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# MICROBIOLOGICAL AND PHARMACEUTICAL ACTIVITY IN GHANA

A review

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## Abstract

## MICROBIOLOGICAL AND PHARMACEUTICAL ACTIVITY IN GHANA: A REVIEW.

The review deals with the microbiological and pharmaceutical activities in Ghana from 1900 to date. It indicates that most microbiological effort has been concentrated on cocoa disease control with comparatively little work on other crops. Similarly, soil microbiology has received practically no attention. Food microbiology, which was previously neglected, is now receiving serious attention. Work in medical microbiology has indicated an increasing drug resistance of <u>Salmonella</u> and <u>Staphylococcus</u> spp. to certain antibiotics.

Pharmaceutical research in Ghana has been concentrated on alkaloid-containing plants to find new substances as well as to identify plants from which existing known substances of medicinal value may be found as alternative sources of supply. Several new finds have been reported in this program of research.

# INTRODUCTION

Microbiological investigations in Ghana are conducted mainly in the University of Ghana, University of Science and Technology and by various departments of the Ghana Ministry of Agriculture. Most of the research functions of the latter department, however, have since 1962 been reorganized into a separate, but Government controlled, Council for Scientific and Industrial Research (C. S. I. R.). The latter body has the responsibility of co-ordinating all research activities in Ghana and directly controls a number of Research Institutes including seven that conduct research in agriculture. These Institutes are the:

> Cocoa Research Institute (Ghana) Crops Research Institute Soil Research Institute Animal Research Institute Forest Products Research Institute Institute of Aquatic Biology Food Research Institute

This paper reviews the microbiological activities in agriculture and medicine in Ghana from 1900 to the present as well as the pharmaceutical activities from 1958 to date.

# COCOA RESEARCH

Ghana is an agricultural country and naturally its research, including microbiological, activities are centered around agriculture and in particular around the cocoa industry which is the main stay of the nation's economy.

Like any crop, cocoa is plagued by many diseases and one of the first to be reported (Dade, 1922, 1927a) was a collar-crack root disease caused by the fungus <u>Armillaria melea</u> (Vahl exFr) Kummer. This fungus develops in the medullary rays and cracks the trunk longitudinally from ground level to a height of 4ft up the cocoa tree; the tree eventually collapses and falls. Several other minor root diseases, characterized by leaf fall and die-back of twigs, are listed by Wharton (1962).

Wharton (1962) also lists two blight diseases. One attacks single leaves and is called white thread disease and the other attacks whole canopies and is called the horse hair blight (Dade 1927d). These diseases are caused by the fungi <u>Marasmius</u> <u>scandens</u> Masse and <u>Marasmius</u> equicrinis Mull respectively.

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Of greater economic importance, but still of less importance in Ghana compared with other countries, is the black pod disease of cocoa. This disease is caused by the fungus Phytophthora palmivora. The disease blackens the pod tissue within 4 days from initial attack and destroys the beans inside the pods within 4 weeks if the pods are left on for so long. This disease has been studied with respect to its mode and conditions for infection (Dade, 1927b; Wharton, 1962; Bimpong, 1969), spread into the stalk and flower cushions, (Dade, 1928-29) pathogenicity (Dade, 1927e) strains and their distribution (Turner 1960a; 1960b, 1961; Wharton, 1962; Ashby, 1929; Dade, 1927b) and dissemination and control measures (Bunting, 1922, 1929; Wharton, 1955, 1958; Owen, 1951; Dade, 1927b and c; Hammond, 1958; Attafuah, 1965). From these studies it has become evident that chemical control of the disease is uneconomic because of the difficulties imposed by heavy rainfall and peasant agricultural practices. Indeed, field sanitation and management practices involving the removal of dead pods and husks from trees have proved the best method of control.

In 1936 a farmer from the eastern region of Ghana showed the Ministry of Agriculture a number of badly deformed cocoa branches from his farm. The disease was investigated and identified by the plant pathologist as a severe plant disease (Stephen, 1936) and was given the name "Swollen shoot and die black".

