Review Paper

Over seventy years of a viral disease of cocoa in Ghana: From researchers' perspective

H. Dzahini-Obiatey*, Owusu Domfeh and F. M. Amoah

Cocoa Research Institute of Ghana, P. O. Box 8, Akim Tafo, Ghana.

Accepted 8 January, 2010

Virus diseases have plagued cocoa (*Theobroma* cacao) production in West Africa for over seven decades. Principal among them is that caused by the cocoa swollen shoot virus (CSSV), which is endemic in Togo, Ghana and Nigeria, and more recently Cote d'Ivoire. The incidence of the disease in Ghana has led to the launch of the costliest and an over ambitious eradication control programme in the world. This review highlights the various research activities conducted mainly in Ghana that influenced the various control strategies as well as those that have the potential to influence future ones. Isolation of newly planted cocoa has been identified as an efficient method of reducing CSSV prevalence and spread in the field. Identification of mealybugs as vectors, the role of alternative host in the spread of the disease, the need for an urgent review of the eradication procedures, breeding specifically for resistance to CSSV as well as some biochemical and molecular biology studies are some of the points highlighted in this paper. The achievements and limitations made in these fields are duly emphasized. The way forward, however, will be to combine most of these strategies into a single or two integrated approaches to control cocoa swollen shoot virus disease (CSSVD). This will then be in tune with the suggestion that no single measure is adequate to solve the swollen shoot disease problem in Ghana, and the rest of West Africa.

Key words: Eradication campaign, mild strain, cross-protection, CSSV, mealybugs, isolation, barriers, non-host crops, molecular biology, biochemistry.

INTRODUCTION

Four types of viral diseases of cocoa (*Theobroma* cacao) occur in Ghana and other cocoa producing countries of West Africa, namely, Cote d'Ivoire, Nigeria and Togo. Two, the cocoa swollen shoot and the mottle leaf viruses are transmitted by mealybugs but the vectors of the other two, cocoa yellow mosaic and cocoa necrosis viruses, remain unknown (Kenten and Legg, 1971). The most economically important and the most studied, the cocoa swollen shoot virus (CSSV) will form the basis of our discussion in this review. The discussion will, however, be mainly focussed on the experiences in Ghana, since most of the work on the virus was done in Ghana. Also, Ghana has had the most dramatic experience with the virus, particularly in the eradication (cutting out)

Abbreviations: CSSV; Cocoa swollen shoot virus.

campaign to control the disease. This control method, which was launched in Ghana in 1946, has been considered as the most ambitious and costliest eradication campaign to control a plant viral disease anywhere in the world (Thresh et al., 1988a). The lost in monetary terms of dead trees is huge since the last estimate put the total number of trees claimed by the disease then at over 200 million trees (Ollennu et al., 1989a, 1999; Dzahini-Obiatey, 1993, 2008), and this in cash could be several millions of US dollars. This lost of income impacts negatively on government through loses in tax revenue from cocoa, which could have been used for development projects. For the peasant cocoa farmers, the effect is even more devastating since whole fields could be lost to the disease, hence, their regular source of income could be curtailed. In the case of settler farmers, things can even get worse due to the likelihood of the landlords taking back their land once the cocoa is gone. Yet the disease is still as prevalent as ever (Figure 1).

Research work on CSSVD or its pathogen, CSSV, has

^{*}Corresponding author. E-mail: h.k.dobiatey@googlemail.com.



Figure 1. Scores of CSSV-infected trees dying back in new outbreak areas in the Western Region of Ghana. Parts of the said Region now have areas where the disease is so common that it is termed an area of mass infection (AMI).



Figure 2. Young cocoa leaves of CSSV infected cocoa showing red vein banding: A = relatively younger leaf B = relatively older leaf.

been reviewed under various topics (Owusu, 1983; Thresh and Owusu, 1986; Thresh et al., 1988 a, b; Ollennu et al., 1989 a, b; Hughes and Ollennu, 1994; Dzahini-Obiatey et al., 2003; Dzahini-Obiatey, 2008). The present review attempts to tackle all the disciplines together and to suggest the way forward in the management of the disease. However, this review will be limited to studies that have led to key findings that have contributed to the present control strategies or those that have the potential to similarly influence future strategies. The eradication campaign and procedures will also be reviewed and recommendations for future control strategies made. Topics discussed include epidemiology, CSSV Management, biochemical and molecular biology.

HISTORY AND DISCOVERY OF COCOA SWOLLEN SHOOT DISEASE

The first record of the swollen shoot and dying back disease was made in Ghana in 1936 (Annonymous, 1936, Steven, 1936) after a plant pathologist had made a detailed description of a reported disease by a farmer. The actual pathogen causing the disease was, however, not identified until Posnette (1940) through experimenttation, attributed the disease to a virus, subsequently named CSSV. Subsequent tests identified its vector as mealybugs (Homoptera: pseudococcidae) (Box, 1945) and later the other modes of transmission were found (Archibald, 1954). Before identifying the pathogen as a virus, recommendations were made to cut and burn all affected trees and their adjoining contacts (Steven, 1936). Thereafter, a systematic survey was made in 1944, which found the virus to be widespread throughout the Country and by 1947 it was estimated that 50 million trees had been affected of which 45 million were in the Eastern Region alone (Thresh et al., 1988a). An eradication campaign to control the disease was officially launched in 1946 and backed by compulsory legislative powers in 1947. The campaign has remained in place ever since, albeit with sporadic reviews and the injection of new ideas.

