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PLEIOTROPY IN SUNFLOWER (*Helianthus annuus* L.)

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SUMMARY

A case of pleiotropy in sunflower was detected while studying the inheritance and inter-relationships of genes governing anthocyanin pigmentation of the petiole, midrib, leaf margin, leaf tip, ray floret and stigma in a cross between the inbreds RM-1 and ITP-1. Pigmentation was dominant in the F_1 generation. Complementary gene action of 9 purple: 7 green was observed for petiole, midrib, leaf margin, leaf tip and ray floret colourations in the F_2 generation. Colour of the stigma was found to be controlled by four pairs of factors which segregated into 207 coloured: 49 couluoress in the F_2. The gene symbols were designated for the above mentioned characters. The genetic ratios obtained in the F_2 generation are verified with the data collected from the F_3 generation. The occurrence of pleiotropism in sunflower is reported here for the first time.

Key words: Pleiotropy, sunflower, *Helianthus annuus* L.

INTRODUCTION

Anthocyanin pigmentation occurs in many parts of sunflower plant. The inheritance of pigmentation seems to be not simple as many genes are involved in colour development. Pleiotropic genes manifest more characters than one alongwith theprimary effect. Hence the selection for a particular trait automatically leads to the selection for other traits too, which is of immense importance to breeders. Most of the available literature deals with the inheritance of floral/morphological variations (Cockrell, 1912; Leclercq, 1968; Fick, 1976; Hiremath and Giriraj, 1984). Though these reports indicate that differences in pigmentation or morphology are qualitative in nature, controlled by one or two major genes (Fick, 1976), the knowledge of their inter-relationships is rather scant. Hence the present investigation was carried out to study the inter-relationships between the genes controlling such characters.

MATERIAL AND METHODS

Two genotypes of sunflower (*Helianthus annuus* L.) viz., RM-1 and ITP-1, maintenained at the sunflower seed production project, GKVK, Bangalore, were selected for the study. Crossing was done during the rabi season of 1990. The F_1 and F_2 progenies were grown during subsequent seasons. The F_2 population consisted of 1554 plants an 59 families were raised in the F_3 generation with 60 plants in each family. X^2 test was employed to assess the independent assortment of genes and pleiotropy. Gene symbols were used as per the recommendations of the International Committee on Genetic Symbols and Nomenclature (Tanaka, 1957).

RESULTS AND DISCUSSION

The line RM-1 was devoid of pigment in all plant parts while the other parent, ITP-1, showed pigmentation on the petiole, midrib, leaf margin, leaf tip, ray floret and stigma. Pigmentation was found to be dominant in the F_1 generation (Table 1). The analysis of the F_2 segregation (9 coloured: 7 colourless/green) followed by the F_3 confirmation (Table 3) revealed the occurrence of two complementary factors for colour on the petiole, mid rib, leaf margin, leaf tip and ray floret. Two sets of complementary genes were responsible for pigmentation on the stigma, out of which either the first or the second set of complementary genes in dominant condition produces the colour. The F_2 plants segregated in the ratio of 207 pigmented: 49 non-pigmented.

Table 1. Frequencies of phenotypic segregations in the F_2 for six characters in the cross RM-1xITP-1

Character		F_2 segregation		Ratio	X^2	'P' value
		Purple	Green			
Petiole	Observed	902.00	652.00			
	Expected	874.13	679.88	9:7	2.0300	0.20-0.10
Midrib	Observed	849.00	705.00			
	Expected	874.13	679.88	9:7	1.6500	0.25-0.10
Leaf margin	Observed	879.00	675.00			
	Expected	874.13	679.88	9:7	0.0620	0.90-0.70
Leaf tip	Observed	876.00	678.00			
	Expected	874.13	679.88	9:7	0.0097	0.95-0.90
		*	**			
Ray floret	Observed	881.00	673.00			
	Expected	874.13	679.88	9:7	0.1236	0.75-0.55
Stigma	Observed	1275.00	279.00			
	Expected	1311.19	242.81	207:49	1.4100	0.25-0.10

*PS – Purple striations **Y – Yellow colour.

