in 13°. In la Victoire this is different: When the preliminary exposure to 20°—28° lasts but 3 weeks, 17° is better, because in that case a greater number of flowers remains rudimentary in 13°. When the preliminary exposure to 20°—28° lasts 5 weeks, the number of rudimentary flowers is smaller and when an after-exposure to 13° or to 17° follows no real difference in number can be pointed out. Accordingly here (with the mean error) it may be more or less traced in the table, where the number of rudimentary flowers is great.

Where these rudimentary flowers occur in la Victoire the following indications are made: In the tables 6 and 6a sub "number of flowers per cluster" an asterisk or an asterisk between brackets has been placed. In the case of la Victoire this means, that in those cases the clusters bore a number of apical flowers (from 1 to 5) which showed aberrations due to the treatment (as in Queen of the Blues, BLAAUW 1924, first part § 8). With (*) is indicated, that it was only distinctly observed on one of the clusters. Only those flowers were taken into consideration, which could be clearly distinguished with the naked eye. It occurs in 3 and 5 weeks only

then, when an after-treatment to 13° took place. In 5 weeks 28° and next 13° it does no more occur; it was observed still however at a cluster in 7½ weeks 17° and next 13°.

Especially when many apical flowers fall behind, this goes together with a thinner common flower-stalk and smaller and paler flowers.

Though therefore in the after-treatment to 13° the celerrima are found, they are no optima here in the sense that the flower-cluster is best developed and richest in flowers.

In the case of l'Innocence * and (*) indicate, that the tepals in those cases were slightly greenish at the apex. Generally this does not occur in the after-treatment to 17°, but it does occur when the bulbs have been kept in the same temperature for 12 weeks.

§ 5. Increase in weight of the bulbs.

On Sept. 27, 1926 (after the treatment in the various temperatures) the sets of bulbs were weighed and next planted in a cistern with equal soil and ground-water-level, as has already been described in § 4. From this date to the date of lifting (July 4, 1927) they have been, though all in an equal measure, exposed to various influences of the weather. The differences, we find in the tables 7 and 7a therefore are due to the different exposures in the previous summer, while after that for months together the climate has influenced them all equally.

The small figures in the tables denote the weight per bulb (an average of 20 bulbs) on Sept. 27, 1926 and on July 5, 1927 (for l'Innocence) and on July 6, 1927 (for la Victoire). So after lifting they were weighed on two subsequent days, which has been recorded, because the loss of weight in one day is already worth mentioning. After each brace the difference, i.e. the average increase in weight, has been given in large figures.

TABLE 7. l'Innocence.

Increase in weight per bulb in grams from Sept. 27, 1926 (date of planting) to July 4, 1927 (date of lifting)

		3 weeks		5 weeks		7½ weeks		12 weeks	
Preliminary exposure	17°	47.1 } 28.4 } 19.7	—	48.1 } 28.5 } 19.6	—	—	—	52.6 } 27.7 } 24.9	and 50.3 } 27.7 } 22.6
	20°	51.8 } 28.1 } 23.7	54.5 } 27.7 } 27.8	50.4 } 28.4 } 22.0	50.0 } 27.4 } 22.6	50.1 } 27.6 } 22.5	49.8 } 27.7 } 22.1	51.9 } 27.9 } 24.0	
	23°	49.4 } 28.3 } 21.1	54.2 } 28.2 } 26.0	49.7 } 28.1 } 21.6	51.7 } 28.2 } 23.5	52.0 } 28.3 } 23.7	54.7 } 27.8 } 26.9	55.1 } 28.6 } 26.5	
	25.5°	46.9 } 28.5 } 18.4	54.1 } 27.4 } 26.7	51.6 } 28.2 } 23.4	58.0 } 28.0 } 30.0	55.3 } 27.9 } 27.4	53.6 } 28.0 } 25.6	53.7 } 28.4 } 25.3	
	28°	57.9 } 27.7 } 30.2	58.3 } 27.7 } 30.6	52.3 } 28.1 } 24.2	56.3 } 28.0 } 238.	53.5 } 27.8 } 25.7	54.3 } 27.9 } 26.4	56.4 } 28.6 } 27.8	
After-exposure to:	13°	17°	13°	17°	13°	17°	—		

TABLE 7a. *la Victoire*, as table 7.

