Botany. — *Plasmatic influences upon the inheritance in Vicia Faba.* III. *The elimination of a definite factor (variegated) as caused by the type of plasm.* By M. J. SIRKS (Instituut voor Plantenveredeling, Wageningen). (Communicated by Prof. J. C. SCHOUTE).

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Chlorophyll-inheritance in the broad bean (Vicia Faba L.) seems to be dependent upon various factors. First of all there is a factor *A*, which has been observed in almost all the strains used in my crosses; it produces normal green colour (typica) and it is epistatic to all other chlorophyll-factors, which can be observed in *a*-plants only. Besides a number of multiple allelomorphs has been detected:

C_3 a green colour, brighter than that which is shown in typica-plants (subtypica), characteristic for strain 4;

C_2 more yellowish green (semichlorina) found in strain 4*a*;

C_1 yellow-green (chlorina) which has been obtained in segregations from crosses between *A*- and *a*-plants only;

c white (albina), seedlings dying within ten days; it occurred in strain 1*a*, in which heterozygous individuals always produce about 25 % of white seedlings.

An exact definition of the colours produced by these factors may be obtained by comparing them with the various types of colours, that have been published in "Ostwalds grosser Farbenatlas" or by means of a colorimeter after separating chlorophylls from xanthophylls on WILLSTAETTERs method. The results of this comparison were:

	Ostwald	Colorimeter	
		Chlorophylls	Xanthophylls
Typica	92 . lg—ng	100.0	100.0
Subtypica	92 . le—ne	98.3	99.0
Semichlorina	96 . le	66.9	71.5
Chlorina	00 . le—ne	45.3	47.8

To these factors, all of which produce plain-coloured phaenotypes, a factor *V* for variegation may be added. It has appeared that this factor exerts its influence on subtypica- and on semichlorina-colours only; phaenotypically it does not appear in *A*-plants, though genotypically it may be present; neither can its presence be observed in C_1-plants. This

type of variegation produces plants with chlorina-coloured leaves, in which spots of subtypica or semichlorina are dispersed. In many cases these variegated plants bear one or more stems, which have plain green leaves only, thus allowing an exact determination of the green colour. First this factor for variegation has been found in strain *4b*; a number of crosses between *A*-types and C-plants further have led to the conclusion that the *V*-factor is present also in the typica-coloured strains 2, *3b*, 11 and 14.

The results of some crosses, in which this factor for variegation was involved, have been summarized in table 1. From these data it may be stated that:

1°. The factor *A* is dominant to *a* (typica to non-typica); C_3 is dominant to C_2, C_2 to C_1 and C_1 to *c*; *V* is dominant to *v* (variegata to plaingreen, non-typica). The factors *A*, (C_3-C_2-C_1-*c*) and *V* do segregate independently from each other.

2°. Cross 69 (genotype of F_1: AaC_3C_3VV) should segregate according to 3 typicae (AC_3V) : 1 variegated (aC_3V); this has been ascertained.

3°. Crosses 2, 8, 50 and 87 (genotype of F_1: AaC_3cVv) should segregate according to 48 typicae (AC_3V and AC_3v) : 9 variegatae (aC_3V) : 3 other greens (aC_3v) : 4 albinae (acV and acv). No conclusive deviations have been found.

4°. Crosses 7, *7a*, 48, 55 and 56 (genotype of F_1: AaC_3C_3Vv or AaC_3C_2Vv) should produce segregations into 12 typicae (AC_3V or AC_2V and AC_3v or AC_2v) : 3 variegatae (aC_3V or aC_2V) : 1 other green (aC_3v or aC_2v). This expectation has been affirmed.

5°. Crosses 46, *46a*, 47, *47a* (genotypes of F_1 : aaC_3C_3Vv or aaC_3C_2Vv) should segregate into 3 variegatae (aC_3V or aC_2V) : 1 plain green (aC_3v or aC_2v). Here the deviations from the expectations are too large; the total of these crosses segregated into 415 variegatae : 36 plaingreen individuals, thus giving $D:m = 8.66$. Some additional cause must have disturbed the regular segregation of factors.