Pleiotropy: Combined segregation data (Table 2) reveal the existence of pleiotropy for pigmentation on the petiole, midrib, leaf margin, leaf tip, ray floret and stigma involving a single gene. The pleiotopic nature of this gene and its inter-relationship with the other genes is shown diagrammatically in Figure 1. The joint ratio of 81:63:63:49 for the petiole with mid-rib, petiole with leaf margin, petiole with leaf tip and petiole with ray floret colour was modified into 27:9:9:19 as one gene is common among these characters (Table 2). But the joint ratio of 1863:441:1449:343 for the petiole with stigma, mid rib with stigma, leaf margin with stigma, leaf tip with stigma and ray floret with stigma respectively, was modified into 513:63:315:133 in the presence of the same single pleiotropic gene. The factors controlling petiole colour were designated by the gene symbols **Ptl**$_a$ and **Ptl**$_b$. One of these two complementary genes of petiole colouration, **Ptl**$_a$, was common for pigmentation on the midrib, leaf margin, leaf tip, ray floret and stigma. This pleiotropic gene, **Ptl**$_a$, acted as basic complementary gene and is responsible for the expression of colour on the aforementioned characteres in combination with the

Table 2. Frequencies of different pairs of characters in the joint segregations for the cross RM-1xITP-1

Character with ratio	Joint ratio F_2		Joint segregation					
			PP	PG	GP	GG	X^2	'P' Value
1	2	3	4	5	6	7	8	9
1. Petiole (9:7)								
		Obs.	660.00	242.00	294.00	458.00		
Midrib (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	288.68	0.25-0.10
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	5.33	
		Obs.	681.00	221.00	198.00	454.00		
Leaf margin (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	312.34	0.50-0.30
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	3.31	
		Obs.	677.00	225.00	199.00	453.00		
Leaf tip (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	303.99	0.50-0.30
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.79	
		Obs.	674.00	228.00	207.00	445.00		
Ray floret (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	283.64	0.70-0.50
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.11	
		Obs.	800.00	102.00	475.00	177.00		
Stigma (207:49)	1863:441:449:343	Exp.Ind.	706.81	167.31	549.74	130.13	64.82	0.25-0.10
	513:63:315:133	Exp.*	778.52	95.61	478.04	201.84	4.20	
2. Midrib (9:7) with:								
		Obs.	648.00	231.00	201.00	474.00		
Leaf margin (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	300.53	
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.55	0.50-0.30
		Obs.	650.00	226.00	199.00	479.00		
Leaf tip (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	313.76	
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.72	0.50-0.30
		Obs.	648.00	201.00	233.00	472.00		
Ray floret (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	296.60	
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.70	0.50-0.30
		Obs.	769.00	80.00	506.00	199.00		
Stigma (207:49)	1863:441:1449:348	Exp.Ind.	706.81	167.31	549.74	130.13	90.96	
	513:63:315:133	Exp.*	778.52	95.61	478.04	201.84	4.34	0.25-0.10
3. Leaf margin (9:7) with:								
		Obs.	673.00	203.00	206.00	472.00		
Leaf tip (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	334.88	
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.53	0.50-0.30
		Obs.	868.00	11.00	292.00	383.00		
Ray floret (9:7)	81:63:63:49	Exp.Ind.	655.59	218.53	509.90	169.59	625.94	
	27:9:9:19	Exp.*	863.66	10.46	301.83	378.04	0.43	0.95-0.90
		Obs. 798.00	81.00	477.00	198.00			
Stigma (207:49)	1863:441:1449:343	Exp.Ind.	706.81	167.31	549.74	130.13	101.31	
	513:63:315:133	Exp.*	778.52	95.61	478.04	201.84	2.79	0.50-0.30
4. Leaf tip (9:7) with:								
		Obs.	647.00	229.00	234.00	444.00		
Ray floret (9:7)	81:63:63:49	Exp.Ind.	491.69	382.43	382.43	297.44	240.44	
	27:9:9:19	Exp.*	655.59	218.53	218.53	461.34	2.36	0.70-0.50
		Obs.	796.00	80.00	479.00	199.00		
Stigma (207:49)	1863:441:1449:343	Exp.Ind.	706.81	167.31	549.74	130.13	102.37	
	513:63:315:133	Exp.*	778.52	95.61	478.04	201.84	2.98	0.50-0.30
5. Ray floret (9:7) with:								
		Obs.	800.00	81.00	475.00	198.00		
Stigma (207:49)	1863:441:1449:348	Exp.Ind.	706.81	167.31	549.74	130.13	102.37	
	513:63:315:133	Exp.*	778.52	95.61	478.04	201.84	2.98	0.50-0.30

Obs: - Observed values. Exp. Ind.: Expected values on independent basis.
Exp.*: Expected values when one gene is common.

