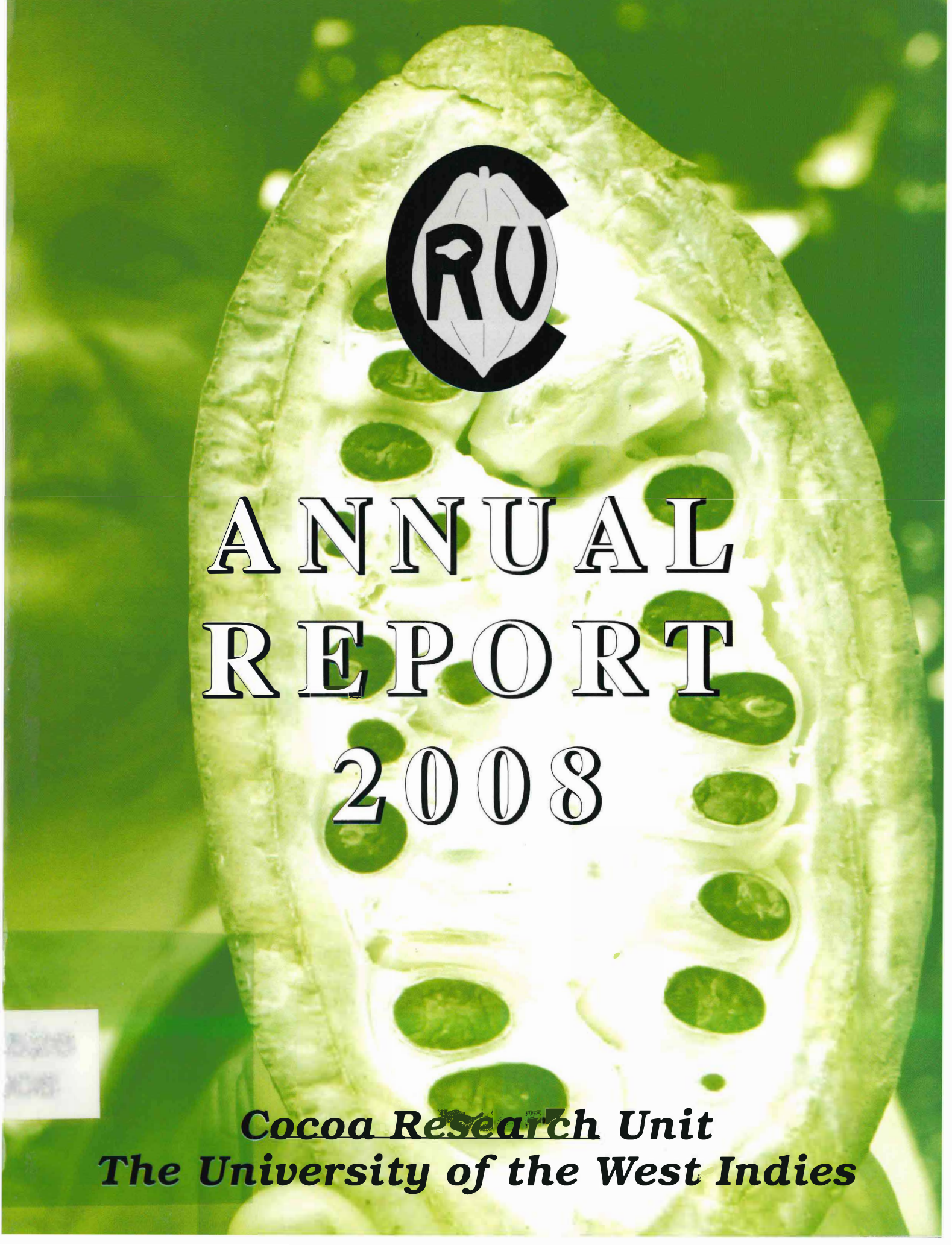

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Cover photograph. Two halves of a fully mature cocoa pod from the ICG,T held by Eusebius Solozano

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An update on the germplasm enhancement for Witches' Broom disease programme

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Introduction

The Witches' Broom germplasm enhancement programme was initiated in July 2004 as part of the CFC/ICCO/Bioversity Cocoa Productivity Project, and resulted in the creation of approximately 5,300 progenies over three years of pollination. Screening of year one progenies was completed and a selection of plants resistant to both WB and BP diseases were planted in the field in 2007. Some of year two crosses were completed in year three, in addition to crosses initially planned for year three. We present here combined results of the screening to Witches' Broom disease for all year two and year three crosses.