The disease was later discovered in other West African cocoa producing countries like, Cote D'Ivoire, Nigeria, Sierra Leone and Togo. Neither the disease nor its pathogen has been reported in the Amazon Basin (South America), where cocoa was purported to have been brought from into West Africa.

PATHOGEN AND SYMPTOMS

CSSV or the pathogen of cocoa swollen shoot virus disease has recently been characterised as having a double-stranded circular DNA genome (Lot et al. 1991). It was subsequently been assigned to the genus badnavirus with the family Caulimoviridae. In Ghana, the pathogen or virus was also isolated from alternative host trees such as Cola chlamydanta, Ceiba pentandra, Adansonia digitata, Cola gigantean and Sterculia tragacantha. CSSV exhibits myriads of symptoms in the cocoa plant. These include red vein banding in young leaves (Figure 2), different shades of chlorosis in mature leaves (Figures 3 and 4), swellings on stem or chupons (Figure 5), and on roots (Figure 6). Most of these symptoms are guite unique to isolates/strains and hence form the basis for identification. and limited characterization of some strain/isolates.





в

Figure 3. CSSV infected cocoa leaves showing fern pattern form of chlorosis with A clearing along major veins and B green vein banding along major veins.





Figure 1. CSSV infected cocoa leaves showing vein clearing.

EPIDEMIOLOGY

Vector-host-pathogen relationships

Important features of epidemiology of the swollen shoot disease became apparent just upon observation of the

first outbreaks even before any decision was made to carry out detail studies. It was noted that outbreaks enlarged and eventually coalesced to form areas of mass infection. The identification of mealybug species as vector (Box, 1945) invigorated studies that explicated the characteristic pattern of spread of the swollen shoot disease. Radial spread over short distances around the periphery of outbreaks was thus attributed to mealybug vectors "walking" between the interlocking branches of adjacent trees (Strickland, 1951; Cornwell, 1958). New outbreaks were shown to be occasioned by "jump spread" over greater distances by wind borne viruliferous mealybugs and mainly by the very active small first instar nymphs (Strickland, 1950; Cornwell, 1960).

Later epidemiological experiments confirmed the circumscribed nature of spread of the swollen shoot virus leading to the proposition of infection gradients. The observations were that infections are greatest around the periphery of the plantings and decrease or are few internally (Benstead, 1951; Thresh, 1958; Thresh and Lister, 1960; Legg and Lockwood, 1981; Ollennu et al., 1989a; Dzahini-Obiatey et al., 2006a). These observations were made at different times, and at different locations in Ghana and Nigeria. It was also noted that the incidence of infected trees and the distances over which they occurred were greater around larger outbreaks than around smaller ones. These findings informed the recommendation and adoption of drastic eradication procedures based on outbreak sizes practised in Nigeria and Ghana (Thresh et al., 1988a; Ollennu et al., 1989b) and the proposition to isolate new plantings from the boundaries (Dzahini-Obiatey et al., 2006 a, b).

Isolation

It has been suggested that short isolation distances can be effective in controlling typical "crowd diseases" (Vanderplank, 1948). However, little attention was given to the benefits of isolation to control CSSVD in farmers' farm. This is unfortunate because records abound, which suggested the advantages to be derived if at least some degree of isolation could be provided between all new plantings and the older cocoa, especially, in those areas where infection is rife (Posnette, 1951a, Thresh et al., 1988 a, b; Ollennu, 1988; Owusu and Ollennu, 1997; Dzahini-Obiatey et al., 2006 a, b). The findings that feeding insects transmit CSSV in semi-persistent mode (Posnette et al., 1950) therefore not only lend credence to the above assertions but also help to explain the slow rate of spread and the infection gradient proposition. One possibility for creating the isolation would be to discourage or prohibit the current practice of planting to the boundary of the land available. Thus, an isolation distance of 10 m around all new plantings was recommended in Nigeria (Are, 1969), and at a much later date in Ghana (Ollennu et al., 1989a). Another possibility is to introduce barriers with non-host crops around the



Figure 5. CSSV infected cocoa trees having large stem swellings on rejuvenated stem and chupon.



Figure 6. Roots from CSSV infected cocoa tree with a swelling shown by red arrow.

new plantings. The choice of non-host crops, their economic benefits and effectiveness in curbing the spread of the virus to new cocoa plantings would however have to be investigated and evaluated before recommendations are made to farmers (Thresh et al., 1988a). Studies to evaluate effectiveness and the economic returns of using some selected plants as barriers to delimit new plantings from the boundaries, which often have old, abandoned and infected cocoa started in 1993 (Ollennu et al., 1995; Dzahini-Obiatey et al., 2006 a, b; Dzahini-Obiatey, 2008). The studies were on three economic crops, citrus, kola and oil palm, which were selected based on their potential of earning the farmer similar incomes to the cocoa he would have planted close to the boundaries. The latest report from the studies showed citrus and oil palm to be effective barriers for preventing early re-infection into newly

established cocoa from adjoining old plantations (Ollennu et al., 2003). The economic analysis on the returns from these two crops has shown them to be equally profitable as cocoa (Ollennu et al., 2005), thus, giving an added advantage to the recommendation for planting them as barriers to prevent re-infections.