Preliminary exposure	3 weeks		5 weeks		7½ weeks		12 weeks	
	17°	37.1 } 11.6 25.5 }	—	40.8 } 15.1 25.7 }	—	42.4 } 16.7 25.7 }	—	46.0 } 20.2 25.8 }
20°	43.3 } 17.9 25.4 }	45.6 } 20.0 25.6 }	45.2 } 19.8 25.4 }	46.0 } 20.6 25.4 }	41.7 } 16.3 25.4 }	45.0 } 19.2 25.8 }	46.1 } 20.5 25.6 }	
23°	43.7 } 18.3 25.4 }	46.7 } 21.1 25.6 }	44.6 } 19.4 25.2 }	44.1 } 18.7 25.4 }	46.4 } 21.0 25.4 }	45.9 } 20.4 25.8 }	46.5 } 20.3 26.2 }	
25.5°	44.6 } 19.4 25.2 }	47.2 } 21.7 25.5 }	47.4 } 22.2 25.2 }	51.3 } 25.8 25.5 }	49.3 } 24.0 25.3 }	48.3 } 22.8 25.5 }	49.1 } 22.7 26.4 }	
28°	46.8 } 21.8 25.0 }	48.5 } 23.1 25.4 }	46.5 } 21.4 25.1 }	48.7 } 23.6 25.1 }	45.8 } 20.7 25.1 }	48.4 } 23.1 25.3 }	49.5 } 23.3 26.2 }	
After-exposure to:	13°	17°	13°	17°	13°	17°	—	

BLAAUW (1924) points out, that little attention must be paid to slight differences in weight. In this case this is evident from the 2 sets of 20 bulbs which (one by mistake) were both left in 17° for 12 weeks. Here there is a difference of 2.3 grms, so that therefore no value must be attached to 10 % increase in weight.

On comparing table 7 with 7a, it strikes us at once, that the increase in weight in l'Innocence is in every case greater than in *la Victoire*. On our starting the experiment the bulbs were equally large for the two varieties; positively the more vigorous leaves of l'Innocence with larger assimilating surface than those of *la Victoire* act an important part here.

Nearly everywhere the after-treatment in 17° appears to be more favourable than in 13°, especially in l'Innocence. An exception to this is for instance 7½ weeks 25.5° + 4½ w. 13°; yet this difference is smaller than 10 %, so that it is a question whether it must be attached any value to. But it is pointed out here, because in Queen of the Blues (BLAAUW 1924, tab. 32) there was also stated greater increase in weight in this case.

Taking the preliminary exposures also into consideration the following exposures are optimal for l'Innocence: 5 w. 25.5° + 7 w. 17° and 3 w. 28° + 9 w. 13° or 17°. For *la Victoire* 5 w. 25.5° + 7 w. 17°. For Queen of the Blues (BLAAUW 1924, tab. 8) there is an optimum at eight weeks 25.5° + 7 w. 17°. In these early varieties therefore optima are found in the higher temperatures (25.5° and 28°), during a shorter period (3 and 5 weeks).

§ 6. *Summary in connection with the application.*
Comparison with Queen of the Blues.

With respect to the growing out of all leaves in the field in spring it has appeared, that the higher the temperature of the preliminary exposure, the shorter it should last. For

l'Innocence (tab. 2) this temperature may be lower, and needs last shorter than for *Queen of the Blues* (BLAAUW 1924, tabs. 2 and 24). For *la Victoire* (tab. 2a) this temperature may be lower yet and the preliminary exposure shorter than for *l'Innocence*. As to the combination of optima (BLAAUW 1924, §§ 1 and 9) this causes no difficulties, as all following optima for this are found as a rule at so high a temperature and for so long a period, that all leaves shoot.