This incident was instrumental in the setting up of the inter-territorial West African Cocoa Research Institute based in Ghana to study cocoa diseases to save the industry.

The apparent sudden onset and the fact that surveys showed its wide occurrence on cocoa farms in Ghana made people believe that it was the result of some environmental change such as decreased humidity and soil deterioration. In 1938, however, Posnette (1940) traced the cause to a virus infection which had for its vector mealybug insects (Cotterell 1943; Posnette and Strickland, 1948) which are associated with <u>Crematogaster</u> ants (Strickland, 1951b; Cornwell, 1957; Hanna et al. 1956).

The importance and alarm caused by the discovery of swollen shoot in Ghana may be judged by comparing the number of published papers on black-pod disease and on swollen shoot. Between 1940 and 1960 the

number of published papers on black-pod and on swollen shoot was in the ratio of one on black-pod to 10 on swollen shoot (Ripley, 1968). In this period over 50 virus strains which possess varying degrees of virulence and induce shoot, leaf and pod symptoms (Entwistle 1958; Dale 1962; Kenten and Legg 1965) were isolated. It was found, for example, that most Ghanaian isolates produce swellings of various sizes but the Kpeve virus also isolated in Ghana only produced leaf-mottle symptoms (Thresh and Tinsley, 1959; Posnette, 1947). Indeed, some avirulent strains of the deadly New Juaben isolate of Ghana do not produce swellings at all.

The pattern of the spread of the cocoa virus has been shown to agree with the movement of the mealybug vector which may be by walk-migration (Cornwell, 1958), passive dispersal by air currents (Strickland, 1950; Cornwell 1960, 1955b) and by other agencies (Thresh 1958b; Posnette, 1943).

While prospects for controlling fungus diseases were good, studies showed that virus diseases of cocoa can only be prevented by the painful measure of grubbing or "cutting out" of diseased trees and the elimination of alternative plant hosts (Attafuah, 1965) on farms. However, this measure was started soon after Posnette (1940) had demonstrated the virus nature of the disease. Several attempts have been made at biological control (Nicol et al. 1950, Rojter et al. 1966), but though successful in the laboratory it has proved a failure in the field (Nicol 1953). The microbiological studies on cocoa have concentrated on disease control with only one publication (Knapp, 1935) on the microbiology of the wet bean with fermentation. Similarly, only three reports on the storage of the dry beans (Powell and Wood, 1951; Scott and Hudson, 1937; Anonymous, 1932) are on record.

## RESEARCH ON OTHER CROPS

The detailed and sustained microbiological research on cocoa, which has enabled the fundamentals of cocoa diseases to be understood and controlled, has not characterized work on other crops. Microbiological studies on these crops have been very fragmentary and unsustained. For example, over the period under review there were three publications on cocoyam root rot (Wright, 1930; Shephered, 1938; Posnette, 1945) and two papers each on tobacco leaf curl (Shephered, 1938; Stephen, 1955) and diseases of para rubber (Dade, 1927f; Robertson, 1947). There were also single publications on Cercospora disease of the bambara groundnut (Teyegagah, 1970), bacterial soft rots of cabbages and lettuce and hard rots of other vegetables caused by Sclerotium rolfsii and Rhizoctonia solani (Dade, 1934b), virus mosaic diseases of legumes (Lister and Thresh, 1956), virus rosette disease of groundnuts (Brunt and Bonney, 1964), a pepper disease caused by the fungus Leveillula taurica (Lev) Ar (Ayesu-Offei, 1966) and a fungal disease of the Carica papaya caused by the fungus Phyllactinia corylea (Clerk and Ankora, 1969a).

Diseases of the tomato fruit caused by the fungus <u>Sclerotium rolfsii</u> (Dakwa, 1965) and its control with herbicides (Clerk and Bimpong, 1969) have also been studied. In the latter studies it was observed that <u>S. rolfsii</u> germinated well in a 300 ppm aqueous solution of atrazine, prometryne and ramrod, but was completely suppressed in 400 ppm aqueous solution

of prometryne and 500 ppm of ramrod. A concentration of up to 1000 ppm of atrazine, however, permitted up to 48% germination.