Methodology

Data analysis for resistance to Witches' Broom

Two variables were used to measure WB resistance: time to first symptom (TFS) and maximum broom diameter (MBD). Since some of the crosses from year two were completed in year three, data for families and parents from pollination years two and three were combined for analysis. Analyses of variance (ANOVA) for TFS and MBD were prepared with the General Linear Model using Statistical System Analysis[1]. Two analyses were carried out, one with all data from crosses in the incomplete factorial design and the other with data from bi–parental crosses.

Witches' Broom field observations

The first round of observations was done in May 2008 on seedlings and grafted parents planted in the field in October 2007, and originating from the Kempthorne and Curnow incomplete diallele design. For each plant, brooms were counted and then removed in order to avoid weakening the plants. The percentage of seedlings having brooms was calculated.

Results

ANOVA of families from the incomplete factorial design revealed significant differences between families for both variables TFS and MBD (Table 1). One family, *i.e.* MOQ 695 x (IMC 67 × GU 353/L) T64 was considered as very good for both criteria and a few families are considered promising for one of the two criteria: CRU 89 × (ICS 1 × GU 175/P) T28 and LP 3/15 [POU] × CL 10/5 for TFS and AM 2/19 [POU] × SJ 1/40 [POU] and B 9/10-25 [POU] × (IMC 67 × GU 353/L) T64 for MBD.

The factorial analysis showed the superiority of parents AM 2/19 [POU] and CRU 89 for both variables when used as females and of CL 10/5 and (IMC 67 × GU 353/L) T64 for both variables when used as males (Table 2).

[1] SAS Institute, USA

Analysis of variance of the 18 bi-parental crosses not included in a specific experimental design also revealed significant differences between families for both TFS and MBD (Table 3). A group of 13 families produced the smallest brooms with no significant differences between them. Among them, four were classed best for TFS, namely MAN 15/60 [BRA] × IMC 31, CRU 80 × MATINA 1/7, IMC 47 × (NA 45 × B 7/21 [POU]) T83 and PA 126 [PER] × AMAZ 6/3 [CHA].

Table 1. Level of resistance to Witches' Broom disease of crosses made using the incomplete factorial experimental design.

Crosses	TFS[1]			MBD[2]		
	Number of plants	Value (days)	Group	Number of plants	Value (mm)	Group
AM 2/19 [POU] × NA 232	112	14.3	bcd	109	10.2	c
AM 2/19 [POU] × SJ 1/40 [POU]	30	14.1	abcd	27	8.8	a
B 9/10-25 [POU] × CL 10/5	94	13.7	abc	94	9.6	abc
B 9/10-25 [POU] × (IMC 67 × GU 353/L) T64	79	13.7	abc	78	8.9	a
CRU 89 × SJ 1/40 [POU]	77	13.6	abc	76	10.0	bc
CRU 89 × (ICS 1 × GU 175/P) T28	139	14.4	cd	134	10.2	c
LP 3/15 [POU] × CL 10/5	35	14.6	cd	34	11.4	de
MOQ 695 × NA 232	83	13.3	a	82	11.6	e
MOQ 695 × (IMC 67 × GU 353/L) T64	38	14.9	d	37	9.0	ab
PA 195 [PER] × LP 3/15	59	13.5	ab	59	11.6	de
PA 195 [PER] × (ICS 1 × GU 175/P) T28	77	12.9	a	77	10.4	cd

[1] Time to first symptom
[2] Maximum broom diameter

Table 2. Level of resistance to Witches' Broom disease of parents used in the incomplete factorial experimental design.