For the stretching of the foliage till January (judged from the outmost foliage-leaf) for *l'Innocence* (tab. 4) a treatment in 25.5° for 3 weeks and next 13° is optimal. For *la Victoire* (table 4a) this may also be 23° and 28°. The after-exposure to 17° is not favourable in this case, especially not for *la Victoire*. For *Queen of the Blues* (BLAAUW 1924, tab. 4 and § 11) this lies in a higher temperature, which is to be applied for a longer period.

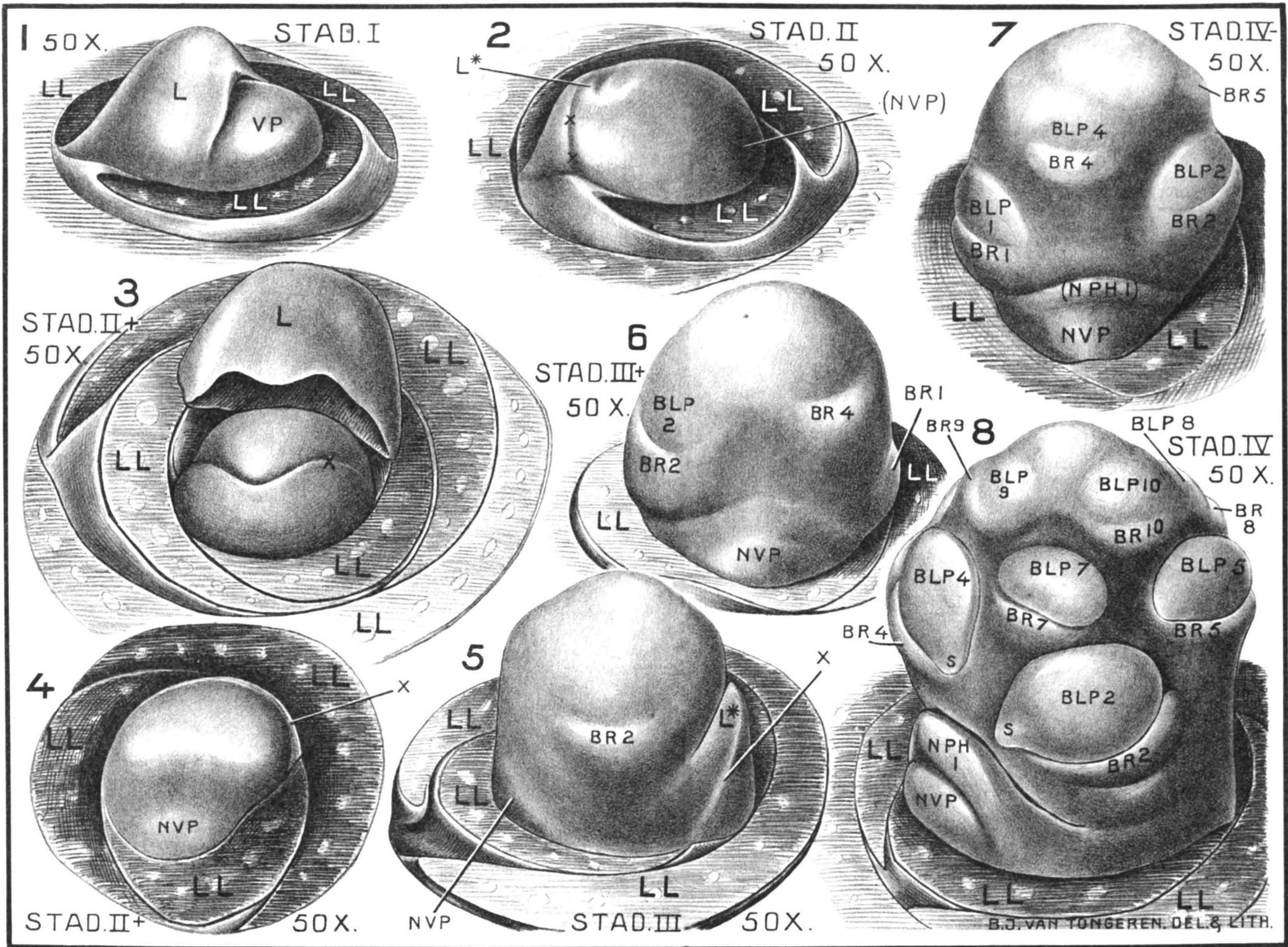
For celerrimal flowering in *l'Innocence* and *la Victoire* 25.5° is necessary for 5 weeks and then 13° till the planting-period (tabs. 6 and 6a). In *l'Innocence* the lower surfaces of the tips of the tepals were slightly green in this case, which does not do much harm to the beautiful appearance of the flower. In *la Victoire* however the apical flowers had remained rudimentary in 13°. This however was stated after a stay for months together outdoors in the field. For a celerrimal flowering (forcing) indoors the results may be more favourable and at any rate an after-treatment in 17° would cause too great a delay i.e. no celerrimal effect. In *Queen of the Blues* (according to experiments not yet published) the celerrimum of flowering is also found at 25.5° or 28° for 5 weeks, followed by an after-treatment in 13°.