The apparent lack of sustained microbiological activity on individual crops may be attributed to the fact that there have been no serious epidemics affecting any of these crops as has been experienced with cocoa and maize which resulted in the setting up of the West African Cocoa Research Institute (now Cocoa Research Institute, Ghana) and the former West African Maize Rust Research Unit. Another reason may be that most of these crops were not economically important to the colonial administration. This policy of waiting till trouble starts may not be good, but it is expedient with the limited amount of manpower available.

The maize Rust Research Unit was established in 1950 to investigate and find a solution to a maize rust epidemic caused by the rust fungus <u>Puccinia polysora</u> that hit West Africa that year. During the short life span (1950-61) of this Unit several technical papers were contributed on maize rust disease by several investigators (Stanton and Cammack, 1953; Stanton 1954; Cammack, 1956a, b, c; Rhind, 1952; Blaine, 1953). Before this Unit was set up, however, Bunting (1925, 1926, 1928) of the Gold Coast (Ghana) Ministry of Agriculture had reported on diseases of maize and other graminaceous plants.

Two economically important crops, whose diseases have received some study in Ghana, are the lime tree and the coconut palm.

The most important disease affecting the lime industry in the Central Region of Ghana is a virus die-back disease, tristeza, transmitted by the black aphis <u>Toxoptera citricidus</u>. This disease has been studied by various authors (Posnette, 1952; Hughes, 1949, 1954; Hughes and Lister, 1953; Martin, 1954a) and has been shown to be effectively controlled by using disease-resistant rootstocks, e.g. Rhangphur lime rootstock.

The Cape St. Paul or coconut wilt disease poses a difficult problem to microbiologists interested in this crop in Ghana. Some years ago Dring (1958) stated that it is not certain whether it is caused by a fungus, a virus or an eelworm. This statement is still applicable. The disease is characterized by nut dropping, yellowing, bronzing and drying out of the lower leaves and at the last stage of the disease the heart leaf dies and rots with the roots. This disease has been investigated by Leather (1959) and Martin (1954b).

Fungus of the <u>Fusarium</u> spp. are responsible for a number of plant diseases and tuber rots and this group of fungi has been taxonomically studied by Gordon (1960).

# SOILS

Crops and soils are closely linked but the microbiology of the latter has been very little investigated. A comparative account of the nitrogen cycle in Ghanaian Savannah and forest soils has been rendered by Meiklejohn (1962). The inhibitory effect of soil extract on the growth of some fungi has also been reported by Clerk (1969b). In this study it was found that aqueous extracts of humus-rich soil inhibited the germination and growth of <u>Beauveria bassiana</u> and <u>Paecilomyces farinosus</u>. The inhibitory substance was fungistatic rather than fungicidal and was inactivated by heat treatment.

## WATER

By building a dam across the Volta River in Ghana, the largest manmade lake has been formed and some microbiological observations on the lake have been made by Biswas (1967, 1969). She found that seasonal changes in temperature did not affect bacterial counts in the lake. She also found that the broad composition of the microbial population resembled that found in temperate lakes. There was, however, an indication of seasonal increase of spore formers especially in the surface and bottom layers. Presumptive coliforms have usually been low on the lake. Microbiological observations on other bodies of water have been studied by Abrahams (1963) and Chelty (1966).

# FOOD

Microbiological investigation into foods in Ghana has, until recently, been very limited. Apart from Fishlock's (1930a) paper on the moulding of copra which was followed by Rasper and Kuskova (1964) on the same subject and Abrahams' (1962) investigation of a local festival food suspected of causing diarrhoea to some subjects, no previous papers had been published on any microbiological studies of foods in Ghana. In the latter investigation a considerable number of <u>Clostridium welchii</u> in some food samples were isolated. With the setting up of the C. S. I. R. and, in particular, of the Food Research Institute in Ghana in the latter half of the sixties, microbiological studies on food are being taken seriously within the limits of our resources.