The land tenure system in Ghana is not helping matters either. Lands have become more fragmented due mainly to inheritance and land leasing arrangements where the already small cocoa farms are further divided among the family members upon the death of the owner or between the care taker and the land lord (Thresh et al., 1988a). This has made the management of the swollen shoot disease by treatment and the isolation of replanted farms more difficult. Treatment of infected trees is done with the consent of all the farmers and by these fragmentations more people become involved and contacting them to get their consent to treat their farms becomes even more daunting. It has been suggested (Thresh, et al., 1988a) that treating and replanting of small farms (less than 0.04 ha) may not be advisable since virtually all the replanted trees in such farms may be vulnerable. This gives credence to the idea that vast lands under one management may be required to manage the disease more efficiently. Unfortunately, such types of lands are rare in Ghana. An urgent review of land tenure and usage in Ghana is very imperative. The recent proposition for land banks is long overdue. The establishment of a system where individuals, families, chiefs, etc, could pool their lands together for a specific usage and under one management will be a good idea (Dzahini-Obiatev et al., 2006a). Each banked land could be valued and a bond or share certificate issued to the owner according to the size of the plot and the returns of whatever project is undertaken on the land shared equitably among the share or bond holders (Dzahini-Obiatev et al., 2006a). Through such arrangements, adequate lands will become available for specific projects that will include rehabilitation of the swollen shoot endemic lands anywhere in Ghana.

Host range studies

Host range studies also contributed to the swollen shoot disease control by providing information that explicated the origin of swollen shoot disease in Ghana and Nigeria. The studies showed that Western Ghana strains of CSSV might have originated from forest trees. Cola chlamydantha trees were frequently found infected with CSSV both in cocoa farms and in forest reserves several miles away from cocoa (Posnette et al., 1950; Todd, 1951; Tinsley, 1955; Dale and Attafuah, 1957). Indeed, about a tenth of these naturally infected C. chlamydantha trees were found in areas where cocoa was rare and not infected. Swollen shoot disease was thus prevalent in farms with C. chlamydantha in and around them than those without (Tinsley, 1971). Other forest trees like, Sterculia tragacantha (Legg and Agbodjan, 1969; Owusu and Lovi, 1970), Adansonia digitata (Attafuah and Tinsley, 1958) and Ceiba pentandra were other likely sources of CSSV into cocoa in other Regions of Ghana, Togo and Nigeria. Removal of such trees was thus recommended as part of the control measures for the disease.

CSSV MANAGEMENT

Since the discovery of CSSV, several interventions were introduced, investigated and/or put in place to manage the virus or the disease it causes. Notable among these are the "Eradication Campaign", breeding for resistance to the virus and mild strain cross-protection.

Eradication campaign

A typical crowd disease like the swollen shoot disease of cocoa could be amenable to control by eradication if properly done. The history of swollen shoot disease control in Ghana is however different. Since its official launching in 1946, the eradication campaign has been fraught with many disruptions and discontinuities (Ollennu et al., 1989b; Dzahini-Obiatey et al., 2006b) and the eradication procedure itself has been full of deficiencies. For example, the initial procedure removed only trees with symptoms even though the value of a more drastic procedure of removing the adjoining contact trees as well was fully appreciated; the drastic procedure was delayed until 1957 (Thresh et al., 1988a). Similarly, the replanting of new cocoa in the treated areas was also done very close to older and often infected trees in the boundaries even though the benefits of isolating new plantings were known. This unfortunate situation has continued up to date and has generated several reviews in memoranda, reports and scientific articles (Owusu, 1983; Thresh and Owusu, 1986; Thresh et al., 1988a; Ollennu et al., 1989b, Dzahini-Obiatey et al., 2006 a, b).

A new control strategy of combining eradication with isolation is being proposed for the Western Region of Ghana where vast cocoa lands still await rehabilitation from CSSV (Dzahini-Obiatey et al., 2006 a, b). Treated and replanted farms will have to be isolated with barriers of CSSV immunce crops such as citrus (Figure 7) or oil palm. Where adequate land is available, the replanting can be done in large blocks as was done for the Suhum Cocoa Project, but with a barrier of non-host crops planted around them and with more tolerant or resistant cocoa.

Breeding for resistance against cocoa swollen shoot virus disease

Cocoa swollen shoot disease in Nigeria was partly managed by replanting with tolerant hybrids (Are, 1969). In Ghana, although it has been suggested that using tolerant or resistant varieties could control the disease, no suitable variety has yet been found (Kenten and Legg, 1971; Thresh et al, 1988b). Attempts to control the disease through breeding started with the introduction of the Upper Amazon varieties (Posnette, 1951b) but these varieties were soon found to be of no immediate practical value due to their very low level of resistance or tolerance (Posnette and Todd, 1951). The F3 Upper Amazon and the series II hybrids (Upper Amazon x Amelonado and Upper Amazon x Trinitario) used in new plantings and in replacing Amelonado trees destroyed by swollen shoot disease did fill the gap by performing well against the disease even though none of the parents was specifically selected for resistance/tolerance. Attention was thus given to the T17 trees (Dale, 1957; Attafuah and Glendinning, 1965a) and later to the other Upper



Figure 7. A replanted field having a barrier of citrus crop shown by red arrows and a replanted cocoa tree shown by a white arrow. Shade plants such as plantain, *Glicidia* and *Papaya* can also be seen in the background.

Amazons (Attafuah and Glendinning, 1965b) and the emphasis in breeding was switched to selecting for tolerance.

