BIOSYSTEMATIC STUDIES IN SOME NIGERIAN SPECIES OF <u>ANTHERICUM</u> LIN. AND <u>CHLOROPHYTUM</u> KER-GAWL. (LILIACEAE).

BY

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A THESIS IN THE DEPARTMENT OF BOTANY SUBMITTED TO THE FACULTY OF SCIENCE IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY OF THE UNIVERSITY OF IBADAN.

MARCH, 1981.

## DEDICATION

In memory of my dear mother Madam Susannah Okuola Adeyemi.

ANLIK

## ABSTRACT

Field surveys and investigations of representatives of Anthericum L. and Chlorophytum Ker-Gawl. complex in Nigeria were carried out in their natural habitats in at least sixteen states of the Nigerian Federation. All herbarium specimens in the Forestry Research Institute, Ibadan and in nine Nigerian Universities visited were examined (ir any). So far ten species of Anthericum and fifteen species of Chlorophytum have been reported and collected in Nigeria. They were all studied morphologically. Detailed ecological analysis including the chemical composition of their soils were carried out on three taxa of Anthericum and ten taxa of Chrolophytum. Many living populations were sampled from their different natural habitats in Nigeria. Their representatives were cultivated in three locations for experimental studies. Data were collected on the morphology by conventional methods and analysed. Anatomical data were amassed from the leaf surface patterns and the roots. Types of leaf margin anatomy were noted. Cytological studies were carried out in five taxa of Anthericum and eleven taxa of Chlorophytum, There were inter-generic hybridization tests.

The use of loaf vein spacing interval and anther: filament ratio as taxonomic criteria have been suggested and their usefulness in the monocotyledon taxonomy, especially in Liliaceae, needed to be further explored. The importance of leaf margin anatomy as a taxonomic criterion above species level was highlighted.

Chromosome counts for seven taxa were confir	med, viz:
C. macrophyllum (A. Rich.) ischers	2n = 28
C. blepharophyllum Schwinf. ex Bak.	2n = 28
C. stenopetalum I Bak.	2n = 14
C. stenopetalum II (sp. nov.)	2n = 14
C. laxum R. Br.	2n = 14
C. inornatum Ker-Gawl. C. togoense Engl.	2n = 14 $2n = 14$
New chromosome counts were recorded in:-	
C. geophilum Peter ex. v. Poelin.	2n = 28
C. alismifolium Bak.	2n = 16
Chlorophytum X (related to C. elatum)	2n = 16
C. caulescens (Bak.) Marais & Reilly	
(formerly A. caulescens Bak.)	2n = 16
<u>A. limosum</u> Bak.	2n = .16
A. pterocaulon I Welw. ex Bak.	2n = 24
A. pterocaulon II "	2n = 16
A. nubicum Bak.	2n = 16
A. pubirhachis Bak.	2n = 16
A. uyuiense	2n = 16

The possible evolution of chromosome numbers in the complex has been postulated. The possible chromosome evolution of the complex based on the available data from this work and existing

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literature has been proposed. Evidence was adduced to support the suggestion that the possible basic chromosome number in the complex was x = 4, and that n = 8 was a secondary basic number. It has also been contended that n = 7 and n = 6 evolved from a more primitive n = 8.

Supplementary evidence in support of Marais and Reilly's (1978) transfer of <u>A</u>. <u>caulescens</u> from <u>Anthericum</u> into <u>Chlorophytum</u> as <u>C</u>. <u>caulescens</u> was adduced. A form of <u>B</u>.<u>stenopetalum</u> has been elevated to species level on account of its morphological, cytological and genetic differences. A new taxonomic key has been proposed for the separation of <u>Anthericum</u> and <u>Chlorophytum</u>.

The possible mode of evolution of some Nigerian species of <u>Anthericum</u> and <u>Chlorophytum</u>, based mainly on their leaf surface patterns, has been proposed.