6°. This disturbing cause may be found in some part played by the plasm of the F_1-plant. Crosses 43 and *43a* produced two genotypes of F_1-plants, owing to the heterozygous *Aaccvv*-nature of one of the parents. Some of these F_1-plants possessed the genotype AaC_3cVv and the typica-phaenotype and here the segregations agreed with the expectation on mendelian lines; in F_2 a segregation into 154 typicae : 27 variegatae : 7 subtypicae : 9 albinae has been stated (48 : 9 : 3 : 4 :). In the other F_1-plants (genotype aaC_3cVv; phaenotype variegated) the segregation again showed to be abnormal : 80 variegatae : 5 subtypicae : 24 albinae have been found, in which the variegatae-subtypicae-ratio deviates considerably from the theoretical proportions 9 : 3 : 4 ($D:m$ for each of the croses about 2.5; for both crosses combined about 3.7).

The above seems to lead to this preliminary conclusion:

In F_1-plants which have the typica-phaenotype the segregation of the

V-factor corresponds with the expectations. In F_2-generations produced by phaenotypically variegated F_1-plants however, there is a considerable excess of variegated plants, combined with a deficiency of plain-green (subtypica or semichlorina) individuals.

A further view on the cause of this abnormal segregation may be obtained by considering the data which have been produced in six generations of descendants from one variegated individual by selfing (table 2). The genotypical formula of this individual has been ascertained to be aaC_3C_1Vv, thus being heterozygous for subtypica-chlorina and for the variegata-factor.

A. Variegata-parents from variegata-grandparents may give a uniform variegated offspring (C_3C_3VV) or their offspring may segregate into variegated + variegated with green stems + subtypica + chlorina (C_3cVv) or into variegated + variegated with green stems + subtypica (C_3C_3Vv) or into variegated + chlorina (C_3cVV). A strong deficiency of subtypica-individuals has been found.

B. Variegated parents which came from subtypica-grandparents (cf. sub *C*) segregate into var. + var. g.s. + subt. + chl. or into var. + var. g.s. + subt.; constant variegated plants or segregations var. : chl. never have been observed.

C. Subtypica-plants from variegated grandparents, segregate in two ways only : var. + var. g.s. + subt. + chl. or var. + var. g.s. + subt. No other segregations have been observed.

D. Subtypica-plants from subtypica-grandparents produce a constant subtypica-offspring or they segregate into subtypica : chlorina only.

E. Offspring from variegated stems on variegated plants with green stems behave in quite the same way like the parents mentioned sub *B*, irrespectively as to their origin from variegated or from green grandparents. Offspring from subtypica-stems on the same plants behave like the plants mentioned sub *C*.

From these data the following conclusions may be drawn :

1°. The segregation (variegata + subtypica) : chlorina is in accordance with a 3 : 1-expectation (totals found 3774 : 1113), though the number of chlorina-plants is too small as a result of the deficiency in germination-capacity for seeds of the chlorina-type when compared with green individuals.

2°. The subtypica-individuals which spring from variegated parents do not represent homozygous recessive *vv*-individuals, but they possess the dominant factor *V*; their genotype thus being *Vv*, which is entirely disguised by a change into phaenotypically plain-coloured subtypica-individuals.

3°. Variegated stems and subtypica-stems on the same plants are found on heterozygous *Vv*-individuals only; genotypically they are identical.

4°. The proportions of the various groups of offspring are regulated by the type of plasm in which the offspring is being formed. In variegated

TABLE 1. INHERITANCE OF THE VARIEGATA-CHARACTER IN CROSSES.

