

COCOA GROWERS' BULLETIN
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COCOA GROWERS' BULLETIN

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EDITORIAL

In recent years, the international cocoa community has debated the need for implementation of more sustainable systems of cocoa cultivation. As highlighted in the Editorial in the last issue of the Cocoa Growers' Bulletin, cocoa is a crop which can make a positive contribution to environmental preservation and indeed to environmental improvement in the humid tropical rainforest zones. As yet these effects are poorly understood, poorly documented and inadequately researched. This research gap needs to be closed and the result will be better methods of growing cocoa into the new millennium.

A number of specific international initiatives have recently been started in addition to ongoing cocoa research and extension activities. Amongst these maybe listed: the International Sustainable Cocoa Programme (ISCP), and the Sustainable Tree Crop Programme financed by USAID and involving cashew, coffee and cocoa actions in four cocoa producing countries in West Africa. These initiatives are intended to supplement the existing efforts on cocoa research and extension and, hopefully, bring better international co-ordination to ensure that the maximum benefit accrues to cocoa growers from the efforts being undertaken in their name.

In an attempt to advance the understanding of this prospect for more sustainable means of growing cocoa in coming decades, we hope to publish a number of articles describing some of the issues and some of the potential solutions to the very real needs. In this issue, we offer the article by Dr Ayanlaja entitled "The Development of an Alley Cropping: A Viable Agroforestry Technology for Sustained Cocoa Producing in Nigeria". Sadly, it was not possible to include this paper in the last issue as was intended. There is also a second article under the sustainable theme in this issue – that of Dr Adeyemi entitled "Effective Management of Cocoa Farms in Nigeria for Profitability".

However, it is clear that the most important contributor to sustainable cocoa production will be the quality of the planting material involved. The article by Drs Adomako and Adu-Ampomah 'Reflections on the yield of Upper Amazon cocoa hybrids in Ghana with reference to breeding for Cocoa Swollen Shoot Virus resistant varieties' adds to our understanding of this issue.

We hope that this will stimulate debate and thus encourage the drive towards more sustainable systems of cocoa cultivation. We would appreciate other articles on sustainable cocoa cultivation systems for future issues of the Cocoa Growers' Bulletin.

Information on the Sustainable Tree Crop Programme can be found on the IITA website (www.iita.org).

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R.A.LASS, EDITOR

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Cocoa Growers' Bulletin No.52

November 2000

REVIEW OF PRODUCTION, CONSUMPTION, STOCKS AND PRICES - I

Mark N. Taylor
(February 1998 - December 1999)

PRODUCTION

E D & F Man Limited Cocoa Market Report No.367 reported that World production in 1998/99 was 2,803 TMT ('000 tonnes), 5.5% higher than the 2,658 TMT produced in 1997/98. Larger crops in Indonesia, Côte d'Ivoire, Ecuador and Nigeria were more than sufficient to offset lower production in Dominican Republic and Brazil.

Production in West Africa was reported to have risen by 5.2% during 1998/99 to reach 1,936 TMT, only 44 TMT below the record crop harvested in 1995/96. Most of this increase came from Côte d'Ivoire and Nigeria. Production in Côte d'Ivoire rose by 62 TMT to 1,175 TMT, the second largest crop ever produced by a single origin. Meanwhile, Nigeria was reported as having produced 202 TMT, its largest crop for 22 years. Outside of West Africa, production in Indonesia and Ecuador recovered from the El Niño-reduced crops in 1997/98. Indonesian production rose to a new record of 393 TMT in 1998/99, while production in Ecuador recovered from the disastrous 1997/98 season (when just 28 TMT was produced) to reach 72 TMT. Production fared less well in Dominican Republic and Brazil. Hurricane Georges devastated production in the former, lowering output from 60 TMT in 1997/98 to just 24 TMT. The decline in Brazilian cocoa production continued with the 1998/99 crop totalling just 130 TMT, the lowest for 35 years.

CONSUMPTION

E D & F Man Limited Cocoa Market Report No.367 stated that World cocoa consumption (as measured by World grind) totalled 2,758 TMT, a fall of 1.9% on the record level seen in 1997/98. They noted that this was the first fall in consumption since 1991/92 and only the second since 1977/78.

Grindings in Europe were reported to have fallen by 5.6% during 1998/99. One of the key factors was the collapse in consumption in Russia. This had the effect of reducing East European grindings by more than 23% and also had a negative impact on grindings in Germany. It was also reported that a major processing company had reduced grinding activity in an attempt to boost cocoa butter prices. This was a key factor behind UK grindings falling to their lowest level since 1994/95 and the first reduction in the Netherlands grind since 1976/77.

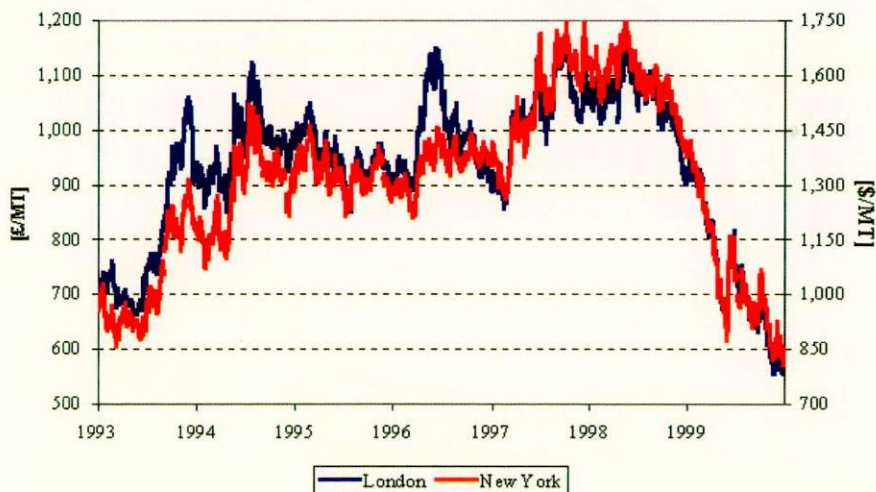
STOCKS

E D & F Man Limited Cocoa Market Report No.367 indicated that for the 1998/99 season there would be a modest surplus of 17 TMT, only the 2nd surplus in 8 years. End of season stocks were expected to remain high at more than 1.2 million tonnes or 44% of Global usage.

PRICES

Prices fell sharply between May 1998 and June 1999 as the economic crisis in Russia sparked a massive reversal in market sentiment. Heavy selling by speculators, coupled with the absence of buying by manufacturers (who had previously held long cover due to fears surrounding liberalisation in Côte d'Ivoire and the threat to production from El Niño), pushed prices continually lower during a period of slightly more than 12 months. This fall was punctuated by a brief rally in June 1999 following the cessation of sales by Ghana. However, in the continued absence of manufacturer buying, the arrival of the West African main crop season saw prices revert to their previous downtrend. In London, the second position price ended 1999 at £571 per tonne, compared with £1,156 on May 15th, 1998.

NEW YORK AND LONDON TERMINAL MARKETS – 2nd POSITION 1993 - 1999



Source: E D & F Man Limited, Cocoa Market Report No.367

REVIEW OF PRODUCTION, CONSUMPTION, STOCKS AND PRICES - II

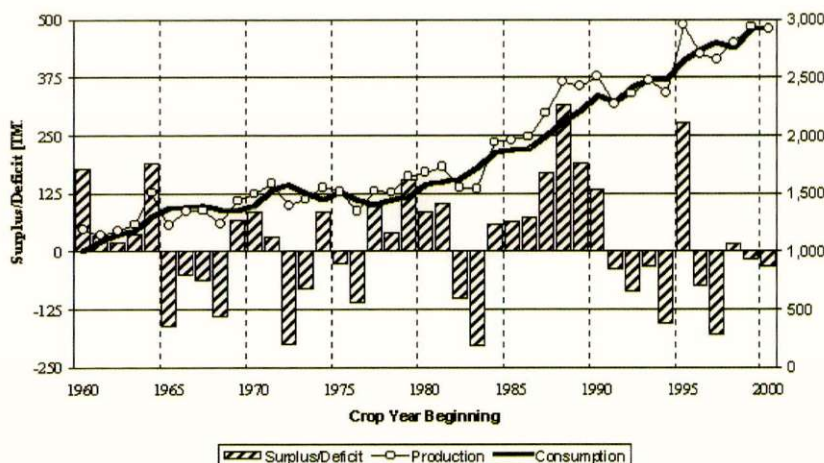
Mark N. Taylor
(January – November 2000)

PRODUCTION

E D & F Man Limited Cocoa Market Report No.367 reported that World production in 1999/00 would be 2,937 TMT ('000 tonnes), the second largest global crop on record and 4.8% higher than the 2,803 TMT produced in 1998/99. The La Niña weather pattern coincided with record crops in Côte d'Ivoire and Indonesia and the largest Ghana crop for 28 years. These increases in supply were more than enough to offset smaller crops in Nigeria (following the record crop in 1998/99) and Malaysia.

Production in West Africa was reported to have totalled more than two million tonnes for the first time. The 1999/2000 West African crop reached 2,064 TMT, 6.6% above the 1,936 TMT produced in 1998/99. This was a result of a production increase of 125 TMT in Côte d'Ivoire to 1.3 million tonnes and an increase of 42 TMT in Ghana to 440 TMT. Meanwhile, Ecuador completed its recovery from the devastating El Niño of 1997/98 to produce 93 TMT in 1999/2000, an increase of 21 TMT on the previous season. The Dominican Republic saw the start of a recovery in its crop, rising 11 TMT to 35 TMT, although this remains well below the level previously considered normal for the island. The 1999/2000 season also saw a further decline in production in Malaysia with output falling to just 50 TMT, the lowest level for 19 years.

WORLD COCOA SUPPLY AND DEMAND



Source: E D & F Man Limited, Cocoa Market Report No.367

E D & F Man Limited Cocoa Market Report No.367 forecasts that 2000/2001 global cocoa production will fall by a 0.6% to 2,920 TMT. Lower production in Côte d'Ivoire is expected to be partly off set by increases in Indonesia, Nigeria, Ghana and the Dominican Republic.

CONSUMPTION

E D & F Man Limited Cocoa Market Report No.367 anticipated that World cocoa consumption (as measured by World grind) would rise to a new record level of 2,924 TMT in 1999/2000, an increase of 6.0% on 1998/99. They noted that this increase in consumption was being led by demand for cocoa powder and, consequently, there had been a significant increase in cocoa butter stocks.

The Americas led the strong growth in grindings, rising by more than 10% during the year. Grindings in the USA rose by 32 TMT to a record level of 438 TMT. However, growth in chocolate consumption has been modest and, consequently, this appears to be the location where the largest increase in butter stocks has occurred. Grindings in Africa also grew at a healthy rate, increasing by 8.6%. This reflects the increase in origin processing, particularly in Côte d'Ivoire which is now the World's third largest grinding nation. Grindings in Europe saw a partial recovery following the sharp decline during the previous year. Grindings were reported to have risen by 3.2% during 1999/2000. A healthy recovery in East European demand was the key factor behind this increase.

STOCKS

E D & F Man Limited Cocoa Market Report No.367 indicated that for the 1999/2000 season there would be a deficit of 16 TMT. Its preliminary expectations are for a deficit in 2000/01 of 33 TMT. Global stocks are therefore expected to remain at around 1.2 million tonnes by the end of September 2001.

PRICES

The year 2000 saw the return of some semblance of stability to cocoa prices. The London second position price traded in a relatively narrow range between January and the end of November, reaching a low of £554 on February 1st and a high of £686 on September 18th. Prices had fallen back to the lows of the year by late November as the market awaited the bulk of the West African main crops.

WORLD COCOA PRODUCTION DATA

Reproduced from E D & F Man Limited Cocoa Market Report No.367
(September 2000)

	1996/ 1997**	1997/ 1998**	1998/ 1999	1999/ 2000	2000/ 2001 (i)
Cameroon	121 ¹	114 ⁻⁶	121	120	120
Côte d'Ivoire	1,130 ⁴	1,113 ⁻⁶²	1,175	1,300	1,225
Ghana	323 ⁻³	409 ⁹	398	440	450
Nigeria	157 ⁻⁵	165 ¹⁰	202	165	180
Others	39 ²	40 ⁸	40	39	39
Total Africa	1,770 ⁻¹	1,841 ⁻⁴¹	1,936	2,064	2,014
Brazil	170 ⁵	168	130	128	123
Colombia	45	46 ¹	39	40	40
Ecuador	101	28 ⁻²²	72	93	95
Mexico	35 ⁻⁸	30 ⁻¹³	25	22	30
Peru	18 ⁵	17 ⁴	15	16	16
Venezuela	15 ¹	14 ⁻¹	15	15	15
Others	12 ⁻²	11 ⁻²	11	11	11
Total C. & S. America	396 ¹	314 ⁻³³	307	325	330
Dominican Rep.	47	60 ⁸	24	35	45
Others	11	9 ⁻²	9	8	9
Total West Indies	58	69 ⁶	33	43	54
Indonesia	327 ¹⁷	331 ¹	393	396	420
Malaysia	102 ⁴	57 ⁻²⁸	79	50	45
Papua New Guinea	28 ²	26 ¹	35	39	37
Others	21 ³	20 ³	20	20	20
Total Asia & Oceania	478 ²⁶	434 ⁻²³	527	505	522
WORLD TOTAL	2,702 ²⁶	2,658 ⁻⁹¹	2,803	2,937	2,920
% change in production from previous season	-8.5 ^{-1.0}	-1.6 ^{-4.3}	+5.5	+4.8	-0.6

(i) Forecast.

** Changes since the last edition of the Cocoa Growers' Bulletin.

Note: All figures in Thousand Metric Tonnes (TMT) unless stated.
Some figures may not add due to rounding.

WORLD COCOA CONSUMPTION DATA

Reproduced from E D & F Man Limited Cocoa Market Report No.367
(September 2000)

	1995/ 1996**	1996/ 1997**	1997/ 1998	1998/ 1999	1999/ 2000(i)
France	111	106	107 ⁻³	124	140
Germany (Fed. Rep)	270 ⁻⁷	255 ⁻¹¹	240 ⁻³⁰	200	200
Italy	73	71	72	72	70
Netherlands	385	402	425 ¹⁵	415	437
United Kingdom	191	172 ⁻¹	174 ⁻¹	166	167
Others	158	164 ⁻⁴	176 ⁵	177	177
Total Western Europe	1,187⁻⁸	1,170⁻¹⁶	1,193⁻¹⁵	1,154	1,191
FSU/USSR	80	85	85	55	65
Other Eastern Europe	69 ⁻³	73 ⁻¹	68 ⁻⁷	61	56
Total Eastern Europe	149⁻³	158⁻¹	153⁻⁷	117	121
Côte d'Ivoire	135	150	205 ²⁵	221	245
Ghana	60 ⁻¹⁰	68 ⁻⁷	67 ⁻¹⁸	65	70
Others	48 ¹	56	51 ⁻¹⁰	62	63
Total Africa	243⁻⁹	273⁻⁸	323²	348	378
Brazil	183 ⁻⁹	180	188 ⁸	200	214
United States	345	394	399 ⁻⁹	406	438
Others	191 ⁻⁷	195 ⁻⁷	186 ⁻¹⁰	167	201
Total America	718⁻¹⁷	769⁻⁷	773⁻¹¹	773	853
Malaysia	96	103 ⁵	94	109	115
Indonesia	60 ⁵	72 ³	74 ⁵	80	76
Japan	49	53	43	47	49
Others	143	151 ⁵	157 ⁻²	129	140
Total Asia & Oceania	347⁴	379¹⁴	369⁵	366	380
WORLD TOTAL	2,645⁻³²	2,748⁸	2,811⁻³¹	2,758	2,924
% change in consumption from previous season	5.9 ^{-1.1}	3.9 ^{0.6}	2.3 ^{-0.4}	-1.9	6.0

(i) Forecast.