#### ACKNOWLEDGEMENTS

I wish to express my profound gratitude to my supervisors: Professor M.B. Scott-Emuakpor and Dr S.O. Oyewole, for their sound guidance and keen supervision of this work. I am particularly grateful and thankful to Dr S.O. Oyewole, who had to continue supervising the later part of this work, despite his transfer of service from University of Ibadan to University of Ilorin, for his personal financial involvements, inconveniences, understanding and for making his unpublished data available to me.

I am very grateful to Dr Joyce Lowe for her help in the taxonomic aspect of this work and her kindness in reading through the draft thesis. I quite appreciate her constructive criticisms and useful suggestions. I am also grateful to all members of the academic staff especially Professor S.O. Alasoadura, the Head of Department, and Dr A.O. Fajola for their kind gestures and encouragements during the period of this work.

I am grateful to all the technicians, and the administrative staff of the Department of Botany, University of Ibadan, especially Messers M.A. Oyebode; E.A. Ogunduyilemi and Mrs Bimbo Aina for their technical assistance. I am greatly indebted to the staff of the Forestry Research Institute of Nigeria, Ibadan for their cooperation. My gratitude is also due to Mrs R.A. Azeez and Mr J. Tyiola of the

- vi -

Department of Agronomy, University of Ibadan, for their technical assistance when the soil chemical analysis was being done.

I am greatly indebted to the Department of Biological Sciences, University of Ilorin, for the facilities made available to me during this programme.

I am grateful to my sister, Arinola, and Mr A. Adesina for their moral and financial support. I am particularly grateful to my wife, Adebowale, my children: Omoniyi, Ademola, Adedola Adetilola and 'Femisi, for their patience, endurance and understanding.

I am grateful to Messers J.A. Ogunleye and J.O. Osasona for typing and Dr V.A. Adisa for proof reading the manuscripts.

Finally, I wish to thank the Federal Government of Nigeria for the award of bursary to carry out this programme.

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## CHAPTER 1

#### INTRODUCTION

Cytogenetics, a relatively new aspect of biological and medical sciences is a hybrid of cytology and genetics (Swanson, 1965). Cytology is undertaken in the hope of determining the presence of heritable characters, establishing the physical bases of heredity so as to throw light on the phylogeny and inter-relationship of multitudinous forms of living organisms, through studying the structure, behaviour, growth, reproduction of cells and exploring the properties of the protoplasm (Darlington, 1928); Sirks, 1956; and Gardner, 1965). Genetics, on the other hand, deals with heredity, a study which seeks to explain the similarities and differences that exist between parents and their offspring (McGraw-Hill, 1977), between closely related species, general or even tribes. In order to establish a comprehensive and convincing cytogenetical relationship between tio morphologically similar organisms at any taxonomic level, studies of ecology, detailed morphology and anatomy, cytology and experimental breeding of the organisms need be investigated. This is necessary because the phenotypic characters are the manifestations of the genes located on the chromosomes, the plasmagenes or cytogenes (i.e. fractional parts of chromosomes found in the cytoplasm which can multiply by themselves completely and

independently (Wright, 1941; 1945; Sirks, 1956; and Webster, 1966) in interplay with environmental factors which play a modifying role. Since this study tries to employ these aspects; ecology, morphology, anatomy, hybridization and cytology - in considering the cytogenetic relationship and the probable mode of evolution of the genera <u>Anthericum L. and Chlorophytum Ker-Gawl.</u>, the word 'Biosystematics', which is ordinarily more inclusive than 'cytogenetics' is used in the title.

The role of cytogenetics in the solution of taxonomic problems is assuming increasing importance Many such recent works include Brighton and Wickens (1976) on Combretum, Brighton and Ferguson (1976) on Melalevea malalena. Oye ole (1972), employed cytogenetics to elucidate the relationship and evolutionary trend among the species of Albuca. Naik (1976) employed cytogenetic data to postulate the probable evolution of Indian species of the genus Chlorophytum. On many occasions, cytology and genetics have helped in supporting or correcting and, in some cases, rejecting early classifications of organisms through chromosome counts, and chromosome morphology. coupled with results of inter-breeding experiments. Buchanan (1958). working on Luzula campestris, Serota (1959), working on the Trilliums of Northern Carolina and Carr (1978), working on Railliardia and Dubautia, two genera of compositae, employed these methods in solving their different taxonomic problems.