Cr.	Parents	Genotype		Phaenotype	nF_2	Segregations				Theoretical proportions	D : m			
		Mother	Father	F_1		Typ.	Var.	Other green	Alb.		Typ.	Var.	Other green	Alb.
69	3b × 4b	AC_3V	aC_3V	typica	362	278	84	—	—	3 : 1	+ 0.79	− 0.79	—	—
2	2 × 4	AcV	aC_3v	typica	189	134	29	10	16	48 : 9 : 3 : 4	− 1.36	+ 0.50	+ 0.40	+ 1.29
8	4 × 14	aC_3v	AcV	typica	185	138	26	8	13	48 : 9 : 3 : 4	− 0.13	− 0.01	− 0.24	+ 0.45
50	4b × 17	aC_3V	Acv	typica	227	178	30	9	10	48 : 9 : 3 : 4	+ 1.21	− 0.35	− 0.53	− 1.16
87	4b × 17b	aC_3V	Acv	typica	198	155	27	7	9	48 : 9 : 3 : 4	+ 1.09	− 0.18	− 0.78	− 1.00
Total					799	605	112	34	48	48 : 9 : 3 : 4	+ 0.45	− 0.03	− 0.55	− 0.28
7	4 × 11	aC_3v	AC_2V	typica	154	119	25	10	—	12 : 3 : 1	+ 0.64	− 0.78	+ 0.13	—
7a	11 × 4	AC_2V	aC_3v	typica	165	122	31	12	—	12 : 3 : 1	− 0.32	+ 0.01	+ 0.54	—
48	4b × 13	aC_3V	AC_2v	typica	122	94	19	9	—	12 : 3 : 1	+ 0.52	− 0.90	+ 0.51	—
55	4b × 8	aC_3V	AC_3v	typica	148	114	26	8	—	12 : 3 : 1	+ 0.56	− 0.37	− 0.41	—
56	6 × 4b	AC_3v	aC_3V	typica	151	116	28	7	—	12 : 3 : 1	+ 0.52	− 0.07	− 0.81	—
Total					740	565	129	46	—	12 : 3 : 1	+ 0.85	− 0.92	− 0.37	—
46	4 × 4b	aC_3v	aC_3V	variegata	153	—	142	11	—	3 : 1	—	+ 5.14	− 5.14	—
46a	4b × 4	aC_3V	aC_3v	variegata	79	—	73	6	—	3 : 1	—	+ 3.62	− 3.62	—
47	4a × 4b	aC_2v	aC_3V	variegata	111	—	102	9	—	3 : 1	—	+ 4.09	− 4.09	—
47a	4b × 4a	aC_3V	aC_2v	variegata	108	—	98	10	—	3 : 1	—	+ 3.85	− 3.85	—
Total					451	—	415	36	—	3 : 1	—	+ 8.66	− 8.66	—
43	1a × 4b	Acv	aC_3V	typica	131	103	16	5	7	48 : 9 : 3 : 4	+ 0.98	− 0.64	− 0.47	− 0.44
		acv	aC_3V	variegata	61	—	44	3	14	9 : 3 : 4	—	+ 2.50	− 2.38	− 0.36
43a	4b × 1a	aC_3V	Acv	typica	66	51	11	2	2	48 : 9 : 3 : 4	+ 0.27	+ 0.60	− 0.64	− 1.14
		aC_3V	acv	variegata	48	—	36	2	10	9 : 3 : 4	—	+ 2.03	− 2.55	− 1.17

TABLE 2. *Summarized offspring from one variegated plant by selfing.*

A. Offspring from variegated plants from variegated grandparents.

Gen.	Var.	Var. g.s.	Subt.	Chl.
D 2	14	1	1	5
D 2	67	5	4	15
D 3	62	8	10	21
D 4	219	27	20	81
D 6	217	33	19	79
Tot.	579	74	54	201

Besides a number of constant variegated plants from D1 on have been obtained.

Gen.	Var.	Var. g.s.	Subt.
D 2	19	1	1
D 2	13	1	—
D 3	171	21	17
D 3	69	9	12
D 4	327	41	32
D 4	216	24	21
D 4	143	17	15
D 6	311	37	29
D 6	173	21	17
D 6	123	13	19
Tot.	1565	185	163

Gen.	Var.	Chl.
D 2	57	18
D 3	17	3
D 3	227	63
D 4	116	35
D 4	167	48
D 6	315	93
Tot.	899	260

B. Offspring from variegated plants from subtypica-grandparents.

Gen.	Var.	Var. g.s.	Subt.	Chl.
D 6	269	17	29	98
Tot.	269	17	29	98

Gen.	Var.	Var. g.s.	Subt.
D 4	101	11	9
D 6	196	17	12
D 6	439	62	93
D 6	399	55	43
Tot.	1135	145	157

C. Offspring from subtypica-plants from variegated grandparents.

Gen.	Var.	Var. g.s.	Subt.	Chl.
D 4	17	13	34	26
D 6	136	31	181	112
Tot.	153	44	215	138