** Changes since the last edition of the Cocoa Growers' Bulletin.

Note: All figures in Thousand Metric Tonnes (TMT) unless stated.
Some figures may not add due to rounding.

WORLD COCOA STOCK DATA

Reproduced from E D & F Man Limited Cocoa Market Report No.367
(January 2000)

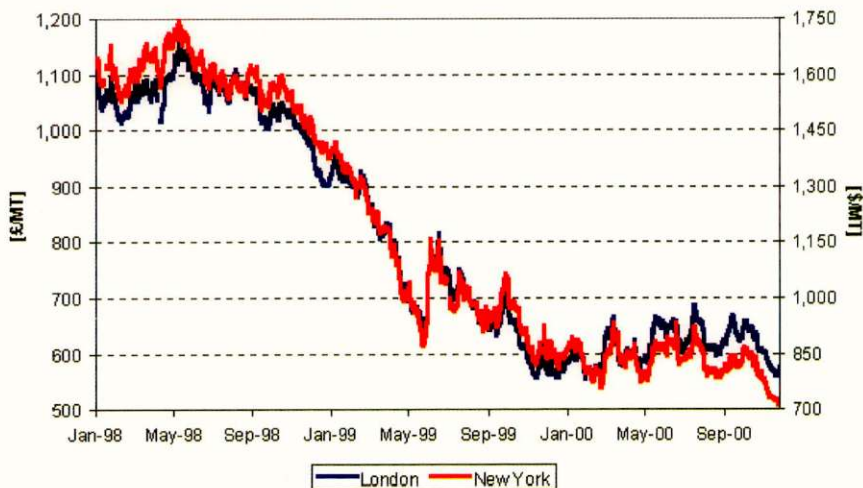
	1996/ 1997**	1997/ 1998**	1998/ 1999	1999/ 2000	2000/ 2001 (i)
Stock change	-73 ₄₄	-179 ₅₉	17	-16	-33
Closing stocks (including buffer stock)	1,373 ₂₂₆	1,194 ₁₆₇	1,211	1,195	1,162
Stock/consumption ratio (including buffer stock)	0.50 _{0.09}	0.43 _{0.07}	0.44	0.41	0.40

(i) Forecast.

** Changes since the last edition of the Cocoa Growers' Bulletin.

Note: All figures in Thousand Metric Tonnes (TMT) unless stated.
Some figures may not add due to rounding.

NEW YORK AND LONDON TERMINAL MARKETS – 2nd POSITION January 1998 – November 2000



Source: E D & F Man Limited, Cocoa Market Report No.367

THE DEVELOPMENT OF AN ALLEY CROPPING SYSTEM: A VIABLE AGROFORESTRY TECHNOLOGY FOR SUSTAINED COCOA PRODUCTION IN NIGERIA

Samson A. Ayanlaja¹

INTRODUCTION

The southern part of Nigeria is in the rainforest agro-ecological zone and contains nearly a million hectares of cocoa plantations, sited on oxisols, ultisols and alfisols which make up 40%, 16% and 5% respectively of tropical soils.

Many workers have been involved in determining the constraints and management problems of upland tropical soils in relation to sustainable crop production - including cocoa. Oxisols and ultisols have problems with acidity, aluminium toxicity, low nutrient reserves, nutrient imbalance and multiple nutrient deficiencies. Ultisols are also prone to erosion particularly on exposed sloping lands. Alfisols have major physical limitations as they are extremely susceptible to crusting, compaction, erosion and their low moisture retention capacity causes frequent moisture stress to crops. They also acidify readily under continuous cropping particularly when acidifying fertilisers are used (Kang and Wilson, 1987; Charreau, 1974; Juo, 1986). Under the present continuous production method, cocoa farms in Nigeria start a yield decline from the age of 10 years and in some cases earlier (Anon, 1963). Thus, cocoa production has declined from 310,176 tonnes in 1965 to only 133,338 tonnes in 1980 (Anon, 1980) and is now around 160,000 tonnes, despite increases in area, use of mineral fertiliser, insecticides, fungicides and introduction of a high yielding F₃ Amazon variety. The production decline has involved the premature death of trees on many farms with very low yields from surviving trees. In addition, replacement seedlings planted in the old cocoa soils have not readily established. Research has shown that these problems are caused by depletion of macro and micro nutrients, loss of organic matter and deterioration of soil physical properties (Omotosho, 1971; Are, 1970; Ayanlaja, 1983; Ayanlaja, 1987).

The future success of cocoa cultivation in Nigeria has to depend on the introduction of an ecologically sound farming system which can prevent the present soil degradation. The system must be able to upgrade the low C.E.C. kaolinitic soils by maintaining an adequate level of organic matter. It should involve low external inputs and be capable of preventing resource depletion and ultimately exhaustion of the inherently fragile tropical soils (Ofori, 1973; Kang and Juo, 1983; Watson, 1983; Nair, 1985).

AGROFORESTRY CONCEPT

The Basis for Sustainability of the System

In a traditional, ecologically stable shifting cultivation system, the soil nutrients are replenished during the fallow period by the regenerating ability of woody perennials and success depends on the length of the fallow period, the nature of the fallow vegetation, soil characteristics and climatic properties - especially temperature and the amount/distribution of rainfall. During the fallow period, plant nutrients are brought up from subsurface horizons by the deep roots of the perennials which are part of the fallow vegetation. These nutrients are ultimately returned to the soil surface through litter fall and decay.

¹ Ogun State University, P.M.B.2002, Ago-Iwoye, Nigeria.

The successful adoption of traditional shifting cultivation requires abundant land, but unfortunately supplies of land are now becoming limited in Nigeria due to the population pressure and the need to increase food production. Modern technology of the temperate regions, involving continuous cultivation with application of mineral fertiliser were found to result in rapid soil deterioration under tropical soil/climatic conditions. In any event in the case of cocoa in Nigeria, the case for fertiliser application is far from proven (Ayanlaja, 1996). It has been concluded that even with the available technology and input, it will still be impossible to sustain productivity of tropical soils (in the absence of substantial fertiliser applications) without incorporation of a "rest" period (Young and Wright, 1980).

The alley cropping concept is based on the higher efficiency of certain tree/shrub species in restoring soil fertility during short fallow periods. These species are encouraged during the fallow rather than just allowing natural vegetation with species which are relatively inefficient at soil rejuvenation.

Alley cropping effectively incorporates a rest or fallow period in the system, while still maximally utilising the land. Energy and nutrients are continually flowing back into the soil, through the fallow to prevent the depletion of the soil such that sustained productivity is achieved. In alley cropping systems, the target crop is planted between hedgerows of suitable shrubs or trees.

The appropriate tree/shrub for alley cropping must be capable of producing considerable quantities of biomass rich in nutrients - absorbed from weathering rock in the sub-surface horizons by its deep roots. When the alleys are pruned, large quantities of nutrient rich residues are added to the soil surface as mulch and green manure. Thus, the trees act as nutrient source, ensuring the regular injection of major and minor nutrients and their efficient cycling within the system.

The mulching material covering the soil surface also serves to de-energise raindrops and reduce or prevent runoff and erosion, as well as to suppress weeds, improve infiltration, aggregation, structure, porosity, water and nutrient retention capacities, earthworm activity and attenuate soil temperature. Furthermore, leguminous woody species add fixed nitrogen into the system, in addition to increasing soil organic matter.

Alley cropping makes possible a longer cropping period with increased land use intensity over time as the cropping and fallow phase are combined and energy flowing into the fallow component is directed towards crop production and soil restoration. Rapid and effective restoration of soil fertility is achieved with efficient fallow plant species. Soil productivity can be sustained, even without additional external inputs.

EXPERIMENTAL EVIDENCE SUPPORTING THE POTENTIAL OF ALLEY CROPPING AS A SUSTAINABLE FARMING SYSTEM

The potential for sustainability of an alley cropping system stems from the production of large quantities of nutrient rich biomass which is returned to the surface. The bigger the biomass yield and the richer the biomass is in nutrients, the better it is able to rejuvenate the soil and so sustain production. The choice of species planted in the alleys is all important.

Many agronomic experiments, with supporting tissue and soil analyses have been carried out in cocoa growing areas of the world (Brazil, Costa Rica, Côte d'Ivoire, Ghana and Nigeria) to screen multi-purpose trees, the so-called agroforestry trees for their suitability with cocoa in terms of climatic/edaphic factors and to monitor nutrient dynamics, soil physical properties, sustainability and the economic viability of alley cropping systems. Woody, leguminous and non-leguminous trees/shrubs found in association with cocoa or adaptable to cocoa soils include: *Cola nitida*, *Cordia alliodora*, *Erythrina poeppigiana*, *Gmelina arborea*, *Hevea brasiliensis* and *Leucaena leucocephala*.

Litter production, rate of decomposition, soil regenerative properties and suitability of the species are being assessed, but Table 1 shows the level of biomass production and the nutrient profile of the biomass for some potential agroforestry species; for example, *L.leucocephala* can add 246.5 kg N to the soil per annum. Omotosho (1975) calculated the amount of nutrients removed from soil in harvested cocoa components and found that 38.97 kg N, 6.29 kg P and 20.38 kg K were removed from soil in each tonne of harvested cocoa. Based on this information and the results of various fertiliser, fractionation, fixation and rate of N mineralisation experiments, Omotosho (1970; 1971) derived a fertiliser recommendation for cocoa in Nigeria as follows: 136 kg N, 70 kg P₂ O₅ and 58 kg K₂O/ha/annum.

Table 1. Biomass Production and Biomass Nutrient Content of Some Agroforestry Species

Species	Biomass Yield (tonne/ha/annum)	Biomass Nutrient Content (kg/ha/annum)				
		N	P	K	Ca	Mg
<i>Acioa barterii</i>	6.0	80.5	7.1	40.8	29.4	10.8
<i>Alchornea chornea</i>	8.0	168.8	12.8	96.8	83.2	16.0
<i>Erythrina poeppigiana</i>	5.0	174.9	8.8	53.6	163.3	53.7
<i>Cordia alliodora</i>	3.4	114.8	13.9	65.5	124.8	50.3
<i>Leucaena leucocephala</i>	15.4	246.5	34	235	142	48
<i>Flemingia macrophylla</i>	12.4	292	24	122	81	25
<i>Gliricida sepium</i>	10.5	333	20	176	140	42

Since *L.leucocephala* can add 246.5 kg N per ha/annum to the soil which is higher than the 136 kg N (ha/annum) recommended as a suitable rate of inorganic fertiliser on cocoa in Nigeria, it follows that alley cropping cocoa with appropriate tree species, would prevent soil depletion and so would more than sustain cocoa production. Nitrogen fixation by *L.leucocephala* in Nigeria was as much as 266 kg/ha/annum (Sanginga *et al.*, 1988). In Costa Rica, when the leguminous shade tree *E.poeppigiana*, planted in combination with cocoa was pruned 2 to 3 times per annum, it returned the same amount of nutrients to the litter layer that are applied to coffee plantations via inorganic fertiliser - even at the highest recommended rates for Costa Rica (i.e. 270 kg N, 60 kg P and 150 kg K/ha/annum). The annual nutrient return in this litter represents 90 to 100% of the nutrient store in the above ground biomass of the *E.poeppigiana* and hence gives no serious competition between the cocoa plants and the tree shade species (Beer, 1988).

In Bahia, Brazil, the yield of cocoa in combination with nitrogen fixing *E.poeppigiana* was 1,000 kg dry bean/ha, compared to cocoa yield of 650 kg dry bean under conventional monoculture. Similarly, in the rubber and cocoa mixture, the cocoa component yielded 645 kg dry bean and the rubber component 588 kg dry rubber/ha/annum (Alvim and Nair, 1986); consequently, tree/crop combinations should be preferred for their sustainable nature and several other advantages in the agricultural and socio-economic context of South East Bahia (Alvim and Nair, 1986). Fassbender *et. al.* (1988), Alpizar *et. al.* (1986), Imbach *et. al.* (1989) (all working in Costa Rica), found that it is possible for this system to sustain soil productivity and crop yield due to the level of organic matter recycling, the high amount of nutrient input into the soil and the prevention of leaching when cocoa is planted with *C.alliodora* and *E.poeppigiana*. Records of seven years of cocoa harvests under *C.alliodora* and *E.poeppigiana* in Costa Rica gave yields of 626 and 712 kg/ha/annum respectively. Harvests were found to gradually increase over the years and losses of nutrients (NPK, Ca and Mg) from the soil were very small due to the high rate of litter decomposition and soil nutrient enrichment from the 5.0 t/ha and 3.3 t/ha annual litter production from the *E.poeppigiana* and *C.alliodora* trees respectively. The timber tree *C.alliodora* reached a height of 14m and a d.b.h. (diameter at breast height) of 26.7cm. The additional advantage of wood production may make such crop/tree combinations preferable in Costa Rica (Heuvelodop *et. al.*, 1988).

In Nigeria, soil properties under monocrop *Cola nitida* were found to improve with time while the reverse was the case for soil under monocrop cocoa; a combination of kola and cocoa tree was therefore beneficial to cocoa (Ekanade, 1987). Prunings from *Acioa barkerii* in Nigerian climatic conditions decompose over a period of 30 weeks. Due to the persistence of the mulch cover on the soil surface, the mulch suppresses weeds, conserves soil moisture, prevents run-off and erosion while helping to maintain adequate soil organic matter (Anon, 1982). Similarly, for cocoa in Côte d'Ivoire, Budelman (1988; 1989), found nine tonnes of *Flemingia macrophylla* mulch survived for 14 weeks and effectively controlled weeds that multiply by seeds. He also found that one tonne dry pruning material of *Gliricidia sp.* would supply as much as 31.8 kg N, 2.4 kg P, 16.8 kg K, 13.4 kg Ca and 4.8 kg Mg to the soil.

In Trinidad, one tonne of the dry prunings of *Gliricidia sp.* was found to similarly add 38.2 kg N, 2.3 kg P, 28.0 kg K, 14.2 kg Ca and 4.6 kg Mg to the soil (Budelman, 1988). Since one tonne of dry cocoa removes 38.97 kg N, 6.29 kg P and 20.38 kg K in harvested cocoa components (Omotoshio, 1975), it follows that in both Côte d'Ivoire and Trinidad, the usual 5.5 tonne biomass yield per annum of *Gliricidia sp.* (Kang, 1981) would supply sufficient nutrients to compensate for nutrient removal from the soil by the cocoa crop, while preventing soil deterioration and leading to sustainable soil/cocoa productivity.