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The close similarities between <u>Anthericum L. and Chlorophytum</u> Ker-Gawl., two genera of the family Liliaceae, have generated confusions. Family Liliaceae, as described by Baker (1888), was more complex than the present day family in that the members of the tribe Dracaeneae have been transferred to form a part of the new family Agavaceae while those with umbellate inflorescence (tribe Allineae) are now members of the family Amaryllidaceae. The family Smilacaceae was formed of the scandent stemmed members of Baker's Liliaceae (Oyewole, 1971).

In spite of these transfers, the present family Liliaceae is still complex, with complexities in similarities and forms among species and genera. The tribe Asphodeleae consists of <u>Anthericum</u>, <u>Chlorophytum</u>, <u>Acrospira</u>, <u>Bulbine</u>, <u>Asphodelus</u>, <u>Dasystachys</u>, <u>Eriospermum</u>, <u>Diuranthera</u>, <u>Echeandia</u>, <u>Trachyandra</u>, <u>Trichopletalum</u> (<u>Bottionea</u>), <u>Verdickia</u> and <u>Schizobasis</u> (Marais and Reilly, 1978). Out of these genera, only <u>Anthericum</u> and <u>Chlorophytum</u> and <u>Eriospermum</u> Jacq. (Hepper 1968) are represented in Tropical West Africa. <u>Eriospermum</u> is represented by only one species, <u>E. abyssinicum</u> Bak. in West Tropical Africa. Hence only <u>Anthericum</u> and <u>Chlorophytum</u>, the two genera with appreciable representatives, are considered in this study. The major problem of the <u>Anthericum</u> L. and <u>Chlorophytum</u> Ker-Gawl. complex is mainly of developing taxonomic criteria, which will separate them or group them together, with convincing, observable and objective characters. In trying to solve this problem, Hepper (1968) separated the two genera in his latest revision of Liliaceae (F.W.T.A., 3(i)) as follows:

"Fruits globose or shallowly lobed,

seeds not flat.

Inflorescence slender, more or less branched, Base of leaves more or less reddish mottled.

.....(5) Anthericum

Fruits deeply lobed, seeds compressed Inflorescence branched or subspicate; Reddish mothling absent from the base of leaves. .....(6) Chlorophytum"

The criteria on which this key is based, are not clearly comparative enough and thus subject to individual judgement. Hepper has admitted that the taxonomy of these two genera calls for a revision in a personal communication (Ngwa, 1979; cited). The criterion which seems to separate the two genera decisively is the reddish mottling of the leaf base; and this is not true of all the

Anthericum specimens. So far, reddish mottling has not been found in the leaf base of A. limosum Bak. A. nigericum Mepper, A. nubicum bak. and A. pterocaulon Welw.ex.Bak. specimens collected by the author or preserved in the Forest Research Institute herbarium (FHI), Ibadan, and at the Botany Depart ment herbarium of University of Ibadan (UIH) and that of University of Ife, (IFE). Hepper, himself only noted reddish mottling in A. dalzielii Hutch ex Hepper out of the fourteen species of Anthericum he listed. However, the base of the inflorescence of specimen of A. pterocaulon in the herbarium of the Forest Research Institute of Nigeria. Ibanan is reddish mottled. Specimens of this species collected on the field trips for this work showed no reddish mottling. This criterion therefore is not very reliable. The so called reddish mottling has been found to be dependent on the stage of development and for environmental changes in A. pubirhachis Bak. The species had reddish mottling before flowering but the reddish mottling gradually faded away as the fruits developed. The reddish mottling also disappeared gradually as from mid-June in the non-flowering representatives of the species.

Hanid (1974), working on Kenya species, and Marais and Reilly (1978), using Kew herbarium collections of <u>Anthericum</u> and <u>Chlorophytum</u>, saw the limitation of Hepper's classification and used the seed and fruit characters as their **b**asis of classifications.