Breeding for resistance again became the main focus of the research activities only after the arrival of the British Research Team (BRT) between 1969 and 1978 (Legg, 1981). Through their efforts, the inter-Upper Amazon hybrids were released in 1986. These hybrids were generally more resistant to infection than the equivalent Series II hybrids (Legg and Lockwood, 1981). Most of the inter-Upper-Amazon hybrids were subsequently used in field trials against CSSV where their apparent higher level of resistance again became evident; even after 20 - 30 years in the neighbourhood of infected cocoa (Ollennu et al., 1989a; Ollennu and Owusu, 2002). Recent efforts to identify CSSV resistant/tolerant parents by screening cocoa germplasm material not previously tested for resistance/tolerance to CSSV has started yielding promising results (Adomako et al., 2003). In the laboratory/gauze house-screening test, the best non-Uppper Amazon parents crossed with the best Upper Amazon selections gave hybrids with enhanced resistance to CSSV. It will be useful to test such promising hybrids in the field in order to be able to select the most promising hybrids that can effectively alleviate economic losses caused by the disease. Posnette (1981) commented that probably no other feasible measures would be as effective in reducing the losses caused by the disease as an increase in resistance which would decrease the rate of spread by say 20%. Another method being pursued is the use of mutation breeding techniques such as irradiation of

cocoa to induce resistance (Adu-Ampomah and Owusu, 1993) and the indication is that the procedure can be used to produce materials that have enhanced resistance/tolerance to the disease that can be used in CSSV breeding programmes (Adu-Ampomah et al., 1996). Randomly amplified polymorphic DNA (RAPD) analysis using PCR can reveal DNA based polymorphism between organisms both at the species and at the individual levels and this can be used to accelerate selection processes during breeding for resistance to the swollen shoot disease. RAPD analysis has the potential for establishing a unique fingerprint for an individual cocoa tree. More importantly, a sub-species, variety, genotype or breeding line can be uniquely characterized. Already a low-density linkage map of Theobroma cacao has been constructed using RAPD markers and an anthocyanin biosynthetic locus (Osei et al., 1995). This knowledge base is being explored in the cocoa swollen shoot virus disease management.

A more recent effort to explore resistant markers in CSSV infected cocoa, has led to the identification of some cellular modifications, which were found to be associated with CSSV infection (Dzahini-Obiatey and Fox, 2006, 2009). Notable among them was the nucleic acid rich inclusion bodies (Figure 8), which will have to be explored as a marker among CSSV-resistant cultivars of cocoa. With the type of outbreaks and number of infections per outbreak being encountered in the Western Region of Ghana, an area hitherto designated as an area of "scattered-outbreaks" due to the fewer outbreaks and infections, breeding for resistance to the disease will be the most plausible proposition to lower the rate of spread.



Figure 8. Periodic acid Schiff toluidine blue stained inclusion bodies observed in CSSV 1 A infected tissue. Note the spherical-shaped inclusion bodies (shown by arrows). They vary in sizes and numbers per cell. They were subsequently tested and found to contain nucleic acids (Dzahini-Obiatey and Fox, 2006).

Although eradication of swollen shoot disease is still ongoing in these areas, replanted cocoa needs to be cordoned with barriers of CSSV non-host crops as recommended (Ollennu et al., 2005). Planting of new cocoa very close to the boundaries of older and obviously infected cocoa should be avoided, and the recommended distance of 10 m (Thresh et al., 1989 a, b; Ollennu 1988; Ollennu et al., 1989; Owusu and Ollennu, 1997; Dzahini-Obiatey et al., 2006) should be left around all newly planted cocoa in the CSSV endemic areas. This message should be disseminated through a strong link between research and farmers. An integrated approach of planting of hybrids purposely selected for resistance/ tolerance to CSSV, coupled with planting of the selected materials in blocks and isolating the blocks with barriers of non-host crops would be a more effective panacea.

Mild strain cross-protection

Attempts to contain the swollen shoot disease by eradication measures have been costly and have led to the diversion of some resources which could otherwise be used to improve standards of husbandry and hence raising cocoa output or for improvement elsewhere in the agricultural sector (Owusu et al., 1996). Nevertheless, no satisfactory control has been achieved and farmers are disillusioned and desperately looking for solutions. Breeding for resistance to the disease has not been very rewarding either, particularly in areas where the swollen shoot disease is rife. It is with these experiences that it was suggested that no single measure is likely to solve the swollen shoot disease problem and that several measures will have to be taken together (Owusu, 1983; Owusu et al., 1996).

Investigations into mild strain cross-protection therefore started with this conviction. Earlier observations by Posnette and Todd (1955) that mild strain of CSSV could protect cocoa against the effect of the severer strains, was thus reviewed and pursued further (Ollennu et al., 1989b; Owusu et al., 1996; Ollennu et al., 1999). These investigations culminated in the identification of two isolates CSSV N1 and CSSV 365B, which are capable of protecting cocoa plants from the adverse effects of the virulent 1A type isolates (Ollennu et al. 2003). Current data from mild-strain cross-protection studies has shown two progenies of cocoa, that is T85/799 x T79/501 and T85/799 x Pa7/808, to be suitable for use in the mild strain cross-protection work (Domfeh et al., 2009). This result reaffirmed the reported high yielding nature of T85/799 X Pa7/808 in the presence of CSSV N1 mild strain (Ollennu and Owusu, 2003). It is envisaged that this mode of control will be suitable only in the "areas of mass infection" (AMI) in the Eastern and Western Regions of Ghana, where the disease is so widespread.