Mulching with *Gliricidia sp.* was found to increase soil moisture content particularly in dry spells. The mulch provide an average 3.9% increase in soil moisture compared to the bare soil situation; this additional moisture accounts for 37% of the available soil moisture. The estimated duration of the effect was up to 65 days. Average maximum soil temperature at 5cm depth was 4.6°C lower than in unmulched plots in Nigeria and this lasted 85 days (Budelman, 1989).

Soil water could be conserved by an alley cropping system and moisture stress which causes death and yield reduction in cocoa (Smith, 1964; Sale, 1970; Hutcheon *et. al.*, 1973) could be prevented (or greatly reduced) if cocoa was grown in alley cropping system. Metzner (1982) reported that alley cropping controlled erosion and improved productivity in the degraded sloping lands of Indonesia. Furthermore, addition of organic material and partial shading in an alley cropping system increased earthworm activity (Yamoah and Mulongoy, 1984).

Agroforestry species can potentially control weeds, add more nutrients to the soil than are removed by the accompanying crops, can increase soil organic matter, improve soil physical and moisture retention properties of the soil, prevent soil erosion and can thereby sustain crop/soil productivity.

ALLEY CROPPING SYSTEM OFFERS SOLUTIONS TO SOCIO-ECONOMIC CONSTRAINTS

The major socio-economic constraints and traditional customs contributing to low cocoa production in Nigeria were studied and enumerated by Ogunfeditimi (1980), Ajobo (1977), Berry (1975) and Olayemi (1970). The constraints identified by Ogunfeditimi are listed in Table 2 which demonstrates that social factors are important. These must be addressed if the results of agricultural research are to be implemented by farmers.

The possibility of an alley cropping system alleviating these problems can be considered under five sub-headings (see below), demonstrating that the application of a system of alley cropping can remove or at least mitigate them. Confirmatory factor analysis has been employed to measure the relationship between a dependant variable (low cocoa production) and a particular independent variable (constraint) when all the effects of other variables are removed.

1. Problems Emanating from Low Soil Fertility

The socio-economic problems of low crop yield, abandoning of cocoa farms in favour of other business, absence of planting, old age of cocoa trees and infrequent harvesting can all be solved by improving soil fertility and productivity. As previously discussed, increased and sustained soil productivity and the resultant higher crop yield can be a feature of an alley cropping system.

The Government of Nigeria, with 50% funding support from a World Bank loan in the 1970s established the Cocoa Development Unit to replant sub-economic cocoa farms with improved, high yielding, early maturing and virus tolerant cocoa varieties (Ajobo, 1977). However, a large part of the replanting operations was unsuccessful (Ogunfeditimi, 1980) as fast growing and vigorous seedlings did not, and cannot, establish in deteriorated old cocoa soils (Ayanlaja, 1983; Are, 1970). Adoption of the alley cropping system could have alleviated the soil fertility problem such that old cocoa trees could be successfully replaced with improved planting material.

Table 2. Significant* Socio-economic Factors Contributing to Low Cocoa Production in Nigeria

Factor Order	Constraints to Cocoa Production (x-factor)	Correlation Value $r_{x_1 - x_{15}}$
1.	Comparative advantage tendency, old age of cocoa trees.	0.84
2.	Land problems.	0.81
3.	Lack of labour supply, transport facilities and other necessary inputs.	0.79
4.	Lack of adequate credit facility, supervision and performance evaluation.	0.72
5.	Low and fluctuating income achieved by cocoa producer and lack of incentives.	0.53
6.	"Get rich quick" euphoria of the society prompting cocoa producer to change occupation and abandon cocoa farming.	0.53
7.	Acts of God (i.e. drought) and pest problems.	0.48
8.	Inadequate extension services.	0.45
9.	Inadequacy or absence of system of provision of replacement cocoa planting material and crude nurseries.	0.44
10.	Instability in the price of cocoa and the role of the middlemen and the Government.	0.42
11.	Increasing age profile of cocoa producers.	0.39
12.	Attitude of cocoa producers to adoption of new varieties, illiteracy and contentment with traditional ways.	0.27
13.	Low or non-crop utilisation, infrequent harvesting and crude method of pod removal from the cocoa tree.	0.25
14.	Inadequacy or absence of intermediate technology such as artificial driers when and where necessary.	0.23
15.	Bad timing of spraying and inadequate knowledge of chemical dosage requirement.	0.21

* Significance at 0.01 level.
 $r_{x_1 - x_{15}}$ = low cocoa production; $x_1 - x_{15}$ = constraints; N = 300.

Source: Ogunfiditimi, 1980.

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2. Problems Relating to High Operational and Maintenance Costs

High labour and chemical costs for weeding, pest and disease control, high cost and scarcity of fertilisers, lack of knowledge of chemical dosages and timing of herbicide applications can be solved under the alley cropping system.

As previously discussed, active mulching and nutrient recycling are features of an alley cropping system. Mulching suppresses weeds (Budelman, 1989) and increases soil fertility (Kang and Wilson, 1987) such that labour cost on weeding and the need to purchase mineral fertiliser and herbicides are drastically reduced; such savings on weeding/fertilising would increase overall profit. Nwonwu (1987) studied costs and benefits of three labour alternatives and found a 47% saving in cost in the taungya system over other alternatives. Decomposing leaf mulch of agroforestry (hedgerow) species like neem (*Azadirachta indica*) has been known to produce extracts having insecticidal and nematocidal properties and has been used for plant protection purposes (Puginier, 1990). A weed has been identified and grown with maize in Asia because its roots give off a compound lethal to a fungus. Odours emanating from some plant species have been observed to repel insects. Selection and incorporation of similar species capable of trapping and killing insects and fungal spores into cocoa agroforestry systems would provide an inexpensive, environmentally non-polluting, cultural solution to pest and disease problems. These possibilities merit further study.

Major diseases (Black pod disease caused by *Phytophthora palmivora* and *Phytophthora megakarya* in Nigeria) or Swollen Shoot Virus disease transmitted by mealybugs might be more effectively controlled culturally at a lower cost under an alley cropping system. Active mulching prevents rain drop splash which is sometimes the process by which *Phytophthora* spores are transported from the soil onto healthy cocoa pods (Gregory, 1981). Alley cropping with appropriate hedgerow species could perhaps help control diseases and reduce cost of labour and the need for chemicals.

3. Problems Emanating from the Custom and Attitude of Farmers

Soil problems such as negative attitude of farmers to new innovations, traditional contentment, act of God (drought) and illiteracy, could be reduced under the alley cropping system.

Farmers in Nigeria have developed negative attitudes as a result of frustration and financial embarrassment that ensued from previous trials of new innovations (chemicals and varieties) introduced to them but which failed to perform as expected. Farmers therefore prefer to be content and farm their land in the traditional way rather than expend their scarce resources on "assumed" productive devices (Lucas, 1980; Ajobo, 1980; Olayemi, 1970).

However, alley cropping is not a new innovation, since traditional West African cocoa cultivation is already an agroforestry system in which cocoa is grown between thinned jungle trees and food crops and farmers raise small ruminant animals (sheep and goats) in their farmsteads (Olayemi, 1970; Ekande, 1987; Okafor and Fernandes, 1987; Adejuwon and Ekande, 1988; Atta-Krah and Sumberg, 1988).

Farmers would probably not be sceptical about adopting the alley cropping system for cocoa since it is only a modification or improvement of the already existing agroforestry system, involving active mulching, instead of passive leaf fall and is not a new or "glorified"

innovation. The alley cropping system therefore fits into the custom and practice of farmers and the system should be readily acceptable especially because of its additional advantage of supplying protein-rich fodder for their ruminant animals and ensuring high yield of the cocoa - food crop - animal production system.

According to Budelman (1989), mulching with *Gliricidia* sp. increases soil moisture content significantly and the estimated duration of the effect was up to 65 days. Adoption of an alley cropping system would mitigate or completely remove soil moisture problems associated with droughts (termed "acts of God" by farmers) since well mulched soil would still have sufficient moisture for over two months without rain.

4. Problem of Lack of Infrastructure

Socio-economic problems that stem from absence of infrastructure include inadequate or lack of supply or complete absence of technological devices like artificial dryers and harvesting equipment. Such areas also have poor road network, inadequate provision of transport facilities and show rural-urban migration of young farm labour.

Provision of infrastructure by Government for a rural area is based on the total quantity of farm produce from that area. Government builds a road network, for example, to connect a rural area to the city if the quantities of farm produce from that area justify this investment.

Adoption of alley cropping system, as previously discussed, would improve soil fertility, soil water content and so increases and sustains total farm produce from that area. This could attract Government attention to build and provide road, pipe water, electricity, schools, industries and markets in that locality. The availability of such infrastructure could help arrest the rural-urban migration of young people in search of schools and jobs and so they would become available to participate in the rewarding cocoa cultivation.

5. Problems of Finance for Cocoa Cultivation

Socio-economic problems including lack of credit facilities, incentives and the fluctuating income now expected from cocoa production can be mitigated by adoption of alley cropping system.

Low incomes from cocoa production is partly a result of expenses incurred on inputs (fertiliser and herbicides etc.), which erode away the net profit on the enterprise. Adoption of the alley cropping system would drastically reduce spending on chemical and fertilisers, and net profit would increase. The need to borrow or the amount of loans received from financial institutions (with interest charge) to purchase chemicals would reduce and overall profit would increase.

Therefore with the adoption of the alley cropping system, some major technical and socio-economic constraints could be mitigated and a boost in cocoa production should follow.

DEVELOPMENT OF SUITABLE ALLEY CROPPING SYSTEM FOR COCOA

Soil chemical and physical deterioration results in drastic yield reduction of cocoa in Nigeria.

As observed by many workers, the major cause of tree death was low soil moisture especially during the long dry season (Wessel, 1969; Ali, 1969; Babalola, 1972; Falade, 1975; Ayanlaja, 1987; Ayanlaja, 1996) and continuous cultivation of cocoa results in gradual depletion of soil organic matter and nutrients with resultant deterioration and yield reduction (Omotosho, 1975; Are, 1970; Ayanlaja, 1983). Mineral fertiliser application has not solved the problem but has actually been found to aggravate it (Egbe and Omotosho, 1971). All these observations confirm that the farming system under which cocoa is currently cultivated in Nigeria (and probably elsewhere too) needs to be modified to prevent the decline in soil productivity, degradation of the environment and needs to become more sustainable. A land management system with a longer cropping period and increased land use intensity, with improved and sustained cocoa productivity, resulting from rapid and effective soil fertility regeneration needs to be developed.

Alley cropping is an agroforestry system which increases overall yield of the land by combining production of crops and forest trees simultaneously. It takes advantage of the biological, environmental and economic benefits of specific tree species rather than relying constantly on expensive agricultural inputs. Research has shown that the system is highly stable and sustainable. Trees act both as a nutrient pump (bringing up nutrients newly released from weathering rock to the surface) and as nutrient source ensuring a constant injection of major and minor nutrients and their effective recycling within the system. The trees, especially legumes, provide mulch which increases organic matter for soil fertility maintenance (Kang and Wilson, 1987). Suitable tree species will improve soil nutrients by nitrogen fixation, erosion control and maintenance of high level of soil organic matter and thus soil structure; in addition, aggregation and water retention properties are improved (Sanginga *et al.*, 1988; Nair, 1985; Kang and Wilson, 1987).

The development of an alley cropping system for cocoa production will require research to screen potential agroforestry trees to select the most suitable species to be combined with cocoa for a given ecosystem and devise management practices for the establishment of the alley cropping system. Integration of such technology into cocoa production systems could increase and sustain production and arrest environmental degradation, even in the face of present forest exploitation (Anon, 1979).

Experiments should be designed to identify optimum spacing for tree and cocoa plants in the combination and assess the effect of the combination on the microclimate. Nutrient recycling, soil water balance and nitrogen fixation under the specific cocoa/forest tree combination should be evaluated. Pruning regimes, designed to optimise biomass production, nutrient profile of prunings, rate of decomposition and release of nutrient by the prunings, all need to be devised.

Availability of this information with knowledge of the specific socio-economic constraints in the given locality, as well as food habits, norms, customs of farmers as they affect cocoa production, would lead to development of an appropriate alley cropping systems for cocoa under which soil/cocoa productivity would be sustained.

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EFFECTIVE MANAGEMENT OF COCOA FARMS IN NIGERIA FOR PROFITABILITY

A.A.Adeyemi²

INTRODUCTION

Cocoa (*Theobroma cacao*) has very high yield potential. Yield as high as 200 pods per tree per annum has been recorded from some F3 Amazon selections in Nigeria (Atanda, 1971; Jacob and Olaniran, 1971). This figure, using Are *et. al.* (1971) conversion ratio of 24 pods to 1 kg dry beans gives about 8 tonnes per hectare of dry beans. In Ghana, Glendinning (1962) also investigating the yield of cocoa reported that a dry bean yield up to about 3.3 tonnes per hectare was realised from some Trinidad introductions. In spite of this good characteristics of cocoa even when the soil is suitable, as described by Smyth and Montgomery (1962), Smith (1966), Egbe and Omotoso (1971) and Egbe *et. al.* (1989), effective management of the farm must still be carried out before good yield can be realised. Lack of proper maintenance of cocoa farms has always resulted in etiolation and poor general performance of cocoa trees leading to high incidence of pests and diseases and low yield. This paper attempts to review the current management practices in cocoa in Nigeria that can lead to high yield and thus make the agricultural venture a profitable and more sustainable one.

ROUTINE MAINTENANCE OF COCOA FARMS

Optimum environmental and edaphic factors (air/oxygen, light, moisture, soil nutrients) as needed for proper growth and yield of cocoa and to reduce incidence of pests and diseases on cocoa farms in Nigeria are needed. Mulching, shade management, weed control, pruning, control of epiphytes and parasites and the establishment of fire traces are the crop husbandry practices recommended for the effective routine maintenance of cocoa farms in Nigeria (Are *et. al.*, 1971; Odegbare, 1977; Adeyemi, 1992b, 1992c, 1993a; Adeyemi *et. al.*, 1992).

Mulching

Mulching is a cultural practice carried out in the early years of cocoa establishment to conserve moisture in the soil during the dry season (November to March). Mulch acts as shade, protecting the soil surface from direct sunlight and is important in the establishment, growth and yield of cocoa in Nigeria (Egbe and Omotoso, 1971; Omotoso, 1973). Dry plantain/banana leaves, grasses and other leafy plant organs are used. The mulch, about 7.5 to 15.0cm thick, is arranged in circular form around the individual cocoa trees ensuring clearance of about 7.5cm radius between the edge of the mulch and the cocoa tree to prevent termite infestation. In addition to soil moisture preservation, mulch can also serve as organic fertiliser after decomposition.

Shade Management

In Nigeria, cocoa requires shade only in the early establishment years (3 to 4 years after planting), when the root system has not fully developed and the tree is unable to tap moisture from any depth of soil. Provision of temporary shade, either natural or planted, reduces water

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loss of from the leaves as well as from the soil surface during the dry period. Young cocoa trees in Nigeria have stunted growth, chlorotic leaves and poor survival rates when grown under no shade conditions (Longworth, 1963; Are et. al., 1971).