Several studies have been conducted on milk. Abrahams and Laryea (1968) have made a comparative study of the bacteriology of raw milk produced on the Accra Plains. This study has established the extent to which the quality of raw milk produced on university farms exceeds that on peasant farms. It has also been established (Caurie, 1970c) that there is considerable post-processing contamination of pasteurized filled milk sold in Accra. The shelf-life of the milk through existing distribution channels was, however, found satisfactory (Caurie, 1970d).

Fermented maize dough is a basic raw material in the preparation of a number of local food items. The microorganisms responsible for the later stages of the fermentation have been shown to be a mixed population of lactic-acid bacteria and yeasts (Christian, 1970). The most common bacterium was the homofermentative <u>Pediococcus</u> cerevisiae.

The tuber of the cassava plant is one of the staple food items in Ghana and one of the methods of preparation is to dry the peeled cut up tubers under field conditions. During the drying, which may last up to 6 or 7 days, a number of fungi grow on the exposed chips. Under humid drying conditions excessive mycofloral growth renders the chips very dirty and unappetising. The mycoflora which develop on the chips have been surveyed (Caurie, 1967). Over eighty percent of the colonising fungi were identified as <u>Cladosporium herbarum</u> together with smaller percentages of <u>Aspergillus</u>, <u>Penicillium</u>, <u>Sporendonema</u> and other species. Chemical changes caused by some <u>Aspergillus</u> species in the root tuber have also been studied (Clerk and Caurie, 1968).

Fermented cassava tuber pulp forms the basis for the preparation of a staple food item, gari, in Ghana. Changes in the microbial population and some concomitant chemical changes during the fermentation process have been studied (Caurie, 1970b). It was found, for example, that there are no successional changes of microorganisms during the fermentation and that both yeasts and bacteria grow together in identical rythms during the fermentation. The end of the fermentation is indicated by a decline in bacterial and an increase in yeast numbers.

The stability of dehydrated foods free of microbial growth or deteriorative chemical reactions resulting from products of microbial metabolism depends on the provision of optimal storage conditions. These conditions have been studied and worked out by Caurie (1970a, 1971a).

The effect of storage temperatures on the microbiological quality of some foods manufactured in Ghana has also received some study (Ako-Addo, 1970).

# FOOD PATHOGENS

Of particular importance and closely related to foods are organisms belonging to the <u>Salmonella-Shigella</u> group which may be taken in with the food to cause enteric fevers, dysentery and, in some cases, death. Gamble and Harris (1953) in their studies have reported that the fever symptom associated with <u>Salmonella</u> infection may be simulated by <u>Pseudomonas pyocynea</u>. Some <u>Salmonella</u> species found in the human have also been isolated from lizard droppings (Vella, 1956), from the python (Rewall et al. 1948) and from chickens (King and Gellatly, 1955). Hughes (1954a) considers beef as a potential source of <u>Salmonella</u> dublin infection in Ghana. The resistance to drugs of several <u>Salmonella</u> species have recently been studied by Sodhi et al. (1970) and shown to maintain a fairly high resistance to antibiotics.

Other organisms likely to contaminate food from human carriers are those of the <u>Staphylococcus</u> species. The bacteriology of wound sepsis which abounds in these species has also been studied (Sodhi et al., 1968). The antibiotic resistance of the most common <u>Staphylococcus</u> species occurring in wounds, <u>S. pyogenes</u>, has similarly been reported by Abrahams and Laing (1964a, 1964b). In the latter studies as much as 84% of the cultures were highly resistant to aureomycin, streptomycin and as high as 96% to chloramphenicol. Sodhi et al. (1968) found as much as 93% of the cultures were resistant to penicillin which indicated an increase of 9% in the antibiotic resistance of <u>Staphylococcus</u> species to penicillin in Ghana within 7 years.

On the distribution of contaminants on the human skin Heman-Ackah (1959b) found no uniform distribution, but reported that the skin may be effectively sterilized with either a solution of mercuric iodide (Heman-Ackah, 1959a) or by the usual scrubbing of the skin followed by treatment with a disinfectant.

# MEDICAL

A search through the medical literature from 1900 to date reveals that though many diseases of microbial origin have been reported, little or no research has been done on the microorganisms themselves.