Gen.	Var.	Var. g.s.	Subt.
D 3	10	4	17
D 3	11	6	19
D 4	71	19	111
D 4	81	13	119
D 4	9	7	19
D 6	97	17	118
D 6	73	19	103
D 6	83	9	99
Tot.	435	94	605

TABLE 2 (*Continued*). *Summarized offspring from one variegated plant by selfing.*

D. Offspring from subtypica-plants from subtypica-grandparents.

Gen.	Subt.	Chl.	Gen.	Subt.
D 6	327	94	D 4	309
			D 5	387
			D 6	927
			D 6	73
			D 6	229
Tot.	327	94	Tot.	1925

E. 1. Offspring from variegated stems on variegated plants with subtypica-stems from variegated grandparents.

Gen.	Var.	Var. g. s.	Subt.	Chl.	Gen.	Var.	Var. g. s.	Subt.
D 4	17	4	5	11	D 3	6	1	1
D 5	171	32	39	72	D 4	141	16	9
D 6	147	12	25	57	D 4	49	6	10
					D 4	15	2	4
					D 5	533	57	61
					D 6	613	48	69
					D 5	119	15	23
					D 6	219	9	21
					D 6	183	21	17
Tot.	335	48	69	140	Tot.	1878	165	215

E. 2. Offspring from subtypica-stems on variegated plants with subtypica-stems from variegated grandparents.

Gen.	Var.	Var. g. s.	Subt.	Chl.	Gen.	Var.	Var. g. s.	Subt.
D 4	13	9	25	17	D 3	5	2	10
D 5	71	16	93	48	D 4	83	12	103
D 6	62	17	81	50	D 4	18	12	33
					D 4	10	8	21
					D 5	279	39	387
					D 6	391	68	467
					D 5	61	16	73
					D 6	121	49	173
					D 6	103	27	119
Tot.	146	42	199	115	Tot.	1071	233	1386

TABLE 2 (*Continued*). *Summarized offspring from one variegated plant by selfing.*

E. 3. Offspring from variegated stems on variegated plants with subtypica-stems from subtypica-grandparents.

Gen.	Var.	Var. g.s.	Subt.	Chl.	Gen.	Var.	Var.g.s.	Subt.
D6	119	7	8	38	D4	47	5	8
					D6	77	5	11
					D6	431	44	50
					D6	75	5	10
Tot.	119	7	8	38	Tot.	630	59	79

E. 4. Offspring from subtypica-stems on variegated plants with subtypica-stems from subtypica-grandparents.

Gen.	Var.	Var. g.s.	Subt.	Chl.	Gen.	Var.	Var.g.s.	Subt.
D6	53	14	74	29	D4	21	13	42
					D6	49	17	74
					D6	253	49	331
					D6	33	8	47
Tot.	53	14	74	29	Tot.	356	87	494

plasm almost all descendants have the variegated type with a minority of phaenotypically subtypica-individuals, whose genotype however is variegated. In subtypica-plasm two genotypical groups of offspring are born: one group of heterozygous variegated plants and one group of real subtypica-individuals; the ratios of both groups being about 1 : 1 with a slight excess of subtypicae (totals 2720 variegatae : 2973 subtypicae).

Thus the main conclusion from the above may be this one:

Plants which are heterozygous for the V-factor (Vv) in their segregation depend upon the type of plasm in this way:

In typica-plasm the segregation is quite regular into 1 VV : 2 Vv : 1 vv.

In variegata-plasm in one of the sexes those gametes that carry the recessive v-factor are discarded, the result being a permanent back-cross between VV and Vv and an apparent constant inheritance of the variegata-character, a number of phaenotypical subtypica-plants excepted.

In subtypica-plasm those gametes that carry the dominant V-factor are eliminated in one of the sexes, thus producing a permanent Vv : vv-segregation according to an 1 : 1-ratio.

It is to be regretted that no back-crosses between *Vv*-individuals and *vv*-plants were available so that the question in what sex this elimination happens, remains undecided. Owing to technical difficulties, the obtainment of a sufficient number of offspring from such backcrosses would have caused a good deal of trouble.