Plantain, banana, tree cassava, pawpaw and palm fronds can be used as temporary shade in cocoa but plantain is preferred. It is relatively cheap to establish, does not compete with cocoa for nutrients and water and is very economical, while serving as a source of income to farmers until the cocoa trees come in to bearing. Plantain at 3m x 3m or one plant per cocoa tree, gives a population of 1,111 plantain trees per hectare. Because of its high rate of multiplication, the population can be as high as five times the initial number, if the suckers are not efficiently and regularly managed. Two young suckers on each adult plantain should be retained to replace the adult plantain after harvest, thus avoiding over-shading. Ideally, plantains should be planted a year before the cocoa, but most often it is planted in the same year as the cocoa. Where the two crops are planted in the same year, plantain is established with the early rains in March/April to allow the plantain to form some shade before cocoa is transplanted to the field in June/July. The population of plantain diminishes with the age of the farm and about 5 to 8 years after planting, it may have almost disappeared due to natural shading (Adeyemi, 1991b).

Where the farm has been initially established through selective thinning of forest, reduction of the forest shade is done gradually by killing the trees with tree killers or by girdling, to prevent over-shading and high incidence of diseases that could occur from the high humidity conditions of the farm. Improved aeration from the reduction of shade leads to increased yield of cocoa. Increased light intensity increases yield, as dry matter accumulation in cocoa leaves increases as the light intensity increases (Omotoso, 1973). Shade is not necessary for mature cocoa under Nigerian conditions, as the roots are well developed and are capable of exploring deeper zones of soil for moisture and nutrients and the canopy at this stage is closed enough to provide adequate shade to minimise moisture loss from the soil.

Weed Control

Weeds are one of the greatest biological problems of cocoa farms in Nigeria. They compete with cocoa for water and nutrients thus hindering its growth, performance and yield. In some cases the young seedlings are completely smothered by weeds, resulting in poor establishment and they also serve as reservoirs for diseases and pests, including squirrels and rodents that devour cocoa pods. Weed infestation is a more serious problem in young cocoa with unclosed canopy during the first 3 to 5 years of establishment, than in mature cocoa with closed canopy. The complete canopy suppresses weed growth, though weeds are a problem in mature cocoa where the canopy is damaged due to leaf abscission (tree defoliation) in senescent cocoa, or when trees are missing due to death, poor establishment or dieback caused by mirids. Weeds can also be a problem where the weed species are shade tolerant such as with *Axonopus compressus*, *Cynthula prostata*, *Asystasia gigantea* which can even grow under a heavy cocoa canopy (Adeyemi, 1991b, 1992a).

Several weed species are important in cocoa farms in Nigeria (Komolafe, 1976; Oladokun and Utulu, 1988; Adeyemi, 1988b, 1992a). They include the species above and *Panicum spp.*, *Setaria spp.*, *Chromolaena odorata*, *Ageratum conyzoides*, *Acalypha ciliata*, *Euphorbia spp.*, *Fluerya aestuans*, *Pouzolzia guineensis*, *Amaranthus spinosus* and *Sida spp.* Weeds are controlled by manual, cultural methods or with chemicals or a combination of these.

Manual Weed Control

Manual weed control is effected by slashing the weeds with sharp strong cutlass. It is initially effective but weed re-growth is very fast, necessitating about 3 to 6 rounds of weeding per annum depending on the ecology and the type of weeds of the area (Adeyemi, 1984a, 1986a, 1992a; Adeyemi and Adenikinju, 1984). Weeds are not removed from the farm but are left as organic mulch to decompose. Hoeing is not encouraged in cocoa farms because of the damage that may be done to the roots of cocoa.

Cultural Weed Control

Mulching of young cocoa farms (i.e. cocoa within 3 to 4 years of establishment) has been found to suppress weeds, conserve soil moisture and improve the vigour and yield of cocoa (Longworth, 1963; Are *et al.*, 1971). Intercropping is popular among peasant farmers of Nigeria especially in the South Western part of the country, using cassava, cocoyam, yam, maize, melon, okra and cowpea cultured with cocoa in different combinations. Cocoa/plantain/yam/maize/cowpea, cocoa/plantain/cassava/melon and cocoa/plantain/cocoyam/okra/melon intercrops have been proved to cause no harm to the cocoa and are also effective in reducing weed incidence (Adeyemi, 1983, 1984a, 1986b, 1988c, 1989). The number of weeding rounds is lower in intercropped plots than when cocoa is grown in pure stands (Adeyemi, 1988b). Maize and cowpea have also been successfully intercropped in rehabilitation plots (Adeyemi, 1984b).

Chemical Weed Control

Initially, only paraquat (at 0.60 kg a.i/ha) was recommended for effective weed control (Freeman and Ashiru, 1966). Recent work has shown that weeds can be effectively controlled by Gramuron (Paraquat and diuron) at 2.0 kg a.i/ha, by glyphosate (1.92 kg/ha) and by tank mixture of Asulam plus Actril DS (ioxynil +2, 4-D) at 3.40 + 1.05 kg/ha (Adeyemi and Adenikinju, 1984, 1988a, 1991a, 1992a). Each of the herbicide rates is diluted with water at the rate of 300 litres per hectare and applied with 4-Oak knapsack sprayer at the pressure of 5.45 kg/cm². These rates are further simplified to 90, 150, 120 and 255 + 75 ml of paraquat, Gramuron, glyphosate and asulam + Actril DS respectively in 9 litres of water for farmers whose farm is less than one hectare. Although the four herbicide formulations are effective in controlling weeds in cocoa, the weed re-growth period in paraquat treated plots is significantly shorter than that of other herbicides (Adeyemi, 1988a). This implies that paraquat has no lasting effect on weeds. No phytotoxicity has been observed on cocoa either young or mature after careful application of these herbicides (Adeyemi, 1991a, 1993b). Paraquat was the most popular herbicide due to its relative cheapness and immediate apparent action on weeds (Adeyemi, 1984c).

Pruning

Pruning is a routine practice in cocoa and carried out to control the height, to give the desired shape and to ventilate farms to reduce disease and pest incidence. All unnecessary branches including chupons, excessive and dry fan-branches are removed with sharp cutlasses, secateurs, pruning saws and shears. To allow for easy operational movements in the farm, jorquetting of cocoa trees from 1.2m upwards is encouraged. Where this occurs

at lower height, branches are removed by pruning which allows further growth of the main stem before the apical stem is removed to give fan-branches.

Light pruning is done any time of the year while heavy pruning is carried out during the rainy season (April to October). All cut surfaces should be painted with a wound dressing paint to prevent entry of rain and pathogens. Pruning is an accepted practice in Nigeria and the frequency of carrying out the operation depends on the age and the growth pattern of cocoa. One main pruning per annum is adequate but excess chupons may be removed 3 to 4 times in a year when cocoa growth is vigorous.

Control of Epiphytes and Parasites

Poorly maintained cocoa farms are often infested with parasites such as mistletoes (*Tapinanthus spp.* and *Euglerina gabonensis*) and epiphytes including bryophytes, liverworts, mosses and lichens. A high level of mistletoe infestation usually results in the degeneration of cocoa trees and high yield loss, because they are parasitic and compete with the host plant for food and nutrients. Epiphytes even though they do not derive their food from their hosts also cover the trunks of cocoa including the flower cushions with their flat or mat-like bodies leading to inhibition of flower production and perhaps enhanced levels of pod attack by *Phytophthora spp.* as they raise the humidity levels. Mistletoes in Nigeria are best controlled by physical removal with sharp tools such as cutlasses, axes and pruning shears (if available). Effective control is achieved when the removal is done at the time of the flowering of the parasites. Epiphytes on the other hand are controlled culturally through prompt removal of unwanted chupons, fan-branches and excess overhead shade. This improves the ventilation and reduces the damping-off condition in the farms.

Fire Trace

Cleared paths of minimum width of three metres should be constructed round the farm to forestall the spread of any bush fire during the dry season. Entry of animal pests into the plantations is also reduced by these paths and so it is useful to keep them cleared throughout the year.

Soil Fertility Maintenance in Cocoa Farms

Under normal conditions, cocoa grown on good soil in Nigeria does not need any nutrient supplementation at the juvenile state (Egbe and Omotoso, 1971; Omotoso, 1973). The available nutrients in suitable cocoa soils are enough to support good growth and yield of cocoa up to 12/20 years for Amazon cocoa and longer than this for Amelonado (Omotoso, 1975; Ojeniyi, 1981). Nitrogen and phosphorus have been shown to be essential for the increased yield of cocoa at the mature stage while significant response has not been shown to K application. Consequently high rates of N and P (337 g each of urea and single superphosphate) and a low rate of K (87 g KCL) per tree are applied to mature Amazon cocoa. For Amelonado cocoa, 275 g urea, 225 g SSP and 36 g KCL per tree are used for soil nutrient supplementation. When expressed in elemental forms the fertiliser requirements of cocoa may range from 120 to 240 kg N, 30 to 140 kg P and 58 to 67 kg K per hectare per annum depending on the age and type of cocoa. Application of about

6g of Boron per tree per annum would also result in increased yield of cocoa. For young cocoa grown on deficient forest soils only phosphorus in the form of single superphosphate at 135 to 270 g per cocoa tree needs to be applied to correct the nutrient imbalance.

DISEASES AND PESTS

Diseases

Phytophthora pod rot (black pod) disease is of real economic importance in Nigeria. Cocoa Swollen Shoot Virus (CSSV) is also present but scarcely causes an economic loss.

Black Pod (Phytophthora pod rot)

Two forms of the fungus - *Ppalmivora* and *Pmegakarya* have now been identified as the causal agents of black pod disease in Nigeria (Sansome *et al.*, 1975) contrary to the initial belief that the disease is caused by *Ppalmivora* only (Gorenz and Okaisabor, 1971). Black pod is the most serious disease of cocoa in Nigeria causing an annual bean loss of about 30 to 35% (Gorenz and Okaisabor, 1971). Yield loss can range from 5% in the dry areas to 80% in the wet parts of the country (Odegbaro, 1977) depending on rainfall and relative humidity.

The pathogens can affect all cocoa plant parts, but it is of most marked effect on the pods. It is first observed as a brown spot or discoloration on the pod. The spot spread rapidly under favourable conditions covering the pod surface within a week or two of its first appearance. In the course of infection, white patches of fungal hyphae coupled with masses of sporangia are formed on the lesions. At the advanced stage of infection, the pathogen penetrates into the centre of the pod and renders the seeds useless (Gorenz and Okaisabor, 1971). The main vehicle of infection is rain splash followed by insect, air currents and vertebrate pests while the sources of inoculum are soil, flower cushions, dry pods from the previous season either on the cocoa trees or on the ground and debris formed from heaping of cocoa pod husks from previous harvests.

The disease is controlled through cultural practices such as farm sanitation through weeding, pruning of chupons and unwanted fan-branches and prompt removal of shade as earlier described to improve the ventilation of the farm and consequently reduce the relative humidity. Application of copper-based fungicides (Bordeaux mixture, Perenox, Kocide 101 and Copper Nordox) at three weekly intervals commencing from the onset of the rains is effective in controlling black pod disease of cocoa in Nigeria. Their active ingredients and rates are given in Table 1.

However, a new fungicide - Ridomil plus 52 W.P. whose active ingredients are cuprous oxide and metalaxyl is presently being studied. In general, the effectiveness of all the fungicides are influenced by the variety of cocoa and location in terms of its climatic conditions (Adegbola and Filani, 1985; Adegbola, 1986). Research in Nigeria to produce resistant/tolerant and escape varieties of cocoa that would be able to resist and avoid the disease respectively is underway.

Table 1. The Composition and Rate of Application of Some Fungicides used in Controlling Black Pod Disease in Nigeria

Trade Name	Common Name / Active Ingredient	Rate (g/9 litres of water)
Lime Bordeaux mixture	Copper sulphate and lime	90.0 + 36.0
Perenox	Cuprous oxide	40.0
Kocide 101	Copper hydroxide	40.0
Copper Nordox	Cuprous oxide	45.0
Brestan	Triphenyltin acetate	13.3

Cocoa Swollen Shoot Virus (CSSV)

The disease is caused by the virus called Cocoa Swollen Shoot Virus (CSSV) and there are many strains of the CSSV in Nigeria named after the localities from where they were isolated (Thresh and Tinsley, 1959; Thresh, 1961). They include Ikire (Egbeda), Offa-Igbo, Ilaro, Abuku, Balogun, Alaparun, Olanla, Ilesa and Ife etc. The symptoms vary according to the strains but range from swellings on stems and roots to leaf chlorosis/necrosis (leaf vein clearing). The attack of CSSV can also lead to complete defoliation and dieback of cocoa trees in more virulent strains. The Ikire isolate is the most virulent though most Nigeria isolates only cause modest swellings (Adegbola, 1971). The Ikire isolate is implicated in the death of young seedlings. The disease is transmitted by mealybugs. Initially, it was controlled by prompt removal of all the cocoa trees around the point of infection up to the radius of 30m away but this has since been stopped due to the low virulence. CSSV is now at a low level with the replacement of the infected cocoa trees with tolerant/resistant varieties, either by complete farm replanting, planting of the improved varieties under the shade of old cocoa or through coppicing of the old cocoa trees. Removal of wild hosts through the reduction of specific shade trees where cocoa is grown under forest trees also assists in reducing the sources of inoculum and the incidence of the disease.

Insect Pests of Cocoa

Mirids (Capsids), are economically the most important pest though some damage is also caused by *Characoma stictigrapta*, *Earias biplaga* and the mealybugs (CSSV vectors) (Adeyemi *et al.*, 1992). Estimated annual losses due to the attack of cocoa by these insects vary between 2 and 25%.

Mirids

Sahlbergella singularis Hagl. (the light mirid), *Distantiella theobroma* Dist. (the dark brown mirid) and *Helopeltis* spp. (the "Cocoa mosquito") are the three major species of mirids in Nigeria. They are shoot feeders that feed mainly on the plant sap of cocoa through their piercing and sucking mouthparts. Damage from mirids are permanent wilting of growing points and leaf flushes, defoliation of leaves giving a staghead appearance to the cocoa trees and the eventual die-back of the trees. The dieback may be further aggravated by infection with a fungus - *Calonectria rigidiscula* whose entry is through the wounds created by the mirids. Yellowing and permanent wilting of cherelles and the appearance of punctures on the

surface of mature pods in mirid infested plantations can also cause losses. It is estimated that up to 15/25% of cocoa trees are lost annually to mirid infestation in Nigeria (Adeyemi *et. al.*, 1992).

Mirid infestation in cocoa farms in Nigeria is reduced by cultural practices and chemical control. Cultural operations involve the pruning of infested branches, regular removal of chupons and infested young cocoa pods to deprive the mirids of suitable feeding and breeding sites. Good establishment of cocoa farms to form a closed canopy and preventing gaps (capsid pockets) through which mirids can enter the farm has always helped in minimising mirid infestation.