Table 3. F_3 breeding behaviour of 59 families for six characters in the cross RM-1 x ITP-1

Character and expected ratio in the F_3 generation with observed figures			Number of families segregating for							
		BTD	3:1	9:7	15:1	57:7	207:49	BTR	X^2	'P' value
Expected ratio										
1:4:4:7	Exp.	3.69	14.75	14.75	-	-	-	25.81	-	-
Observed figures for petiole (9:7)	Obs.	5.00	16.00	16.00	-	-	-	22.00	1.24	0.75-0.70
Midrib (9:7	Obs.	4.00	17.00	16.00	-	-	-	22.00	1.04	0.90-0.75
Leaf margin (9:7)	Obs.	5.00	16.00	16.00	-	-	-	22.00	1.24	0.75-0.70
Leaf tip (9:7)	Obs.	5.00	16.00	16.00	-	-	-	22.00	1.24	0.75-0.70
Ray floret (9:7)	Obs.	4.00	13.00	15.00	-	-	-	27.00	0.29	>0.95
Expected ratio										
31:56:56:16:32:16:49	Exp.	7.14	12.91	12.91	3.69	7.37	3.69	11.29	-	-
Observed figures for stigma (207:49)		8.00	13.00	12.00	4.00	7.00	3.00	12.00	0.39	>0.95

Exp – Expected values
Obs – Observed values
BTD – Breeds true for dominant character
BTR – Breeds true for recessive character

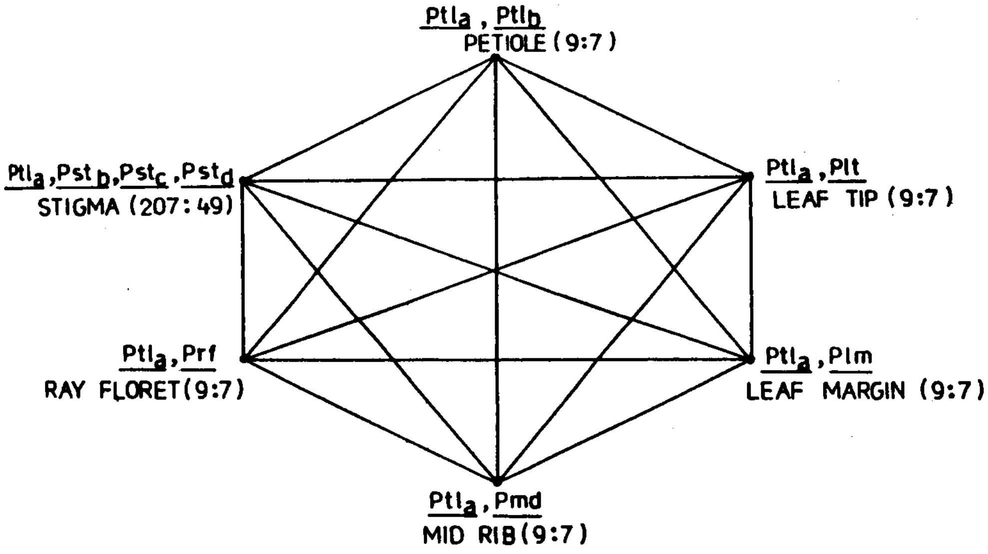

Fig. 1. Pleiotropic gene for anthocyanin pigmented characters in the cross RM$_1$ x ITP-1

other character genes. Taking **Ptl$_a$** as the pleiotropic gene, the gene symbols for these six pigmented plant parts was as follows:

1. Petiole – **Ptl$_a$, (Pl), Ptl$_b$.**
2. Midrib – **Ptl$_a$, (Pl), Pmd$_b$.**
3. Leaf margin – **Ptl$_a$, (Pl), Plm$_a$.**
4. Leaf tip – **Ptl$_a$, (Pl), Plt$_b$.**
5. Ray floret – **Ptl$_a$, (Pl), Prf$_b$.**
6. Stigma – **Ptl$_a$, (Pl), Pst$_b$, Pst$_c$, Pst$_a$.**