Salles and Sodhi (1965) have, however, studied the characteristics of tubercle bacilli isolated in Accra while the resistance of these organisms in chemotheraphy has been studied by Bell and Brown (1961, 1963). Other microorganisms isolated and studied include <u>Norcadia</u> spp. from heart blood (Macfie and Ingram, 1921b), a number of anonymous mycobacteria (Sodhi et al., 1966), bacteria responsible for bacteriuria in pregnant women in Accra (Sefcovic, et al., 1967), and for the first time in Ghana medical history the much dreaded <u>Cholera</u> bacilli were isolated in Ghana last year (Pobee et al., 1970).

In plague control, Burgess (1927) was able to prepare vaccines from selected strains of <u>Bacillus pestis</u> and later Burgess (1930) studied the bacteriological variation in <u>Pasteurella pestis</u>, the causative agent of the disease.

Some bacteriological tests and techniques have also been developed and investigated by Hughes (1953d) and Eddington (1953c).

On the veterinary front, Oppong (1969) has studied the susceptibility of pigs, sheep and goats to the type A virus of foot and mouth disease. The exposure of these animals to the virus showed infection in the pig and subclinical or inapparent infection in the sheep and goats.

In addition to the regular microbiological research activity that goes on in Ghana a considerable amount of non-research, routine, microbiological activity goes on in private establishments and in Government Public Health Reference Laboratories, and in teaching especially at the University level.

# PHARMACEUTICAL RESEARCH

Pharmaceutical investigations are conducted at both the Chemistry Department of the University of Ghana and at the Pharmacy Department of the University of Science and Technology (U.S.T.). It is, however, at the latter University that the bulk of the investigations are carried out. Here, the Pharmacy Department has, since 1958, been specialising in alkaloid-containing plants in a special program on medicinal and toxic herbs which are abundant in Ghana and which may provide useful starting material for the development of new substances of medicinal value. This program is not only out to look for new substances but also to identify alternative plant sources from which known substances of medicinal value may be extracted.

In this program at the U.S.T., several new alkaloids have already been extracted, characterized and named. From the root bark of the plant <u>Teclea</u> <u>verdoorniana</u> Excell and Mendonca (Syn. <u>T</u>. <u>grandifolia</u>) a total of eight alkaloids have been identified (Tackie, Kofi-tsepo and Hadzija, 1967). Out of this number three have been crystallized. One of these has been shown to be identical to the known alkaloid evoxanthine while the other two are new alkaloids (Tackie and Martin, 1970). One of these new alkaloids has been shown to be a furoquinolone base and the other an acridone-type base and have been named tecleadine and verdoornine respectively; tentative structures have been assigned to them.

Another new alkaloid, funiferine, has been isolated from the root bark of <u>Tiliacora funifera</u> by Tackie and Thomas (1965) and these investigators are working on its structure.

The pharmacological value of some alkaloids extracted from local herbs have been studied. The alkaloid, isorotundifoline, extracted from the leaves of certain <u>Mitragyna</u> plant species has been shown to be comparable to papaverine hydrochloride in its musculotropic and spasmolytic effect (Szreniawski and Tacker, 1964).

Githens (1948) has stated that many <u>Solanum</u> species in West Africa contain the alkaloid solanine. One of the commonest <u>Solanum</u> species in West Africa is <u>S</u>. torvum and its leaf was investigated to find the level of solanine, if any, present in it. Investigations (Tackie, 1959a) showed no alkaloidal content but it appeared that the active principle in the <u>S</u>. torvum leaves is a glycoside which yields a mixture of sugars of which dextrose is the principal component. This active principle was shown by Gyan (1959) to have a direct depressant effect on frog heart muscle and an atropine-like effect on an isolated guinea pig ileum.

Talalaj (1964-67) has extracted a number of essential oils from various Ghanaian plant species. From the dried leaf of the lemon grass <u>Cymbopogon citratus</u> (D.C.) Stapf, 2-2.5% oil has been distilled which contains 70-78% citral (Talalaj, 1964a). Citral is a starting material for the synthesis of vitamin A and some synthetic perfumes.