BIOCHEMICAL AND MOLECULAR STUDIES OF THE VIRUS

The bulk of research work on CSSV is in the disciplines of biochemical and molecular biology of the virus. Although the immediate application of some of these works in the swollen shoot disease control is not too obvious, their future field application potential is without any doubt. The success of the first biochemical purification of the virus (Brunt and Kenten, 1960) generated interest for more work, and by 1965, several improvements had been made in the purification procedures (Brunt and Kenten, 1962, 1963; Brunt et al., 1964; Kenten and Legg, 1965) to a point where infectivity rates as high as 90% were frequently attained. The successful purification of the virus enabled mechanical (manual) inoculation to be used in screening plant materials during resistant breeding experiments and this was shown to be better than mealybug inoculations (Kenten and Legg, 1971). Improvement in the purification procedures also showed the virus to consist of bacilliform particles (Brunt et al., 1964; Adomako et al., 1983). Other biochemical studies of the virus led to the production of antiserum for the development of several serological methods like enzyme linked immunosorbent assay (ELISA) (Sagemann et al., 1983, 1985; Dzahini-Obiatey et al., 2002), immunosorbent electron microscopy (ISEM) (Sagemann et al., 1985) and virobacterial agglutination test (VBA) (Hughes and Ollennu, 1993) for the detection of the virus. Some of these serological methods may have high diagnostic potentials for use in the field. Effective diagnosis of CSSV in the field will enhance the screening techniques in resistant breeding experiments and hence accelerate the process of identifying resistant/tolerant varieties. Its use in the field will also speed up the detection of re-infections after eradication so the effective remedies can be planned quickly.

Works on molecular studies of cocoa viruses have centred on the nucleic acid and proteins of CSSV. A cocoa swollen shoot virus isolate was first determined to be a double-stranded DNA virus with an approximate genome size of 7.4 kb by using nuclease activity with gel electrophoresis, restriction enzyme digest and Southern blot analysis (Lot et al., 1991). The genome was thereafter cloned and sequenced to determine segments that encode for specific activities such as the cell-to-cell movement and coat protein gene (Hagen et al., 1993). The full-length genomes were also subsequently developed into infectious clones (Hagen et al., 1994).

Noting that CSSV is a DNA virus, Sackey et al. (1995a) also started parallel studies on the numerous Ghanaian isolates of CSSV that apparently vary from the Agou 1 strain [the isolate from which the first clone was made (Lot et al., 1991; Hagen et al., 1993, 1994)]. Nucleic acid techniques such as polymerase chain reaction (PCR) (Sackey et al., 1995b; Dzahini-Obiatey et al., 1996c), DNA hybridisation with radioactive (Dzahini-Obiatey, 1993; Sackey and Hull, 1994) and non radioactivelabelled probes (Sackey et al., 1995b, 1996b, 1999) and a combination of PCR and DNA hybridisation (Sackey et al., 1995 a, b, c, 1996 a, b) were employed to study the Ghanaian isolates of CSSV. Limited classification of some isolates was thus achieved. Some of these techniques can now be used in routine diagnosing and further characterization of the numerous CSSV isolates in Ghana. Dot blot hybridisation technique in particular can have large-scale application in the field and can thus fit into CSSV control programmes. Low yield of CSSV DNA from

cocoa tissues usually encountered during extraction will however have to be addressed before nucleic acid techniques in general can become useful for field applications. Attempts are currently underway to overcome this problem by finding other ways of extracting CSSV DNA as well as looking for other tissues for culturing CSSV (Dzahini-Obiatey et al., 2001). Another possibility is to clone the CSSV DNA before any biochemical manipulations are done. A very recent work done using microscopic and molecular techniques has determined the cotyledons of CSSV-infected cocoa to have contents of the virus for most analytical work (Dzahini-Obiatey and Fox, 2009). This information will be exploited in the bid to identify resistant markers for breeding purpose.

THE FUTURE

Cocoa swollen shoot virus disease remains by far the greatest challenge by a viral disease to cocoa production in West Africa. This review has highlighted the major efforts made at developing or improving upon existing methods aimed at controlling the disease since its discovery in Ghana and the other West African producing countries. The critical question is that: are the various control strategies in place meeting the guest to find a lasting solution to the disease? All the control strategies, that is the present and future will have to be critically reviewed or thought through carefully. Eradication and replanting may have to be limited to areas where it can be done well, especially, in areas where new plantings can conveniently be isolated with barriers of non-host crops. The Western Region of Ghana where vast infected cocoa lands exist and await rehabilitation may be the ideal place to implement these proposals. New and resistant varieties to the swollen shoot disease could also be bred specifically for the disease endemic areas and these could be used in conjunction with the isolation method mentioned above. The ultimate in the future strategies will be to produce genetically engineered plants by introducing the resistant genes against the virus through non-conventional cocoa breeding into techniques.

ACKNOWLEDGEMENT

We are grateful to colleagues for their critical comments on the manuscript. This paper (Ref. CRIG/05/2009/038/ 002) is published with the kind permission of the Executive Director, Cocoa Research Institute of Ghana.

REFERENCES

Adomako D, Lessmann DE, Paul HL, Owusu GK (1983). Improved methods for the purification and detection of Cocoa swollen shoot virus. Annals. Appl. Biol. 103: 109-116.