Clones	TFS[1]		MBD[2]	
	Value (days)	Group	Value (mm)	Group
Female parents				
AM 2/19 [POU]	15.0	c	8.7	a
B 9/10-5 [POU]	12.9	a	10.6	b
CRU 89	14.5	c	9.9	ab
LP 3/15 [POU]	13.9	abc	12.9	c
MOQ 695	14.1	bc	10.6	b
PA 195 [PER]	13.0	ab	10.0	ab
Male parents				
CL 10/5	14.6	c	9.1	ab
LP 3/15 [POU]	14.4	bc	12.4	e
NA 232	13.1	ab	11.6	de
SJ 1/40 [POU]	12.9	a	10.2	bcd
(IMC 67 × GU 353/L) T64	14.6	c	8.5	a
(ICS 1 × GU 175/P) T28	13.9	bc	11.0	cd

[1] Time to first symptom
[2] Maximum broom diameter

Witches' Broom field observations

Twenty–nine percent of the 134 plants found resistant to both WB and BP in the nursery showed symptoms of WB in the field seven months after planting in the field. This is significantly less than the 68.2% of the 55 progeny found susceptible to WB in the greenhouse and now developing brooms in the field.

To ensure that the genotypes planted in the field from year one crosses with combined resistance to WB and BP are not lost, the plants are being replicated by micro-grafting. To date twenty-five percent of this task has been completed.

Table 3. Level of resistance to Witches' Broom disease of bi-parental crosses not belonging to any specific experimental design.

	TFS[1]			MBD[2]		
Crosses	Number of plants	Value (days)	Group	Number of plants	Value (mm)	Group
NA 399 × (SCA 6 × IMC 67) T12	122	15.3	abc	118	9.0	c
ICS 35 × SCA 24	13	15.2	abc	13	9.0	c
CRU 80 × MATINA 1/7	58	16.2	ab	58	9.3	c
TRD 32 × NA 471	27	13.1	c	27	9.5	c
MO 9 × PA 150 [PER]	74	13.6	bc	72	9.5	c
TRD 45 × NA 471	57	14.6	abc	53	9.6	c
PA 126 [PER] × AMAZ 6/3 [CHA]	75	15.7	abc	74	9.6	c
CL 10/5 × (ICS 84 × TSH 1077) T49	84	14.9	abc	83	9.7	c
IMC 47 × (NA 45 × B 7/21 [POU]) T83	101	16.0	ab	101	9.7	c
PA 171 [PER] ×TRD 109	135	13.9	abc	133	9.9	c
CC 71 × NA 33	35	15.4	abc	34	10.0	c
ICS 35 × CL 10/3	24	14.7	abc	24	10.2	c
MAN 15/60 [BRA] × IMC 31	94	16.4	a	92	10.5	c
MAN 15/60 × GU 261/P	34	15.6	abc	32	12.2	b
JA 5/5 [POU] × CC 41	37	15.0	abc	37	12.3	b
MO 9 × LCT EEN 46	122	15.5	abc	119	12.3	b
LV 20 [POU] × LP 34 [POU]	31	13.9	abc	31	12.9	ab
LV 20 [POU] × NA 702	19	14.8	abc	18	13.9	a

[1] Time to first symptom
[2] Maximum broom diameter

Conclusion

The incomplete factorial design used in years two and three identified parents with a longer TFS and those with smaller MBD. TFS and MBD did not always correlate, suggesting that resistance in cocoa operates at pre- and post- penetration stages in the infection process. Surujdeo-Maharaj *et. al* , 2004 also noted the possibility of different mechanisms of resistance in cocoa. The female (AM 2/19 [POU], CRU 89) and male ((IMC 67 × GU 353/L) T64, CL 10/5) parents had both smaller MBD and longer TFS. These aforementioned parents are good candidates for use in a breeding programme to enhance WB resistance based on both mechanisms of resistance.

References

Surujdeo-Maharaj, S., Umaharan, P. and Butler, D.R. (2004) Assessment of resistance to Witches' Broom disease in clonal and segregating populations of *Theobroma cacao*. *Plant Disease* **88:** 797-803.