This gene action may be explained in terms of biochemical pathways as follows. It is possible that the pleiotropic gene **Ptl$_a$** may be producing a common enzyme which is essential in different biochemical pathways operated by several other genes. Leaving the pleiotropic gene **Ptl$_a$**, as such, the remaining genes for the characters mentioned earlier, such as, **Ptl$_b$, Pmd$_b$, Plm$_b$, Plt$_b$, Prf$_b$** and **Pst$_b$** were ,all localizing genes of their respective plant parts. The enzyme produced by **Ptl$_a$** may mediate the pathways leading to pigmentation in all five plant parts. Experimental proof supporting such explanations of the gene action through biochemical pathways is yet to be advanced.

Inheritance of characters. There are no previous reports available on the inheritance of leaf tip colour and the present finding, that it is because of the interaction of two complementary genes, appears to be the first of its kind. Luckiewich (1975) reported a monogenic control of the petiole and leaf margin colour but the cross between RM1 and ITP-1 has revealed that it is digenically controlled with a complementary gene action. A ratio of 3 purple: 1 green for midrib colour was reported by Dominguez Gimenez (1974) and Stoenescu (1974) in contrast to the present finding of the 9 purple: 7 green ratio. Ray floret colour has been studied by many researchers. Cockrell (1912) reported a single dominant gene for red colour. Leclercq (1986) and Škaloud and Kováčik (1974) obtained a single dominant gene for yellow flowers, whereas Fick (1976) indicated that yellow was dominant to orange and two complementary genes were responsible for the expression of red colour and some yellow lines might have had one of the genes for red colour. Anand and Malik (1977) observed that the exspression of lemon white ray floret is controlled by two recessive genes. Mahacek (1980) concluded that yellow (LL) colour is dominant over sulphur yellow (LL). The orange colour depends on the presence of both "L" and a dominant allele "LO" but a dominant inhibitory gene (II) inhibits the expression of orange colour. The present study reveals that the red\purple ray floret colour was caused by two complementary genes, designated by the gene symbols **Prf$_a$** and **Prf$_b$**. The present study revealed that the inheritance of stigma colour involves four pairs of alleles and not a single allele as reported by Luckiewicz (1975).

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PLEIOTROPIA EN GIRASOL (*Helianthus annuus* L.)

RESUMEN

Un caso de pleiotropia en girasol fue detectado mientras se estudiaba la herencia y la interralación entre los genes que controlan la pigmentación antociánica del peciolo margen y extremo de la hoja y ITP-1. La pigmentación fue dominante en la generación F1. Una acción génica complementaria de 9 purpura: 1 verde fue observada en la generación F2. El color del estigma fue encontrado estar controlado por cuatro pares de factores los cuales secregaron en 207 coloreados: 49 sin color en la generación F2. Los simbolos de los genes fueron de signados para los caracteres mencionados. La segregaciones obtenidas en la generación F2 fueron verificadas con los datos coleccionados de esta generación. La existencia de pleiotropismo en girasol es presentada por primera vez.

PLÉIOTROPIE CHEZ LE TOURNESOL (*Helianthus annuus* L.)

RÉSUMÉ

Un cas de pléiotropie a été détecté chez le tournesol au cours d'une étude concernant l'héritabilité et l'interaction de gènes gouvernant la pigmentation anthocyanique sur les pétioles, nervures, bord des feuilles, feuilles et ITP-1. La pigmentation est un caractére dominant dans la génération F1. Pour la génération F2, nous avons observé l'action de gènes complémentaires, 9 pourpres: 7 verts pour la coloration du pétiole, de la nervure, du bord des feuilles, de l'aprex des feuilles et des fleurs tubulaires. La couleur des stigmates est contrôlée par quatre paires de facteurs qui ségrégent en 207 individus colorés et 49 incolores en F2. Les symboles du gène sont désignés par les caractéres mentionnés précédemment. Les ratios génétiques obtenus en F2 ont été vérifiés à partir des données observées en F3. Cette publication signale pour la premiére fois l'existence de pléiotropisme chez le tournesol.