The leaves, flower tops and flower heads of <u>Lippia multiflora</u> have yielded 0.80%, 1.5% and 2.1% oil, respectively, on distillation (Talalaj, 1964b). The oil contained 2.6 to 3.9% camphor.

The content of essential oil from the dried leaves and flowers of Ocimum viride (Willd) were found to lie between 2.2 and 2.4%. This oil contained 39-41% of thymol as the main constituent (Talalaj, 1964c).

Among the medicinal plants in Ghana are some members of the <u>Callistemon</u> species. By steam distillation of the dried leaves of <u>C</u>. <u>lanceolatus</u> and <u>C</u>. <u>rigidus</u> 1% oil was recovered which had a cineole content of 63% and 54% respectively (Talalaj, 1965a).

The nutmeg seed is a common spice in Ghana from which Talalaj (1965b) has extracted 4% oil containing 2% phenols which are probably carvacrol.

Unlike the plants so far studied the leaves of the <u>Cedrela</u> <u>mexicana</u> contain very small amounts of oil. The oil content was found rather concentrated in the wood and it contained only small amounts of phenols which are also believed to be carvacrol. (Talalaj, 1965c).

The vetiver oil content of <u>Vetiveria</u> <u>zizanioides</u> Stapf grass roots in Ghana (Talalaj, 1965d) contained 19-20% ketones.

The essential oil content (3-4%) of ginger grown in Ghana has been found to be of very high quality (Talalaj, 1966a).

Like the <u>Callistemon</u> species the cineole is the main constituent of the oil extracted from the <u>Melaleuca leucadendron L</u> (Talalaj, 1966b). The oil content of the fruits of <u>Xylopia aethiopica</u> (Talalaj, 1966b) also contains 6-8% of cineole.

Some fruits of the lime tree grown in Ghana have been reported (Talalaj, 1966c) to contain much higher levels of oil extracted by distillation than is reported in the literature.

Elemi oleo - resin collected from incised wounds on <u>Canarium</u> <u>schweinfurthii</u> grown in Ghana - has been shown (Talalaj, 1966d) to be a very good source of elimi oil. This oil was reported to contain phellandrene and high amounts of free alcohols and low levels of ester. There was a complete absence of aldehydes and phenols.

The resin from the <u>Daniella</u> <u>oliveri</u> plant has been reported to contain as much as 50% oil (Talalaj, 1966f).

The oil content of the dried leaves of <u>Eucalyptus citriodora</u> has similarly been shown to be a good source of citronellal (60.4%) (Talalaj, 1966g).

Though the characteristics of the oil from the leaf of the <u>Cinnamomum</u> <u>zeylamicum</u> Nees grown in Ghana bear close identity to Ceylon cinnamon oil, the oils from the bark of species from the two localities show differences in certain essential characters (Talalaj, 1967). These differences have been attributed to soil differences.

The activity of the Pharmacy Department at U.S.T. is not only limited to medicinal herbs but extends to the investigation of non-herbal pharmaceutical study as well. Thus, the efficacy of some corticoid drugs (Liniecka, 1968) and the use of certain clay preparations in traditional medicine (Tackie, 1959b; Heman-Ackah, 1960) have been reported. Tackie (1960) has also identified the active principle of a popular hair dye in Ghana and advised on its proper application.

The group of workers at the Chemistry Department at the University of Ghana has, in parallel investigations, also extracted a number of alkaloids from <u>Fagara</u> spp. (Torto et al., 1969) and several coumarins from <u>Afraegle paniculata</u> (Schum and Thom) (Adjangba et al., 1968). In the latter studies three furo-coumarins, imperatorin, heraclenin and xanthotoxin as well as two chromenocoumarins, xanthyletin and xanthoxyletin, were extracted from various parts of the <u>A</u>. <u>paniculata</u> plant.

# CONCLUSION

This review has dealt with the microbiological activities in the fields of agriculture, food and medicine from 1900 to date. It has also reported on some pharmaceutical research in Ghana. Some aspects of certain subjects have received more attention than others. The reason for this is economic since at our present stage of development it is not possible to have men, in sufficient numbers, to tackle problems that are not considered of prior importance.

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