Mirid infestation in Nigeria has been historically controlled through the application of contact insecticides. These include Gammalin 20 E.C. and Uden 20 E.C. each applied at 113 mls in 9 litres of water, Mipcin 75 W.P., at 3.3 g a.i./litre of water, Dursban 4 E.C. at 48 mls per 9 litres of water and Elocron 75 and 50 W.P. applied at 0.25% concentration. They are applied with knapsack sprayers or motorised knapsack sprayers of the "Motoblo" type or with Swing-fog machines. Best results are obtained when the applications are done in August, September and October every year. But using an Integrated Pest Management (IPM) approach, it is advisable to only spray chemicals to control mirids when their number is at economic threshold so as to minimise the quantity of insecticides used (Idowu, 1993 *pers. comm.*). Biological control of mirids through the use of parasites such as braconidae and mamilionides is still under research in Nigeria. Alternative management systems for capsid control are needed.

Characoma stictigrapta

This is a moth with pod boring larvae that attacks the cocoa cherelles and pods (Ojo, 1980). Annual pod loss due to its attack varies between 10 and 15% (Adeyemi *et. al.*, 1992). The insect, especially the larvae, which causes the damage to pods can be controlled by the application of Gammalin 20 E.C. and Elocron 50 E.P. at 0.25% concentration.

Earias biplaga

The insect is a moth and it is the larva, a spiny boll worm that causes damage to buds and young leaves of cocoa. It is very prevalent in young unshaded cocoa farms. The annual loss in Nigeria due to its infestation is about 5 to 10% (Adeyemi *et. al.*, 1992). The cultural practices described for mirids can also effectively control this pest as can the application of insecticides.

Planococcoides njalensis

Cocoa mealybugs are very important in Nigeria especially in areas of high infection for Cocoa Swollen Shoot Virus disease (CSSV) because they act as the vectors of the disease. In addition, mealybugs also feed on plant sap, as do mirids. Effective control through chemical sprays is difficult to achieve since the mealybugs are protected by the waxy coverings. They can be controlled indirectly by controlling the ants with which each species is associated.

Table 2. Annual Cost of Maintaining One Hectare of a Mature Cocoa Farm in Nigeria

(a) Cost of Labour for the Maintenance Operations

Operations	Man-day	Unit Cost		Total Amount	
		N	\$	N	\$
Spraying against termites	3	60	2.93	180	8.78
Gapping up and shade management	8	60	2.93	480	23.41
Weeding	45	60	2.93	2,700	131.11
Removal of chupons, excessive and dead branches	5	60	2.93	300	14.63
Spraying against mirids	10	60	2.93	600	29.20
Application of fungicides	25	60	2.93	1,500	73.80
Harvesting and processing	45	60	2.93	2,700	131.00
Cutting of fire traces	5	60	2.93	300	14.63
Removal of mistletoes (parasites)	10.33	60	2.93	200	9.76
Total	149.2	—	—	8,960	437.07

(b) Cost of Inputs for the Maintenance Operations

Items of Inputs	Quantity Required	Unit Cost	Total Amount	
		N	N	\$
Wheel barrows	2(1/5)	1,000	400	19.51
Aldrex 40 (litre)	2	100	200	9.76
Copper sulphate (kg)	30	65	1,950	9.51
Insecticide 20 (litre)	4.5	160	720	35.12
Hydrated lime (kg)	15	35	525	25.61
Fertiliser N P K (15-15-15) kg	400	1.6	640	31.22
Cutlasses	2(1/2)	85	85	4.14
CP3 Spraying pump	1/5	1,500	300	14.65
Water drum	1/5	350	70	3.41
Strainer	(2)1/5	10	4	0.20
Pair of gloves	2(1/5)	50	20	0.98
Apron/overall coat	2(1/3)	110	73	3.58
Iron file	1(1/3)	30	10	0.49
Water (litre)	500	0.1	50	2.44
Baskets	510	5	25	1.22
Harvesting knife	(4)1/5	10	8	0.40
Fermenting trays	(5)1/5	75	75	3.66
Plastic bucket	5	10	50	2.445
Drying slab	1/10	7,000	700	34.15
Incidental expenses			5,000	243.90
Total	—	—	10,905	446.40

Termites

These are important group of insects commonly found in newly established cocoa farms in Nigeria. They feed on the roots and barks of young cocoa trees. They are controlled culturally by intercropping young cocoa with dense-canopy, farming arable crops such as cocoyam and cassava that form additional shade and also act as baffles to the insects (Idowu, 1993). Application of Aldrex 40 E.C. at 1% concentration to the soil around an attacked cocoa tree is very effective in controlling termites in cocoa farms.

PROFITABILITY OF COCOA PRODUCTION IN NIGERIA

With the yield potentials of cocoa given at 4 to 8 tonnes per annum, actual yield of up to 2.0/2.5 tonnes can be expected when all the maintenance operations and pest control measures as described in this paper are effectively carried out and weather is favourable. Under the present economic situation in the country, cost of such operations in terms of labour and material inputs is as high as N18,110 or \$883.47 (Table 2). The revenue from the farms under the present management practices based on the given yield is however, N40,000 to 50,000 per hectare. Consequently, a net annual profit range of about N21,889.67 to 31,889.67 (\$1,951.22 to 2,439.02) per hectare can be realised by the farmer. In economic terms when the cash flow of the project in consideration to cost and income (benefit) are compared, a benefit/cost ratio (BCR) of 2.21 to 2.76 (Table 3) is obtained indicating that the agricultural venture is profitable.

Table 3. Cost/benefit Analysis from the Cash Flow on the Maintenance, Harvesting Fermentation and Drying of Cocoa in Nigeria

Variables	t/ha	Rate	Revenue		BCR
		N	N	\$	
Yield	2.0-2.5	20,000	40,000-N50,000	1,951-\$2,439	—
Total Cost of Production			18,110.33		
Benefit/Cost Ratio (B.C.R.)			—	—	2.21-2.76

One can therefore assume that the cultivation of cocoa in a humid tropical environment like that of Nigeria can be very profitable, if all the necessary components of improved production technologies are harnessed.

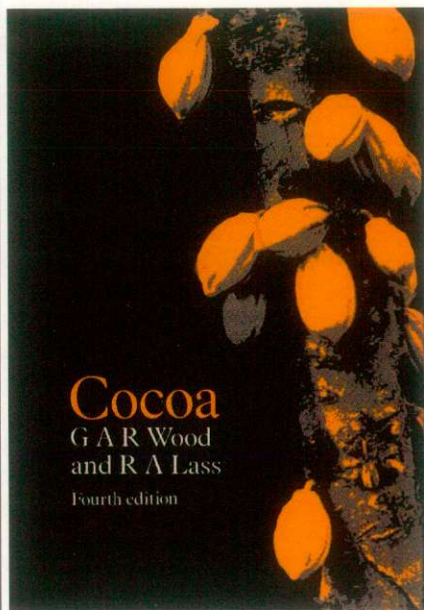
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REFLECTIONS ON THE YIELD OF UPPER AMAZON COCOA HYBRIDS IN GHANA WITH REFERENCE TO BREEDING FOR COCOA SWOLLEN SHOOT VIRUS RESISTANT VARIETIES

B. Adomako and Y. Adu-Ampomah³

INTRODUCTION

When cocoa research begun in Ghana in the mid-thirties it soon became apparent that the limited locally available germplasm would not permit a successful breeding programme. The germplasm available appeared to have been derived from a restricted area of the Amazon, so that there was very little genetic variation. The primary object of introducing Pound's Upper Amazon cocoa collections to West Africa (Posnette, 1948) was, therefore, to have available a greater genetic variability for breeding programmes.

Cocoa hybrids based on the Pound's (1940) Upper Amazon collections have been the basis of cocoa improvement in many countries ever since Posnette (1948) first observed heterosis in crosses involving them. This Upper Amazon material has also proved to be a rich source of disease resistance. In Ghana, a measure of resistance to infection with Cocoa Swollen Shoot Virus (CSSV) was observed in Upper Amazon material by Posnette and Todd (1951). The general observation was that Pound's Iquitos Mixed Calabacillo (IMC) and Nanay (NA) populations were the best sources of CSSV resistance (Attafuah and Glendinning, 1965; Glendinning *et al.*, 1966; Legg and Kenten, 1971a; Legg and Kenten, 1971b; Legg and Lockwood, 1977).

The significance of the discovery of a lethal virus disease of cocoa in Ghana in 1936 became fully appreciated in the 1940s (Posnette, 1947; Voelcker, 1948) and because of the enormous scale which the disease had reached, particularly in the Eastern Region, urgent attention and immediate solution to the problem became necessary. A great deal of work has been done on all aspects of cocoa cultivation at various times but the swollen shoot disease and its control has dominated research activities at CRIG until recently. One of the solutions to the CSSV problem was to breed for varieties resistant to the virus. It has long been apparent that successful hybrids with considerable resistance to infection could be produced by inter-crossing selected Upper Amazon parents. The problem has been how to produce suitably resistant hybrids that combine heterosis with many other attributes required if they were to perform better than the hybrids already being used (Thresh *et al.*, 1988).

The minimum period for producing entirely new hybrids was estimated to be about 12 years. To expedite progress, a breeding strategy was adopted to identify pollen parents, which, when crossed with the Upper Amazon female parents in the already established seed gardens in the Eastern Region, would give hybrids with higher levels of CSSV resistance and higher yield with at least the same levels of other agronomic and quality characteristics as the hybrids already being produced.

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In a study on trends in cocoa production, the International Cocoa Organisation (ICCO) deduced that at maturity (15-19 years) the maximum yield of Amelonado cocoa in farms in Ghana was 443 kg/ha whereas hybrids would give 841 kg/ha after seven years (Lockwood, 1977). But even in the well managed breeding trials the hybrids are generally yielding below the 841 kg/ha. It is, therefore, necessary to find out the factors that limit the yielding capacity of the hybrids. Only then can a meaningful breeding strategy be suggested for improvement. This paper examines the apparent causes of the low yields of the hybrids in four of the breeding trials established at Apedwa Cocoa Station in the 1970s and two others planted later in the 1980s at Tafo.

MATERIALS AND METHODS

In the first phase of the breeding programme initiated by the British Research Team (BRT) in Ghana between 1969 and 1978 (Legg, 1981), the four existing seed garden female parents, T85/799, T63/967, T63/971 and T79/467, were crossed to a wide range of promising pollen parents selected mainly from IMC and Nanay populations. The resulting hybrids were inoculated as beans in the laboratory to evaluate their resistance to virus. The progenies selected for field trials were mainly those which performed best in the CSSV screening tests. The field trials were established at Apedwa Cocoa Station (16 km from Tafo) to assess yield and agronomic features (Lockwood, 1977). Two similar progeny trials were later established at Tafo after the first phase of the BRT programme, by crossing two seed garden female parents, T60/887 and T85/799, to a range of pollen parents which included some non-Pound's Upper Amazon parents (Adomako and Enuson, 1987).

Each of the four trials (10th – 13th progeny trials) established at Apedwa consisted of six rows of seven trees spaced at 2.44 m², except for Amelonado in the 10th progeny trial area (PTA) which was planted at 2.44m x 1.22m to give 90 tree plots. The trials were planted at the onset of the main rains in May/June 1971 (10th PTA), 1972 (11th PTA), 1973 (12th PTA) and 1974 (13th PTA). The Tafo trials, 25th and 26th PTAs, were planted in June 1983 and 1985 respectively (Adomako and Enuson, 1987). In all the trials the trees were planted in a completely randomised block design with four replications in the 10th, 12th and 13th PTAs and five replications in the 25th and 26th PTAs where the plots consisted of 25 trees spaced at 2.5m². The 11th PTA was made up of two sub-trials, each with three replications. All the trials at both Apedwa and Tafo were surrounded by perimeter guard trees of cocoa. The cocoa was grown under shade of mixed species of forest trees, supplemented with *Gliricidia sepium*. Missing cocoa trees were replaced for up to two years after planting. Insect damage was controlled with insecticides, but neither fungicides nor fertilisers were used.

Observations made included:

- i. Trunk circumference 15cm above leaf litter was measured three years after planting and again at the end of the trials (adult plant girth).
- ii. Height at first jorquette.
- iii. Pods harvested at intervals of approximately three weeks.

Net yield (kg/ha of dry cocoa) was based on healthy ripe pods containing useable beans and was calculated using weighted mean pod values (number of pods required to give 1kg dry cocoa beans) of crosses obtained by the method of sample fermentation as described by Lockwood and Edwards (1980).

Table 1a. Origin of Trees Selected from Pound's Upper Amazon Material

T Number: These are seedlings within pod progenies introduced from Trinidad in 1944. Thus T17/524 is a seedling of pod number T17:

T17	:	IMC 53	open	pollinated
T44	:	Pound's refractario	selection	made in 1937 at Hacienda Bolivar, Ecuador
T60	:	PA 7	x	NA 32
T61	:	NA 33	x	NA 32
T63	:	PA 35	x	NA 32
T65	:	PA 7	x	IMC 47
T72	:	NA 32	x	IMC 60
T73	:	NA 33	x	IMC 60
T79	:	NA 32	x	PA 7
T85 & T87	:	IMC 60	x	NA 34
T90 & T101	:	IMC 76	x	NA 32
T92	:	NA 32	x	NA 31

Introduced Clones

1. POUND (PD) : Material collected as budwood in the headwaters of the Amazon by Pound. He described the clones as Nanay, Napo, Iquitos Island or Contamana depending on origin: POUND 7 (Nanay), POUND 10 (Nanay), POUND 15 (Nanay), POUND 21 (Napo).
2. Pound's Iquitos Mixed Calabacillos or Mixed Collection (IMC) : IMC 23, IMC 47, IMC 60, IMC 67, IMC 76.
3. Pound's Nanay Selections (NA) : Material collected on the riverbanks between Rio Amazon and Rio Nanay, Ecuador: NA 33, NA 34, NA 440.
4. Pound's Parinari Selections (PA) : Found on the lower Rio Marañon, Ecuador: PA 7, PA 107, PA 150.
5. Pound's Scavina population (SCA) : SCA 6, SCA 9.

Table 1b. Origin of Parent Trees Selected from Non-Pound's Upper Amazon Material

AMEL	:	West African Amelonado
ALPH. B	:	T60 x T60 crosses made at Tafo, Ghana.
AMAZ	:	Material collected by Chalmers about one hour up River from Iquitos.
BE	:	Selections made at different localities around Belem.
CATONGO	:	A white seeded 'Comum' type found near Urucuca, Bahia, Brazil.
C-SUL	:	Selections made in the vicinity of Cruzeiro do Sul, Acre, Brazil on the Jurua river.
EQX	:	Selections made in the Oriente region of Ecuador.
GA	:	Grande Anse, Haiti selections (GA 11 : Trinitario)
ICS	:	Imperial College Selections. (ICS 6 : Trinitario).
RB	:	Selections made on the Rio Branco, Brazil.

The parents used in the breeding programme are listed in Table 1a and Table 1b and the crosses made in Table 2. Two hybrids were chosen as standards for comparison: T85/799 x Amelonado because it was the highest yielding hybrid in earlier series of trials and T85/799 x T79/501 because it was moderately high yielding and showed exceptionally low losses from blackpod disease (Lockwood, 1976).