- Adomako BAY, Ollennu LA, Dzahini-Obiatey H, Takrama JF (2003). Breeding for cocoa varieties resistant/tolerance to cocoa swollen shoot virus (CSSV) (STABEX) (EU FUNDED) PROJECT). Progress Report for 2002-2003, Cocoa Research Institute of Ghana pp. 201-203.
- Adu-Ampomah Y, Owusu GK (1993). Induction of cocoa swollen shoot disease resistant mutants by gamma irradiation. Proceedings of the 11th International Cocoa Research Conference, Yamoussoukro, Cote D'Ivoire, 18th - 24th July, 1993.
- Adu-Ampomah Y, Owusu, GK, Sackey S, Padi B, Abdul-Karimu A (1996). Use of gamma rays to induce mutants resistant to cocoa swollen shoot disease in *Theobroma cacao* L. Plant Breeding 115: 74-76.
- Annonymous (1936). A new disease of cocoa in the Gold Coast. Gold Coast Farmer 5: 122.
- Archibald JF (1954). Botany and Physiology. Ann. Rep. W. Afr. Cocoa Res. Inst. 1953-1954 p. 22.
- Are LA (1969). Rehabilitation of cocoa farms. Cocoa Growers Bulletin 13: 11-13.
- Attafuah A, Glendinning DR (1965a). Studies on resistance and tolerance to cocoa varieties in Ghana. I. A survey of the T17 progeny. Annals. Appl. Biol. 56: 219-225.
- Attafuah A, Glendinning DR (1965b). Studies on resistance and tolerance to cocoa varieties in Ghana II A survey of breeding material. Annals. Appl. Biol. 56: 227-230.
- Attafuah A, Tinsley TW (1958). Virus disease of *Adansonia digitata* L. (Bombacaceae) and their relation to cacao in Ghana. Annals. Appl. Biol. 46: 20-22.
- Benstead J (1951). Cocoa re-establishment. Rep. Cocoa Conference pp. 111-115. Cocoa, Chocolate and Confectionery Alliance, London.
- Box HE (1945). Insect transmission of the "swollen shoot" virus in West African cacao. Nature 155: 608-609.
- Brunt AA, Kenten RH (1960). Mechanical transmission of cocoa swollen shoot virus. Virol. 12: 328-330.
- Brunt AA, Kenten RH (1962). The mechanical transmission of cocoa swollen shoot virus to and from cocoa and other hosts. Annals. Appl. Biol. 50: 749-754.
- Brunt AA, Kenten RH (1963). The use of protein in the extraction of cocoa swollen shoot virus from cocoa leaves. Virol. 19: 388-392.
- Brunt AA, Kenten RH, Nixon HL (1964). Some properties of cocoa swollen shoot virus. J. Gen. Microbiol. 36: 303-309.
- Cornwell PB (1958). Movement of vectors of virus disease of cacao in Ghana. I. Canopy movement in and between trees. Bull. Entomol. Res. 49: 613-630.

Cornwell PB (1960). Movement of vectors of virus disease of cacao in Ghana. II. Wind movement and aerial dispersal. Bull. Entomol. Res. 51: 175-201.

- Dale WT (1957). Studies on resistance and tolerance to cacao viruses. In: Proceedings Cacao Breeding Conference 1956 pp. 3-6. Tafo, Ghana: West African Cocoa Research Institute.
- Dale WT, Attafuah A (1957). Virus research: the host range of cocoa viruses. Report of the West African Cocoa Research Institute for 1955-1956, pp. 28-30.
- Domfeh O, Dzahini-Obiatey H, Ameyaw Akumfi G, Opoku IY (2009). The effect of mild strains on growth and yield of cocoa. 2008/2009 Progress Report and Work plan, Cocoa Research Institute of Ghana, July 2009, Akim Tafo pp. 151-153.
- Dzahini-Obiatey H (1993). The development and use of molecular biological techniques in the diagnosis of cocoa swollen shoot virus (CSSV): Application to viruses from Malaysian cocoa leaves and *Commelina* species from cocoa fields in Ghana. M.Sc. Thesis, University of East Anglia, Norwich, UK.
- Dzahini-Obiatey H (2008). Cytopathological and Molecular studies of Cacao swollen shoot Badnavirus (CSSV) infected cocoa plants. PhD thesis, University of Reading.
- Dzahini-Obiatey H, Fox RTV (2006). Early developments in hostpathogen interaction between *C*acao swollen shoot virus (CSSV) and the cocoa plant. Proceedings of the 15th International Cocoa Research Conference, Herradura Plazza Hotel, San Jose, Costa Rica, October 9 -14, 2006 pp. 885-897.
- Dzahini-Obiatey H, Fox RTV (2009). Early signs of infection in cacao swollen shoot virus (CSSV) inoculated seeds, and the discovery of