Non-flowering bulbs (tabs. 6 and 6a) occur in lower temperatures than the above-mentioned optima, so that — with a bulb-circumference of 12 to 13 cms — we need not take this into account for the combination of the optima.

Queen of the Blues of 7.5—9 cms (BLAAUW 1924, tab. 14) is much closer to the limit of flowering-ability; those of 12—13 cms (ibid. tab. 23) are perfectly parallel, as far as the exposure was identical, with these early varieties. The *Queens* of 7.5—9 cms must not be compared in this respect with the older bulbs of the early varieties, because these have a more vigorous flowering-capacity and consequently yield a greater number of flowering bulbs.

Of *la Victoire* and *l'Innocence* it may be said, that with a short preliminary exposure in this respect the former can endure a lower temperature better than *l'Innocence*.

With respect to the greatest number of flowers per cluster for *l'Innocence* (tab. 6) ca 25.5° for 5 weeks or 7½ weeks and next 13° or 17° is optimal. In the same way the *Queen of the Blues* of



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12—13 cms (BLAAUW 1924, tab. 23) showed an optimum at 8 w. $25.5^{\circ} + 4$ w. 17° . It should be mentioned here, that in order to be compared, the bulbs must be of equal age, as is evident from comparison of tables 19 and 23 in BLAAUW 1924. For la Victoire (tab. 6a) a preliminary exposure to 20° for a long period (up to 12 weeks) is favourable, but for the combination of the optima, especially with a view to increase in weight, we shall have to raise the temperature (23° , still better: 25.5°), but a still higher temperature is markedly detrimental to the number of flowers.

Optima for the increase in weight (tabs. 7 and 7a) for the two varieties lie at 25.5° for 5 weeks and next 17° (besides for l'Innocence at 28° for 3 weeks and next 13° or 17°). An after-treatment in 17° is desirable for those. On comparing with Queen of the Blues (see BLAAUW 1924, tabs. 8 and 26), we are inclined to say that these early varieties should be exposed to the high temperatures for a shorter period than this Queen.

Final conclusion. For an optimal field-culture ca 25.5° for *eight weeks* and next 17° has appeared to be the best treatment for the late variety Queen of the Blues. The early varieties l'Innocence and la Victoire we had probably better expose to 25.5° for a shorter period (e.g. 5 weeks) and next to 17° .

This is also proved (with l'Innocence) by the fact that the flower-cluster after 8 weeks in 23° and 25.5° has already progressed so far, as is evident from the curve of dots (fig. 15).

We emphasize that we refer to healthy bulbs. For severely diseased sets quite a different treatment may be desirable as a cure.

Laboratory for
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Wageningen, October 1927.

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