Table 2. Crosses Made for the Progeny Trials at Adepwa and Tafo

1) Adepwa Trials

Female Parent	x	Male Parent
<u>10th P.T.A.</u> T85/799	x	PA7, SCA 6, T65/326, T63/967, T63/971, T61/1313, T65/238, T17/524, T92/1615, T101/2540, T90/344, IMC 60, NA 34
<u>11th P.T.A.</u> T63/967	x	T17/524, T65/238, T87/1312, T79/378, IMC 76, T79/501
<u>11th P.T.A.</u> T63/467	x	T17/524, T87/1312, T101/2540, T65/202, T92/1615, T61/1313, IMC 47, IMC 60, IMC 67, IMC 76, NA 34
<u>12th P.T.A.</u> T63/967	x	T65/326, T60/887, T101/2540, T73/612, T85/797, IMC 47, IMC 60, IMC 76, GA 11
<u>13th P.T.A.</u> T63/971	x	T65/238, T60/887, T65/326, T72/1765, T85/797, T87/1312, T101/2540, IMC 47, IMC 60, IMC 76, NA 34

2) Tafo Trials

<u>25th P.T.A.</u> T60/887	x	BE 8, POUND 7, POUND 10, POUND 15, POUND 21, EQX 3338, EQX 3356, EQX 3364, CATONGO, MA 12, AMEL, ICS 6, RB 49, C-SUL 7, AMAZ 3-2, IMC 60, IMC 78, T17/524
<u>26th P.T.A.</u> T85/799	x	PA7, PA 107, PA 150, MA 12, SCA 9, GA 11, IMC 23, IMC 78, ALPH.B36, NA 440, T44/547

RESULTS AND DISCUSSION

Yield

Mean net yields from all the trials, particularly those established at Adepwa, were very low (Table 3), with average yields generally below 500 kg/ha/yr. The low yields may be due to several factors including inbreeding depression, poor general combining ability (GCA), unfavourable yield-vigour relationships, environmental factors and genotype-environment interactions.

Table 3. Net Yield (Kg/Ha) for Progenies in the Apedwa and Tafo PTAs

Yield Parameters	Apedwa 10th PTA – T85/799 Progenies 1974 – 1986	Apedwa 11th PTA – T63/967 Progenies 1975 - 1986	Apedwa 11th PTA – T79/467 Progenies 1975 - 1986	Apedwa 12th PTA – T63/967 Progenies 1976 - 1986	Apedwa 13th PTA – T63/971 Progenies 1977 - 1986	Tafo 25th PTA – T60/887 Progenies 1987 – 1998	Tafo 26th PTA – T85/799 Progenies 1989 - 1998
Mean net yield (including controls)	469.8	420.5	336.0	320.8	184.1	813.9	443.2
Mean net yield (excluding controls)	452.1	408.3	327.7	316.6	167.2	805.7	449.4
Mean net yield for control 1: (T85/799 x Amel) 2: (T85/799 x T79/501)	574.0 631.8	515.6 410.5	410.5 370.4	343.2 440.5	252.3 299.7	943.7 831.0	438.7 379.4
Range for all crosses (net yield)	342.6–720.8	260.3–515.6	210.2–445.5	223.1–440.5	124.1–299.7	612.4–965.8	189.7–704.9
Highest yielding crosses	T85/799 x PA 7 (720.8) T85/799 x T79/501 (631.8) T85/799 x SCA 6 (582.9)	T63/967 x T17/524 (515.6) T85/799 x Amel (515.6) T63/967 x T65/238 (500.7)	T79/467 x T17/524 (445.5) T85/799 x Amel (410.5) T79/467 x IMC 47 (400.4)	T85/799 x T79/501 (440.5) T63/967 x IMC 60 (383.3) T63/967 x T65/326 (343.2)	T85/799 x T79/501 (299.7) T85/799 x Amel (252.3) T63/971 x T65/238 (212.2)	T60/887 x BE 8 (965.8) T85/799 x Amel (943.7) T60/887 x POUND 10 (912.5)	T85/799 x PA 150 (704.9) T85/799 x PA 7 (646.9) T85/799 x MA 12 (594.1)
Lowest yielding crosses	T85/799 x NA 34 (342.6) T85/799 x T90/344 (342.6) T85/799 x IMC 60 (360.4)	T63/967 x T79/501 (260.3) T63/967 x IMC 76 (370.4) T63/967 x T79/378 (380.4)	T79/467 x T61/1313 (210.2) T79/467 x T92/1615 (230.4) T79/467 x NA 34 (255.3)	T63/967 x T85/797 (223.1) T63/967 x IMC 47 (263.1) T63/971 x T73/612 (280.3)	T63/971 x T101/2540 (124.1) T63/971 x T87/1312 (134.8) T63/971 x T85/797 (145.5)	T60/887 x POUND 21 (753.0) T60/887 x C-SUL 7 (627.7) T60/887 x EQX 3364 (612.4)	T85/799 x IMC 23 (189.7) T85/799 x NA 440 (269.7) T85/799 x IMC 78 (279.3)

Table 4. Yield-Vigour Relationships (Yield-Indices) Based on Total Yield and Adult Tree Circumference for Progenies in the Apedwa and Tafo PTAs

Yield Index (I)	Apedwa 10th PTA – T85/799 Progenies	Apedwa 11th PTA – T63/967 Progenies	Apedwa 11th PTA – T79/467 Progenies	Apedwa 12th PTA – T63/967 Progenies	Apedwa 13th PTA – T63/971 Progenies	Tafo 25th PTA – T60/887 Progenies	Tafo 26th PTA – T85/799 Progenies
Mean yield-index (including controls)	3.4	3.1	3.0	2.4	1.5	6.4	4.2
Mean yield-index (excluding controls)	3.3	3.1	2.9	2.3	1.4	6.6	4.1
Mean yield-index for Control 1: (T85/799 x Amel)	4.6	3.9	3.4	2.7	2.4	9.7	5.3
Control 2: (T85/799 x T79/501)	3.9	2.6	2.7	2.6	2.1	6.2	4.0
Range for all crosses	2.0-5.8	1.5-4.5	1.7-4.8	1.7-3.3 0	8-2.5	4.4-9.9	1.7-6.1
Crosses with highest yield indices	T85/799 x SCA 6 (5.8) T85/799 x PA 7 (5.0) T85/799 x Amel (4.6)	T63/967 x T17/524 (4.5) T63/967 x T65/238 (4.4) T63/799 x Amel (3.9)	T79/467 x T17/524 (4.8) T79/467 x T87/1312 (3.6) T79/467 x T65/202 (3.6)	T63/967 x T60/887 (3.3) T63/967 x IMC 60 (3.0) T63/967 x T73/612 (2.8)	T63/967 x T60/887 (2.5) T85/799 x Amel (2.4) T63/971 x T65/238 (2.1)	T60/887 x T17/524 (9.9) T85/799 x Amel (9.7) T60/887 x EQX 3364 (9.6)	T85/799 x PA 150 (6.1) T85/799 x PA 7 (6.1) T85/799 x SCA 9 (5.4)
Crosses with lowest yield indices	T85/799 x NA 34 (2.0) T85/799 x T101/2540 (2.2) Amel selfed (2.2)	T63/967 x T79/501 (1.5) T63/967 x Amel (2.7) T63/967 x T79/501 (2.6)	T79/467 x T61/1313 (1.7) T79/467 x NA 34 (2.2) T79/467 x IMC 67 (2.4)	T63/967 x T101/2540 (1.7) T63/967 x T85/797 (1.8) T63/971 x GA 11 (1.9)	T63/971 x T101/2540 (0.8) T63/971 x T87/1312 (1.0) T63/971 x T85/797 (1.2)	T60/887 x C-SUL 7 (4.4) T60/887 x POUND 21 (5.1) T60/887 x EQX 3364 (5.5)	T85/799 x IMC 23 (1.7) T85/799 x NA 440 (2.3) T85/799 x IMC 78 (2.4)

- (1) Total yield (g) divided by adult-tree trunk girth (cm) to the cube. Adult-tree trunk girths were measured in November 1986 for Apedwa trials and 1998 for Tafo trials.

Inbreeding Depression

Hybrids which gave the lowest yields were, invariably, intra-population crosses. For instance, in the 10th and 26th PTAs, the hybrids which obtained the lowest yields were mostly NA and IMC parents, indicating the effects due to inbreeding since the female parent T85/799 is a seedling of IMC 60 x NA 34. On the other hand, crosses of T85/799 with the Parinari pollen parents were the best for yield which may be due to some level of heterosis. The problem of how to produce suitably CSSV resistant hybrids with heterosis was apparent when the CSSV breeding programme was initiated. However, because of the limited range of germplasm material available at CRIG at the time and the need to find an immediate solution to the devastating Cocoa Swollen Shoot Virus (CSSV) problem, inbreeding could not be avoided.

An analysis of the Ghanaian breeding trials planted between 1952 and 1974 shows that a narrow range of parents have been used and that many were closely related to each other and involved mostly Pound's (1940) Upper Amazon parents. The Pound's Upper Amazon material is derived from only a few wild ancestors from one region of Peru and therefore the genetic base was not broad. This means that plant breeders who try to produce disease resistant and higher yielding varieties by selection and hybridisation from within the narrow-base genetic material can only make limited progress. In a breeding programme a limit is often reached when a gene pool responsible for yield components gets stabilised. At this stage hybridisation within the same pool may not result in any further improvement in yield.

Yield-Vigour Relationships

Yield-vigour relationships measured as yield indices are presented in Table 4. Yield-index was calculated as yield of dry cocoa beans (in grams) divided by trunk girth (cm) to the cube. The cube of the girth is considered a better representation of the mass of the tree than the girth *per se* or the square of the girth (Albertus Eskes, *pers. comm.*). Generally crosses that had poor yield indices recorded very low yields. The low yields appear to be associated with an unfavourable partitioning of assimilate between growth and crop. The trees had the capacity to produce and retain adequate quantity of vegetative material (biomass) but the partition, the capacity to divert biomass to the desired product, the crop, was unsatisfactory.

Vegetative vigour in the pre-bearing phase is an important characteristic as this leads to ease of establishment. Varieties with such traits are particularly required for areas which originally carried cocoa but have become marginal. However, after bearing commences vegetative growth should, ideally, reduce or be suppressed in favour of reproductive product. Glendinning (1966) observed that the relationship between yield and trunk diameter increment of cocoa generally becomes negative, thus indicating that yield was being obtained at the expense of current vegetative growth. Results from the present study, however, suggest that vegetative growth was being obtained at the expense of yield, partially accounting for the low yields of the hybrids.

General Combining Ability (GCA) Effects

Among the desirable traits of a clone which is to be used as the female parent for seed production in Ghana are:

- i. Self-incompatibility.
- ii. Ease of propagation, ready establishment and vigour (characteristics which normally go together).
- iii. Profuse flowering.
- iv. High GCA for as many economic characteristics as possible.

The first three of the four characteristics listed can be evaluated within five years of planting, but assessing GCA for yield and pod and bean traits would take a longer period. The urgent need for new seed production plots when no new varieties were available prevented such a lengthy evaluation of female parents. Because of the good observations on vigour, precocity and favourable flavour of beans, pods from 11 of the introduced Upper Amazon material (T types) were approved for distribution to farmers in the 1950s (Rogers, 1955; Toxopeus, 1964). The four female parents used in the breeding trials were among the approved T types of planting material (Table 5). Thus their selection was not based on detailed evaluation of their GCA's.

Table 5. The Eleven Approved T Type Upper Amazon Introductions

Type	Percentage		
T12	Scavina (SCA 12) open-pollinated		
T60*	Parinari (PA 7)	X	Nanay (NA 32)
T63*	Parinari (PA 35)	X	Nanay (NA 32)
T65	Parinari (PA 7)	X	Iquitos (IMC 47)
T72	Nanay (NA 32)	X	Iquitos (IMC 60)
T73	Nanay (NA 33)	X	Iquitos (IMC 60)
T76	Parinari (PA 35)	X	Nanay (NA 31)
T79*	Nanay (NA 32)	X	Parinari (PA 7)
T82	Nanay (NA 32)	X	Parinari (PA 35)
T85*	Iquitos (IMC 60)	X	Nanay (NA 34)
T87	Iquitos (IMC 60)	X	Nanay (NA 34)

* Female parents in the Eastern Region seed gardens used as the pod bearing parents in the breeding trials.

In a field trial which evaluated the breeding value of 10 Upper Amazon cocoa selections in Ghana by estimating their GCA's for yield, pod and bean traits as well as blackpod (*Phytophthora palmivora*) disease and vegetative characteristics (Adomako *et al.*, 1999), PA 150, POUND 10 and PA 7 were found to be better, especially in terms of overall net yield, than the two widely used seed garden female parents T85/799 and T60/887 which were among the four female parents used in the breeding trials of the present study (Table 6).

Table 6. Mean, Range, Coefficient of Variation and GCA Values for Yield (kg/ha) and Percentage Blackpod for Female and Male Parents of Upper Amazon Coconas in Ghana

Parent	GCA Values For			
	Yield (11 yrs)	Yield (first 3 yrs)	Yield (last 8 yrs)	Blackpod Incidence
Female				
PA 150	139.35***	66.73	166.59***	-1.88**
POUND 7	61.29*	47.76	66.36*	-1.41
PA 7	23.65	-23.94	41.52	-0.08
T60/887 ⁺	13.25	78.43***	-11.21	0.92
IMC 60	-1.11	-40.71*	13.72	0.79
T85/799 ⁺	-18.01	-6.01*	-22.51	1.16**
ALPH. B36	-20.41	-26.17	-18.28	1.58***
POUND 26	-57.18* ⁽¹⁾	-52.34** ⁽¹⁾	-58.98*	2.92*** ⁽¹⁾
T65/238	-64.78***	-16.13	-95.11**	-0.28
T17/524	-76.05***	-59.87**	-82.11**	-0.54
S.E.	21.62	16.64	25.58	0.49
Males				
SCA 9	18.00	103.94*	-14.23	0.36
P30	-3.63	-38.60	9.48	-0.44
ICS 6	-14.37	-65.33	4.75	0.07
			6.92	0.22
S.E.	8.77	42.90	437.8	
Grand mean	392.4	271.2	437.8	11.0
Range ⁽²⁾	266.3-586.5	103.8-638.9	242.1-714.4	7.5 - 14.9
C.V.	15.1	27.7	18.6	16.8

(1) *, **, ***: GCA values significantly different from zero at the 0.05, 0.01, and 0.001 probability level respectively.

(2) Range for means of individual crosses.

+ Female parents already being used in the seed gardens.

C.V. Coefficient of variation.

Even though T60/887 was among the best female parents for yield in Côte d'Ivoire (Paulin *et al.*, 1994) and also in Ghana (Adomako *et al.*, 1999), especially when early yields are considered, its tendency of not sustaining yields in later years makes it a less favourable choice as a female parent. It appears, therefore, that the low yields of the hybrids may partly be due to poor GCA's of the female parents used.

Environmental Effects

In nature, ideal conditions for plant growth are seldom achieved. There are always defects of climate, local environment and management (Simmonds, 1981). These defects may alter the manufacture, translocation, partition and utilisation of plant photosynthates, mineral nutrients and water. Two of the environmental factors that might have affected the yields of the hybrids in the trials are related to soil and shade.

Cocoa trees are more demanding in terms of soil conditions than other tropical tree crops such as oil palm, coffee and rubber, since cocoa requires well-drained deep soils and a top soil rich in organic matter and fairly rich in minerals (Petithuguenin, 1998).