- the cotyledons of the resultant plants as rich sources of CSSV. Afr. J. Biotechnol. (In press).
- Dzahini-Obiatey H, Sackey ST, Lowor ST, Donkor AQ (1996). Poymerase chain reaction. Rep. Cocoa Res. Inst. Ghana 1995/1996 pp. 163-164.
- Dzahini-Obiatey H, Aculey PC, Owusu GK (2001). An alternative source for the extraction and purification of cocoa swollen shoot badnavirus (CSSV). J. Ghana. Sci. Associ. 3(2): 102-109.
- Dzahini-Obiatey H, Aculey PC, Takrama JF, Lowor ST, Ollennu LA (2002). A method for the elucidation of pathogenic related proteins of cocoa swollen shoot badnavirus (CSSV). J. Ghana. Sci. Assoc. (In press).
- Dzahini-Obiatey H, Ollennu LA, Aculey PC (2003). Cocoa swollen shoot virus in Ghana: A review of diagnostic procedures. J. Ghana. Sci. Assoc. (In press).
- Dzahini-Obiatey H, Akumfi Ameyaw G, Ollennu LA (2006a). Control of cocoa swollen shoot disease by eradication of infected trees in Ghana: a survey of treated and replanted. Crop Protection 25: 647-652.
- Dzahini-Obiatey H, Ameyaw Akumfi G, Ollennu LA (2006b). Eradication of cocoa swollen shoot disease in Ghana: Is the battle being won or lost? Proceedings of the 15th International Cocoa Research Conference, San Jose, Costa Rica, October 9 -14, 2006 pp. 899-906.
- Hagen LS, Jacquemod M, Lepingle A, Lot H, Tepfer M (1993). Nucleotide sequence and genome organisation of cacao swollen shoot virus. Virol. 196: 619-628.
- Hagen LS, Lot H, Godon C, Tepfer M, Jacquemond M (1994). Infection of *Theobroma cacao* using cloned DNA of cacao swollen shoot virus particle bombardment. Photopathol. 84: 1239-1243.
- Hughes Jd'A, Ollennu LA (1993). The virobacterial agglutination test as a rapid means of detecting cocoa swollen shoot virus. Annals. Appl. Biol. 122: 299-310.
- Hughes Jd'A., Ollennu LA (1994).Mild strain protection of cocoa in Ghana against cocoa swollen shoot virus: a review. Plant. Pathol. 43: 442-457.
- Kenten RH, Legg JT (1965). Observations on the the purification and properties of cocoa swollen shoot virus. Ghana. J. Sci. 5: 221-225.
- Kenten RH, Legg JT (1971). Varietal resistance of cocoa to swollen shoot disease in West Africa. FAO Plant Protection Bulletin 19: 2-12.
- Legg JT (1981). The cocoa swollen-shoot Research Project at the Cocoa Research Institute, Tafo, Ghana 1969-1978. Technical Report Volumes I-IV. London: Overseas Development Administration.
- Legg JT, Agbodjan FX (1969). Swollen shoot disease. Report of the Cocoa Research Institute of Ghana for 1967-1968, pp. 23-25.
- Legg JT, Lockwood G (1981). Resistance of cocoa to swollen-shoot virus in Ghana. I. Field Trials. Annals. Appl. Biol. 97: 75-89.
- Lot H, Djiekpor E, Jacquuemond M (1991). Characterization of the genome of cacao swollen shoot virus. J. Gen. Virol. 72: 1735-1739.
- Ollennu LAA (1988). An update on the swollen shoot disease problem in Ghana. In: Proceedings of the seminar on National Preparedness for Disasters and Workshop on cocoa Pp. 94-106.
- Ollennu LAA, Owusu GK (2002). Spread of cocoa swollen shoot virus to cacao (*Theobroma cacao*) plantings in Ghana. Trop. Agric. 79: 224-230.
- Ollennu LAA, Owusu GK (2003). Field evaluation of the protective capability of CSSV N1 against severe strain New Juaben (1A) isolate. Ghana. J. Agric. Sci. 36: 3-12.
- Ollennu LA, Owusu GK, Thresh JM (1989a). Spread of cocoa swollen shoot virus to recent plantings in Ghana. Crop Protection 8: 251-264.
- Ollennu LA, Owusu GK, Thresh JM (1989b). The control of cocoa swollen shoot disease in Ghana. Cocoa Growers Bulletin 42: 25-35.
- Ollennu LA, Osei BK, Acheampong K (1995). The use of non-host crops as barrier between cocoa plantings. Summary of Thrust Reports 1994-1995. Cocoa Research Institute of Ghana p. 42.
- Ollennu LA, Owusu GK, Dzahini-Obiatey H (1999). Recent studies of mild strain cross-protection with cocoa swollen shoot virus. J. Ghana. Sci. Assoc. (Special Edition) 2(3): 5-11.
- Ollennu L A, Osei Bonsu K, Acheampong K, Aneani F, Ackonor JB (2003). The use of non-host crops as barrier between cocoa plantings. Progress Report 2002-2003, Cocoa Research Institute of Ghana pp. 180-181.
- Ollennu LAA, Osei-Bonsu K, Aneani F, Acheampong K (2005).