All the trials in the present study were established on land which had previously carried cocoa, thus the soils were degraded to some extent and this could have contributed to the low yields of the hybrids since no fertilisers were applied when the new trials were established.

The cocoa was grown under dense shade as is the normal practice in Ghana. The dense shade is to prevent loss of soil moisture during the long dry season, and also to mitigate attack by insect pests. The dry seasons in Ghana have tended to become more protracted since the 1970s. In West Africa, bean size has been found to decrease in the event of severe water stress during the July-August short dry season, at the time of the main crop development (Petithuguenin, 1998). Trials in West Africa have also shown that potential yields of cocoa can be doubled by removing permanent shade, provided fertilisers are applied. If cocoa trees are in full sunlight, without fertilisers, yields are satisfactory in the short term, but fall in the medium-term (Ahenkorah *et al.*, 1974).

If soil fertility and management conditions are limiting, no amount of plant breeding can improve yields substantially. The appropriate variety – agronomy ‘package’ is, therefore, essential in bringing about improved yields.

Genotype-Environment (GE) Interactions

The Series II hybrids used as standards for comparison in this series of trials are known to be high yielding when given appropriate environmental conditions and management (Lockwood, 1971). The yields from these hybrids were very low particularly in the 12th and 13th PTAs established at Apedwa but were relatively higher in the 25th PTA at Tafo (Table 3). This indicates that the disappointingly low average yields did not arise solely from the experimental hybrids having limited genetic potential but GE interactions contributed to the low yields. The hybrids appear to be less stable, varying widely and doing well in good conditions but badly in poor conditions.

There may be the need to define sub-environments (narrow range of environments) for the hybrids to which they will be well adapted and perform well or breed for more stable, widely adapted genotypes. However, as ecological conditions in the southern forest zone of Ghana continue to deteriorate, it would be better to seek widely adapted genotypes.

CONCLUSIONS

This study examined the apparent causes of the low yields of Upper Amazon cocoa hybrids in six of the breeding trials established at Apedwa Cocoa Station and CRIG, Tafo between 1970 and 1986.

Hybrids which gave the lowest yields were, invariably, intra-population crosses involving mostly NA and IMC parents, indicating effects due to inbreeding depression. Inbreeding could not completely be avoided when the breeding programme was initiated because of the limited range of germplasm material then available. The range of cocoa germplasm material at CRIG has now been increased, with the introduction of several non-Pound's Upper Amazon accessions. To avoid the problem of inbreeding in future breeding trials, it is necessary that these latter introductions are thoroughly screened and evaluated to identify other possible sources of CSSV resistance and desirable traits. Breeding for disease resistant varieties and yield should be major selection criteria.

Hybrids that had unfavourable yield-vigour relationships recorded very low yields. It appeared that vegetative growth was being obtained at the expense of yield. Since partition of plant photosynthate is an important factor that controls yield, breeders should aim to breed for varieties that maximise the partition towards yield - such as precocious-bearing varieties with moderate vigour.

The urgent need for seed production plots when new varieties were available prevented a thorough evaluation of the breeding values of the female parents. Thus, some of the parents used in the breeding trials and which are also being used in the seed gardens have poor GCA's for yield, contributing to the poor yields of their progenies. However, parents with good GCA's have now been identified. Thus a new series of superior hybrids in terms of yield can be selected for farmers using the female parents with better GCA's in crosses with unrelated pollen parents.

Establishment of new cocoa farms in Ghana from now will largely be replantings on land that had previously carried cocoa. Some of the farms that need to be replanted are very old and unproductive or have been destroyed by CSSV disease and/or by capsids. It is very important that farmers are encouraged to replant cocoa on the same land that was previously used for cocoa cultivation since they should not be allowed to move into the country's dwindling forest reserves because of the potentially negative impact on the environment.

There is the need, therefore, to assure farmers that they can replant successfully on land previously used for cocoa cultivation. This can best be done by providing them with cocoa varieties that can establish easily and give satisfactory yields under such seemingly marginal areas. However, if soil fertility and management conditions are lacking, no plant breeding *per se* can significantly improve yields. Farmers would, therefore, require in addition to improved planting material, the knowledge and the capacity to use the appropriate agronomic practices to ensure increased cocoa productivity in the 21st century.

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APPLICATION OF THE TECHNIQUES OF MOLECULAR BIOLOGY TO COCOA PATHOLOGY

Gareth Griffith ⁴

INTRODUCTION

The overwhelming public perception of the recent advances in molecular genetics is that such techniques are primarily used in the production of transgenic crop plants or in animal cloning (e.g. FlavrSavrTM tomatoes, Dolly the sheep). However, the same technological advances have revolutionised many areas of biological research, where they do not give rise to similar ethical or ecological concerns. This article will illustrate the impact of this technology on cocoa pathology and give an indication of its future potential.

Plant pathology, like all aspects of microbiological research, faces the inherent problem that the organisms under study are microscopic and generally only visible to the naked eye when causing disease lesions on a host plant or producing macroscopic fruiting structures (e.g. mushrooms). Even during microscopic examination, few phenotypic characters are discernible and the reliable identification of microbial pathogens to the species level requires a significant degree of expertise. For the cocoa-grower, it can often be important to obtain a correct and precise identification of the causal agent of a disease (preferably at an early stage in the disease cycle), so that appropriate control measures can be implemented. It may also be important to achieve identification below the species level, for example where two races of a given pathogen differing in their pathogenicity to particular hosts may be present.

Where morphological identification is problematic or unreliable, pathologists have developed a range of biochemical, microscopic and genetical tools to aid the investigative process. The most powerful of these tools are those of molecular biology, which permit direct analysis of the genetic material (the DNA) of the pathogen. At the simplest level, genetic features characteristic of a particular pathogen can be elucidated, though the power of these techniques is such that it is also possible to gain an insight at the molecular level into the processes which underlie pathogenesis. Such a detailed understanding of the events which take place during the process of host infection can identify targets for disease control such that pesticides with specific activity against the target pathogen can be developed.

Cocoa trees are susceptible to a panoply of microbial pathogens, many of which are still poorly understood. Of these, the three which are considered most important and have thus received the greatest attention are *Phytophthora* pod rot (PPR or Black Pod caused by Oomycete pseudofungi), witches' broom disease (caused by the basidiomycete fungus *Crinipellis perniciosus*) and swollen shoot disease (caused by the Cocoa Swollen Shoot Virus (CSSV), a non-enveloped bacilliform badnavirus). All three have been the subject of study by molecular plant pathologists to varying degrees. The advances permitted by the advent of molecular biology for each of these are described below. *Phytophthora* Pod Rot (PPR), also widely known as black pod, is found in all cocoa-growing regions of the world, though its severity varies depending on climatic conditions, the variety of cocoa being grown, the relative importance of PPR compared to other diseases in a particular areas (e.g. South America) and also the precise nature of the causal agent itself. PPR disease of cocoa

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causes some 45% of the total global crop loss attributed to pest and diseases though locally, however, it can cause losses of up to 80% (Lass, 1985).

It is now recognised that PPR is caused by several species belonging to the genus *Phytophthora* rather than by a single pathogen (*P. palmivora*), as was originally believed (Waterhouse, 1963). Brasier and Griffin (1977) identified consistent morphological and karyotypic differences between isolates morphologically distinct forms of *P. palmivora*. This evidence resulted in the erection of a new species, *P. megakarya* (Sansome *et al.*, 1975), members of which possessed 5-6 larger chromosomes (L-type = large chromosomes, hence *mega-karya*) as opposed to the 9-12 smaller chromosomes (S-type = small chromosomes) found in isolates more typical of *P. palmivora*. The various morphological forms (denoted MF1 to MF4) of *Phytophthora* pathogens which are found on cocoa have now been identified as *P. palmivora* (MF1 and MF2), *P. megakarya* (MF3) and *P. capsici* (MF4) (Förster *et al.*, 1987; Griffin, 1977; Oudemans and Coffey, 1991). However, there remains some confusion about the extent to which MF4 and *P. capsici* are entirely synonymous (Ortiz Garcia, 1996). An additional complication is that PPR is occasionally caused by other *Phytophthora* species, such as *P. citrophthora* in Brazil and some parts of West Africa (Campêlo and Luz, 1981; Kellam and Zentmyer, 1981; Ortiz Garcia, 1996).

Of the three main species of *Phytophthora* associated with PPR, *P. palmivora* is world-wide in its distribution and *P. capsici* is locally important in several areas, notably parts of Brazil, Mexico, the Caribbean and India (Chowdappa and Mohanan, 1994; Lass, 1985; Ortiz Garcia, 1996). Of greatest concern however (particularly in the context of cocoa supplied to Europe), is *P. megakarya*, which causes the most severe symptoms and over recent decades has spread progressively through the cocoa-growing areas of West Africa. At present, *P. megakarya* has not been reported from the more westerly parts of West Africa (e.g. Ivory Coast), nor has it spread to cocoa-growing regions outside Africa (Luterbacher and Akrofi, 1994).

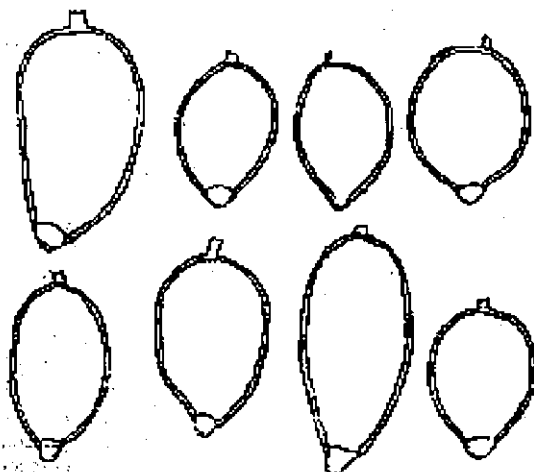
It is possible to distinguish *P. palmivora* and *P. megakarya* on the basis of sporangial morphology (*P. palmivora* isolates have smaller protrusions [papillae and pedicels] on their sporangia) (Figure 1), as well as macroscopic features, such as lesion morphology (*P. megakarya* lesions have a more powdery appearance) and colony appearance on agar (*P. palmivora* generally has a stellate appearance in culture). However, reliable differentiation of the two species based on these features is a specialist skill, usually requiring the time-consuming isolation of the pathogen into pure culture on selective media. Characterisation of the two species using biochemical markers was first reported by Idosu and Zentmyer (1978) and Erselius and Shaw (1982), who found differences in isoenzyme and total protein profiles respectively between the two species. A more detailed study of isoenzyme variability by Oudemans and Coffey (1991) found *P. palmivora* isolates from a wide variety of hosts throughout the world to be genetically very homogeneous, while isolates of *P. megakarya* were polymorphic (variable) at several loci. They defined two groupings within *P. megakarya*, with group MK1 being found in Nigeria and Equatorial Guinea and group MK2 being found in Cameroon.

The advent of molecular biology has provided the technology for more direct and detailed analysis of genetic variability at the DNA level within and between PPR pathogens. A number of more general investigations have been conducted to examine genetic variation within and between various *Phytophthora* species (many of which are important pathogens of crops other than cocoa).

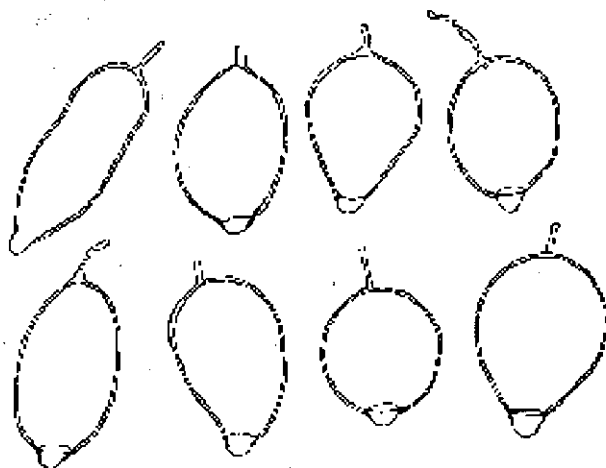
Figure 1: Variability in sporangium morphology of (A) *Phytophthora palmivora* and (B) *P. megakarya*

Figure 1

(A)



(B)



Source: Redrawn from Brasier and Griffin (1977). Note the variability both in sporangium size and shape. Bar equals 50 µm.

Förster *et. al.* (1990) purified mitochondrial DNA from a large number of *Phytophthora* isolates and the restriction maps (obtained by locating the cutting sites for several restriction enzymes) for these broadly confirmed those of previous isoenzyme studies (Oudemans and Coffey, 1991), in that *P. palmivora* was found to be genetically very homogeneous and that *P. megakarya* isolates were split into two groups. They suggested that *P. megakarya* was indigenous to West Africa on an unknown host and that present populations found causing black pod in different areas might have independent origins. A more detailed study of isoenzyme polymorphisms alone (Blaha, 1994) and together with RAPD (see below) polymorphisms Nyassé (1997; 1999) confirmed the existence of two distinct populations in West and Central Africa, with the greatest genetic diversity being found in Cameroon. These data further suggested that though most populations were clonal in structure, there was evidence of sexual reproduction in field populations from Cameroon.

Earlier studies on PPR pathogens were originally prompted by a desire to establish the relatedness of the various pathogens to each other and to other *Phytophthora* species. The emphasis has now shifted towards the development of rapid methods of distinguishing the various pathogens. In Ghana (where *P. megakarya* is locally present) and Ivory Coast (where currently only *P. palmivora* is reported to occur), such methods will permit accurate monitoring of the distribution of *P. megakarya*. One such diagnostic assay has been published by Sackey and Luterbacher (1994), in which DNA probes specific to *P. megakarya* and *P. palmivora* were isolated and cloned. By hybridisation of labelled probes against DNA from PPR isolates collected from the field in a dot blot assay, it was possible to distinguish the two species from as little as 0.1 ng of DNA from a pure culture (it should also serve to identify pathogens from disease lesions).

The PCR technique (see Appendix I – attached to this article) has revolutionised diagnostic assays in plant pathology. It is both sensitive enough to use on small quantities of biological material yet specific enough to be highly discriminating. The first use of PCR amplification to differentiate PPR pathogens involved the RAPD technique (see below) though this can only be used with DNA from pure cultures (John Spence, *pers. comm.*; Denis Despréaux, *pers. comm.*).

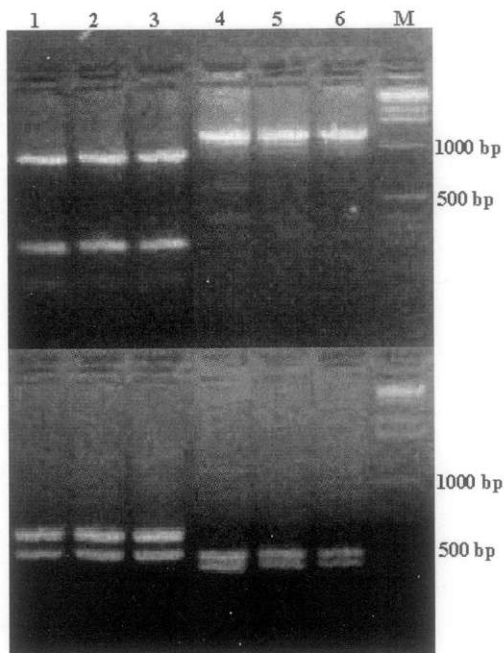
A feature of the all methods described above is that not only must the pathogen be isolated into pure culture, a process which may take several weeks to complete but also in some protocols, large quantities of mycelium are required for analysis. For instance, purification of mitochondrial DNA (mtDNA) is a protracted process which initially requires that several grams of mycelium be grown up in liquid culture (e.g. Förster *et. al.*, 1990). Protocols such as isoenzyme analysis also involve the use of hazardous stain compounds to test for enzyme activity. Since the aim of a diagnostic assay is to provide an identification both rapidly and economically, an ideal method would permit pathogen identification directly from disease lesions within a few days of sampling.