- Preliminary studies of the control of cocoa swollen shoot disease by the use of non-host crops as barriers. Proceedings of the 14th International Cocoa Research Conference, Accra, Ghana, 2003 pp. 839-843.
- Osei JK, Furtek DB, Goodin M, Rodriguez H, Lastra R, Fritz PJ ((1995). Construction of a low-density linkage map of *Theobroma cacao* using random amplified polymorphic DNA markers and an anthocyanin biosynthesis locus. *Turrialba* 45(3-4): 128-132.
- Owusu GK (1983). The cocoa swollen shoot disease problem in Ghana: In: Plant Virus Epidemiology (R. T. Plumb and J. M. Thresh eds), Oxford: Blackwell Scientific pp. 73-83.
- Owusu GK, Lovi NK (1970). Study of swollen shoot outbreaks. Report of the Cocoa Research Institute of Ghana for 1968-1969 p. 33.
- Owusu GK, Ollennu LAA (1997). The problem of reinfection of replanted cocoa by cocoa swollen shoot virus in Ghana. Proceedings of the 1st International Cocoa Pests and Diseases Seminar, 1995, Accra, Ghana pp. 179-189.
- Owusu GK, Ollennu LAA, Dzahini-Obiatey H (1996). The prospect of mild strain cross-protection to control cocoa swollen shoot disease in Ghana. Proceedings of the 12TH International Cocoa Research Conference. Salvador, Brazil 18-22: 121-127.
- Posnette AF (1940). Transmission of swollen shoot disease of cacao. Trop. Agric. (Trinidad) 17: 98.
- Posnette AF (1951a). Virus research at the West African Cacao Research Institute. Tropical Agriculture Trinidad 20: 113-142.
- Posnette AF (1951b). Progeny trials with cacao in the Gold Coast. Empire. J. Exp. Agric.19: 242-252.
- Posnette AF (1981). Viruses and resistance to virus disease in cocoa. In: Proceedings 6th International Cocoa Research Conference, Caracas, Venezuela, 1977, pp. 262-266. Nigeria: Cocoa Producers Alliance, Lagos.
- Postnette AF, Todd J McA (1951). Virus diseases of cacao in West Africa VIII. The search for virus-resistant cacao. Annals. Appl. Biol. 38: 785-800.
- Posnette AF, Rorbertson NF, Todd JMcA (1950). Virus disease of cacao in West Africa. V. Alterntive host plants. Annals. Appl. Biol. 37: 229.
- Sackey ST, Hull R (1994). The use of dot blot hybridisation methods to detect cocoa swollen shoot virus isolates. Rep. Cocoa Res. Inst. Ghana 1991/1992 p. 126.
- Sackey ST, Lowor ST, Dzahini-Obiatey H, Owusu GK, Adomako D, Hoffman K, Vettten HJ, Maiss E (1995a). Polymerase chain reaction and immunocapture polymerase chain reaction (ICPCR) for the detection and identification of cocoa swollen shoot virus isolates. Ghana. J. Biochem. Biotechnol. Molecular Biol. 3: 57-66.
- Sackey ST, Bartels PK, Amponsah RK (1995b). Clone PCR amplification products for identification of CSSV 1A and Nsaba by DNA hybridisation. Rep. Cocoa Res. Inst. Ghana 1993/1994 pp. 119-123.
- Sackey ST, Lowor ST, Dzahini-Obiatey HK, Owusu GK, Adomako D, Hoffmann K, Vetten HJ, Mais E (1995c). Polymerase chain reaction and differentiation of cocoa swollen shoot virus isolates. Proceedings of the First International Cocoa Pests and Diseases Seminar. Accra, Ghana, 6-10 November p. 191.

- Sackey ST, Dzahini-Obiatey HK, Owusu, Adomako D (1996a). Variation in cocoa swollen shoot virus isolates. Proceedings of the 12TH International Cocoa Research conference. Salvador, Brazil.18-22 November, 1996 pp. 107-112.
- Sackey ST, Lowor ST, Dzahini-Obiatey H (1996b). Classification of CSSV isolates by PCR and DNA hybridisation. Rep. Cocoa Res. Inst. Ghana 1994/1995 pp. 141-149.
- Sackey ST, Dzahini-Öbiatey H, Lowor ST (1999). The use of cloned DNA probes for the detection of CSSV 1A and CSSV Nsaba. J. Ghana. Sci. Assoc. (Special Edition) 2(3): 102-109.
- Sagemann W, Paul HL, Adomako, Owusu GK (1983). The use of enzyme linked immunosorbent assay (ELISA) for detection of cacao swollen shoot virus (CSSV). *Phytopathologische* Zeitschrift 106: 281-284.
- Sagemann W, Lesemann DE, Paul HL, Adomako D, Owusu GK (1985). Detection and comparison of some Ghanaian isolates of cacao swollen shoot virus (CSSV) by enzyme-linked immunosorbent assay (ELISA) and immunoelectron microscopy (IEM) using an antiserum to CSSV strain 1A. *Phytopathologische* Zeitschrift 114: 79-89.
- Steven WF (1936). Swollen shoot and die-back a new disease of cocoa. Gold Coast Farmer 5: 144.
- Strickland AH (1950). The dispersal of pseudocococcidae (Hemiptera-Homoptera) by air currents in the Gold Coast. Proceedings of the Royal Entomological Society, London (A) 25: 1-9.
- Strickland AH (1951). The entomology of swollen shoot of cacao. I. The insect species involved with notes on their biology. Bull. Entomol. Res. 41: 725-748.
- Thresh JM (1958). The spread of virus disease in cacao. Technical Bulletin West African Cocoa Research Institute 5: 36.
- Thresh JM (1988). Eradication as a virus disease control measure. In B. C. Clifford and E. Lester (eds.): Control of plant diseases: Costs and benefits, pp 155-194. Blackwell Scientific Publications, Oxford.
- Thresh JM, Lister RM (1960). Copping experiments on the spread and control of cacao swollen-shoot disease in Nigeria, Annals. Appl. Biol. 48: 65-74.
- Thresh JM, Owusu GK (1986). The control of cocao swollen shoot disease in Ghana: an evaluation of eradication procedures. Crop Protection 51: 41-52.
- Thresh JM, Owusu GK,Ollennu LA (1988a). Cocoa swollen shoot: An archetypal crowd disease. Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz 95: 428-446.
- Thresh JM, Owusu GK, Adomako B, Lockwood G (1988b). Ghanaian cocoa varieties and swollen shoot virus. Crop Protection 7: 219-231.
- Tinsley TW (1955). Virus infection in Cola chlamydantha. *Report* of the West African Cocoa Research Institute for 1954-1955 p. 32.
- Tinsley TW (1971). The ecology of cocoa viruses. I. The role of the wild hosts in the incidence of swollen shoot virus in West Africa. J. Appl. Ecol. 8: 491-495.
- Todd JM (1951). An indigenous source of swollen shoot disease of cacaol Nature (London) 167: 952-953.
- Van der Plank JE (1948). The relation between the size of the plant and the spread of systemic diseases: A discussion of ideal cases and a new approach to problems of control. Annals. Appl. Biol. 34: 376-387.