Based on the observations of Förster *et. al.* (1990) that polymorphisms exist between the mitochondrial DNA (mtDNA) of *P. palmivora* and *P. megakarya*, a recent study funded by the BCCCA (Griffith and Jones, 1997) has developed a number of PCR probes to amplify polymorphic regions of mtDNA and which permit differentiation of the two pathogens. Primers were designed based on sequence information available for the mtDNA genome of *Phytophthora infestans* (which causes late-blight of potato). When cut with restriction enzymes the amplified fragments reveal polymorphisms between the two species in the form of distinctive banding patterns on an agarose gel. The specificity of this assay (due to the fact

that *Phytophthora* spp. are only very distantly related to both other fungi and plants) is such that DNA extracted directly from disease lesions can be analysed directly.

Small pieces of tissue cut from black pod lesions (and which may be preserved by air-drying) are used as the starting material. DNA is extracted from this tissue using a simplified extraction method and a small amount of the purified DNA is used in the PCR reaction. After the PCR amplification, the resultant PCR products are cut with a restriction enzyme and subjected to gel electrophoresis to separate fragments of different length (Figure 2). Samples are generally analysed in sets of 24 or 48 and a single operator can obtain results from the samples within 24 hours. The assay is economic (consumable cost of ca. £1-2 per sample) and although specialised equipment is required, such equipment is now commonplace in many biological research laboratories.

Figure 2. PCR-RFLP Digest of *Phytophthora megakarya* and *P. palmivora*



A 1300 bp fragment of mitochondrial DNA was amplified by PCR (with the primers F3A and R3A) and digested with the restriction enzymes *HaeIII*. (above) and *NdeII* (below). Lanes 1-3 *P. palmivora* (isolates TR1 [from Trinidad], CIV01, CIV04 [both from Ivory Coast]); lanes 4-6 *P. megakarya* (isolates GHBA6 [from Ghana], TG3, TG7 [both from Togo]); Lane M denotes a molecular weight marker (1 kb ladder; Gibco BRL Ltd.). Following agarose gel electrophoresis, isolates belonging to the two species clearly show distinct banding patterns.

WITCHES' BROOM DISEASE (WBD)

Of all the diseases of cocoa, WBD is currently the source of greatest concern. Its recent spread to Bahia State has decimated the Brazilian cocoa industry, as it did earlier this century in other Latin American countries, notably Ecuador. The possibility, however remote that it might spread beyond South America is viewed with considerable trepidation by the world's cocoa growers.

Unlike PPR, there is only one causal agent of WBD, namely the basidiomycete *Crinipellis perniciosa* though there is evidence that the fungus varies in pathogenicity in different parts of South America (Wheeler and Mepsted, 1984). Genetic differences between pathogen populations have previously been examined indirectly by investigation of basidiocarp morphology (Hedger *et al.*, 1987), isoenzyme polymorphisms (Griffith, 1989) and the identification of somatic incompatibility groupings (Griffith and Hedger, 1994a,b; McGeary and Wheeler, 1988; Wheeler and Mepsted, 1982; Wheeler and Mepsted, 1984, 1988).

The application of molecular biology techniques to the study of WBD is in its infancy. By Southern hybridisation with a mitochondrial DNA probe from *Coprinus cinereus*, Wilson (1995) (also detailed in Griffith *et al.*, 1994) identified RFLPs between isolates of *Crinipellis perniciosa* belonging to the three different biotypes (Cocoa (C-), Liana (L-) and *Solanum* (S-) biotypes; Griffith and Hedger, 1994a) though this method did not differentiate between isolates belonging to the pathogenic C-biotype. Andebrhan and Furtak (1994a,b) conducted RAPD analysis (see below) to assess levels of genetic variability between Brazilian isolates *C. perniciosa* from different host plants. They found an isolate of the S-biotype to be distinct from isolates of the C-biotype (including isolates found on relatives of *Theobroma cacao* and the shrub *Bixa orellana*) but that within the C-biotype isolates genetic relatedness was more dependent on geographical proximity than occurrence on a particular host.

Future work of this nature making use of recent advances in molecular technology (for example using the Amplified Fragment Length Polymorphism (AFLP) technique or DNA sequencing) will provide more detailed information about how the spread of the pathogen between different areas, the relationship between the various biotypes and the rate at which current populations are evolving in the field. This last aspect is of particular importance for the development of durable control strategies in the future. Analysis of the process of pathogenicity by more detailed study of the process of meristem infection and pathogen development *in planta* (for example, which host or pathogen genes are turned on and off during pathogenesis) may also elucidate new targets for disease control. Research into this last aspect is currently being funded by Cocoa Research UK Ltd. at University of Wales, Aberystwyth.

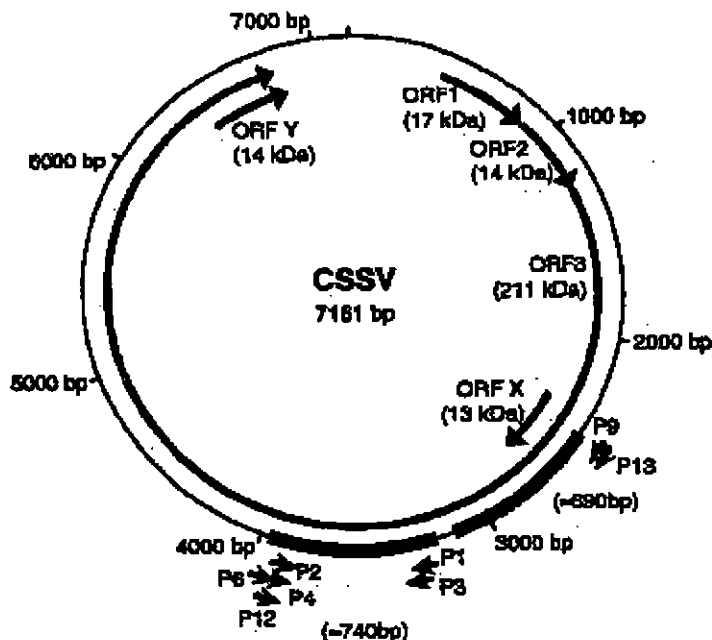
COCOA SWOLLEN SHOOT DISEASE

Cocoa swollen shoot disease was first reported on cocoa in West Africa in 1936, and although it was fairly quickly established that control could be achieved by total removal of infected trees, the difficulty of achieving this and the neglect of sanitation programmes during the Second World War resulted in a significant exacerbation of the disease problem for many years thereafter. The removal of almost 200 million trees has been necessary to bring the situation under control in the intervening period. The pathogen was identified as a virus by Posnette in 1940 and its mechanism of spread via mealybugs has also been long appreciated (Posnette, 1940; Posnette, 1947). Since viral infections cannot be cured (i.e. the pathogen cannot be killed, since viruses are not 'alive' in the normal sense of the word), preventative strategies (as opposed to the control strategy of removing infected trees) rely on breeding

resistant varieties, control of the vector or early identification of disease. The latent infection of trees is known to be a problem and it has been established that latently infected trees (i.e. not yet showing symptoms) can act as a source for further transmission (Legg, 1982).

The emphasis of recent CSSV research has been on detection of infection in the latent stage before any symptoms are manifest. Various immunological tests have been devised both to detect latent infection and also to differentiate the various strains of CSSV. An Enzyme-Linked Immunosorbent-Assay (ELISA) test devised by (Sagemann *et al.*, 1985; Sagemann *et al.*, 1983) was able to distinguish five CSSV serotypes in Ghana which correlated with differences in symptom production. However, this test is rather insensitive and only identifies the presence of CSSV in trees already showing symptoms. A more sensitive assay, the Virobacterial Agglutination (VBA) test has recently been developed (Hughes and Ollennu, 1993). This test differentiates 8 serotypes, as well as four subtypes of the well-characterised CSSV-1A group. CSSV (strain CSSV-1A) has the distinction of being the first cocoa pathogen to have its genome completely sequenced (Figure 3) (7161 bp - ca. 0.0001% of the size of the human

Figure 3. Map Of The Genome Of The Cocoa Swollen Shoot Virus



Genome organisation of the CSSV-Agouli based on the published nucleotide sequence (Hagen *et al.*, 1993; modified). The open reading frames (ORF) are indicated by long arrows. The predicted molecular weight (in kDa) of the proteins encoded by the individual ORF's is stated in parentheses under the ORF designation. The location of the primers (P1 to P4, P6, P9, P12 and P13) used for PCR amplification are shown by arrowheads outside the DNA molecule. The black and grey regions within the outer circle denote the location of the viral DNA of pCSSV9 (Maiss, unpublished) and that of the genome part sequenced after PCR respectively. Reproduced from (Hoffmann *et al.*, 1997).

genome!). This was achieved by Hagen *et al.* (1993) and the complete sequence can be viewed at <http://www.ncbi.nlm.nih.gov/Entrez/nucleotide.html> (type accession number: L14546). Five putative protein coding regions (open reading frames or ORFs) were identified within the genome, including a reverse transcriptase (for replication of the viral genome via RNA) and a coat protein.

By combining immunological purification of viral particles with PCR amplification (the IC-PCR method; immunocapture-PCR) based on information from the complete genome sequence, Hoffmann *et al.* (1997) have devised an detection assay for identifying latent infection. However, this method is effective only for the CSSV-1A serotype and it remains to be established whether this method is more sensitive than the VBA test of Hughes and Ollennu (1993). Nevertheless, a significant degree of sequence polymorphism was detected within the ORF3 gene of CSSV-1A strains indicating a high level of genetic diversity within CSSV populations.

It has been proposed by Hughes and Ollennu (1994) and others that, in the light of the incomplete effectiveness of other control strategies it may be feasible to 'immunise' trees against infection by virulent strains of CSSV by inoculation with attenuated ('mild') strains of the virus. Some symptoms would be caused by these 'mild' strains but it is anticipated that overall crop losses would be reduced (Ollennu *et al.*, 1994a; Ollennu *et al.*, 1994b). However, such a strategy must be adopted with great care, since it must first be established that the cross-protection offered by 'mild' strains will be effective in controlling all the more virulent CSSV strains and also that 'mild' strains once inoculated will be able to evolve higher levels of aggressiveness. Further, strain characterisation or even the construction of transgenic attenuated strains suggests a central role for molecular biology in these developments.

FUTURE DEVELOPMENTS

The progress outlined above has hitherto be made predominantly in the areas of discrimination and detection. Future investigation may use similar techniques to analyse in detail the process of pathogenesis (particularly in the case of WBD and CSSV, as described above) such that through a better understanding of the pathogens physiology, specific control strategies can be devised.

It is also important to consider the progress being made by plant geneticists and breeders. The advent of marker-assisted selection will ultimately speed up the process of plant breeding by identifying quantitative traits associated with degrees of resistance to certain pathogens. Again, the knowledge currently being obtained with regard to pathogen variability will play an important role in these breeding programme by permitting pathogen populations to be closely monitored. Although antibody-based ELISA assays (as used in pregnancy testing kits etc.) would provide a useful tool for cocoa farmers in accurate on-site disease diagnosis, the requirement for animal experimentation in the production of such antibodies can render such approaches ethically unacceptable. However, the combination of molecular genetics with immunology is likely to bypass such issues, as has recently been shown by He and Taussig (1997) who have developed an *in vitro* method for antibody selection.

The final area of advance is likely to stem from the interaction between molecular biology and information technology. The release of DNA sequence data is now a prerequisite for the publication of results and as a result substantial databases of both sequence and other data (for example, information on the International Cocoa Germplasm Database can be found on

the ICGD website - http://www.icgd.rdg.ac.uk/netscape_index.htm) are now in the public domain and readily available via the Internet. Comparison of data between labs in different continents and the use of standardised methods for monitoring pathogen populations will thus be greatly facilitated.

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Appendix I: Description of PCR Methodologies

The polymerase chain reaction (PCR) is by now extensively used as a tool for identification of microbial pathogens. Only nanogram ($1 \text{ ng} = 10^{-9} \text{ gram}$) quantities (or less) of DNA as a template for amplification (which can be isolated from milligram or microgram quantities of biological material) are needed and the process can be made sufficiently specific that the DNA used need not necessarily be derived from a pure culture. It is thus possible to design methods of detection that are both highly sensitive (in theory able to detect as little as a single copy of the target DNA sequence), yet highly specific.

A technique that has been used extensively to date by plant pathologists is RAPD (Random Amplified Polymorphic DNA). In contrast to most applications of PCR, this technique does not require any *a priori* DNA sequence information about the organism being studied. It also only requires a single, short (usually 10 bp) random primer (a primer is a short single-stranded DNA sequence manufactured synthetically and which sticks to matching sequences on the target DNA to 'prime' DNA amplification) is used in the PCR reaction. By using a low primer annealing temperature, multiple-sized bands are amplified and these can be used as a 'bar-code' to type isolates. Similarity indices can be generated for comparison by scoring the frequency of shared bands between isolates. However, the technique has been bedevilled by difficulties in standardisation and there are many examples in the literature of different banding patterns being obtained from identical biological samples by different laboratories or when minor alterations are made to the DNA isolation or amplification methodology (e.g. McEwan *et al.*, 1998).

Most applications of PCR rely on the use of two short synthetic pieces of DNA (called 'primers', usually ca. 20 basepairs long and costing under £10 each to purchase) which match the two ends of the target region of DNA. The specificity of the PCR amplification reaction relies on designing suitable primers to avoid non-specific amplification of any DNA except the desired sequence. The design of these primers requires at least some DNA sequence information about the organisms under study. With the exception of a few 'model' organisms (e.g. humans, yeast, *E. coli* etc.), the amount of DNA sequence information for most organisms is limited or absent. However, primers can be designed using sequence information from related organisms, provided that reasonably conserved regions of DNA are chosen for study.

The most widely used target sequence for PCR-based studies in plant pathology is the rDNA (ribosomal DNA) locus. The rDNA locus is present in all living organisms (since all contain ribosomes) and certain regions of the locus are highly conserved between distantly related organisms. For instance, primers pairs have been designed which amplify parts of the rDNA locus from any fungi (White *et al.*, 1990). Once the PCR product is produced, genetic differences can be revealed either by restriction digestion of the amplified product or by sequencing all or part of the product. Cooke and Duncan (1997) have produced a phylogenetic tree from the genus *Phytophthora* based on rDNA sequences. Although PCR amplification of DNA from pure cultures works well with such primers, they are often non-specific and are thus of limited use for amplification of DNA from complex substrates. In disease lesions, for instance, it is possible that rDNA from other organisms (e.g. the host plant or contaminating fungi) may be also amplified in addition to that of the pathogen.

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