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It is interesting to note that Marais and Reilly, working on almost all species so far collected in the world did not include any of the eight species of <u>Anthericum</u> that Hanid transferred to <u>Chlorophytum</u>. Thus their work did not complement each other. Nor was reference made by Marais and Reilly (1978) to any of the representatives of the genera in Tropical East Africa. The criteria put forth by Ngwa (1979) for separating the two genera are not more reliable than what Hepper and other workers have advocated.

The confusion in the two genera - Anthericum and Chlorophytum does not stop at the generic level Poellnitz (1951) reported seventy species of Anthericum in Tanzania, while Moore (1951) added six new species. Hepper (1968) listed twenty-four species of Anthericum in Tropical West Africa, which he reduced to fourteen by synonym, out of these 14, ten species have been collected in Nigeria. Willis (1945) put Chlorophytum species as one hundred and fifty, with seventy-five species reported to have been found in Tropical Africa, seventy-three species from South America and South Asia while the remaining two species were from Australia. Hepper (1968) recorded fifty-one taga of Chlorophytum and five taga of <u>Dasystachys</u> in Tropical West Africa, which he reduced to twenty-one species of <u>Chlorophytum</u>. Fourteen out of these have been collected

The large number of species recorded by both Poellnitz and Willis might have been due to the similarities between the two genera. Hence

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the same taxan might have been named as a species of both <u>Anthericum</u> and <u>Chlorophytum</u>. The other possibility is that these plants manifest different vegetative morphology in different environmental conditions, at different times of the year (i.e. seasonal variations) or at different ages. Thus a single taxon may possibly have four to six different identifications: two or three as Chlorophytum and two or three as <u>Anthericum</u>. It is therefore not surprising when Obsermeyer (1962) reduced five species of <u>Chlorophytum</u> to synonyms. Hanid (1974), in Kenya lumped eight species of <u>Anthericum</u> and two species of <u>Chlorophytum</u> into <u>C. affine</u> Bak. Ooststroom (194) also grouped <u>A. rouwonertii</u> De:Corter with <u>C. capense</u> Kuntze.

Except for Ngwa (1979) who has worked on the cytogenetics of nine taxa of <u>Chlorophytum</u> in Aigoria, no other work has been recorded on Nigerian species of <u>Chlorophytum</u> and <u>Anthericum</u>. The present study, therefore aimed

- (i) to examine and analyse the morphology, anatomy and ecology of some of the Nigerian representatives of the two genera with a view to establishing the relationships between the two genera;
- (ii) to examine the cytological similarities or differences
   between the representatives of the two genera in Nigeria
   with a view to defining their relationships cytologically;

- (iii) to attempt to establish genetic relationships between the two genera by attempting inter-generic crosses between representatives of both genera;
  - (iv) to critically examine and analyse their similarities and/or differences to see whether they are two distinct genera with clear, convincing, comparative and definitive criteria, or whether they should be merged as one genus, and
  - (v) to consider a probable evolutionary pathway for the Nigerian species of the two genera.

#### CHAPTER 2

## COLLECTION AND HABITAT STULY.

## A. MATERIALS:

Preliminary investigations of vegetative morphology of the herbarium specimens of the two genera were made at the Forest Research Institute, Ibadan, and Botany Department, University of Ibadan, Nigeria. In all, over one hundred and fifty specimens comprising nine species of Anthericum L. and fifteen species of <u>Chlorophytum Ker-Gawl</u>. were examined. This preliminary study was considered necessary in order to become familiar with the members of the two genera. The habitats and areas of collection of each specimen were noted so as to aid in future collection trips.

Some cultivated speciments of both genera, growing in the Nursery of Botany Department, University of Ibadan, were also examined, to aid field identification. These live specimens belonged to <u>Anthericum limosum</u> Bak. <u>A. pterocaulon</u> Welw. ex. Bak., <u>C. caulescen</u>s (Bak.) Marais & Reilly (<u>A. caulescens</u> Bak. <u>C. laxum</u> R.Br., <u>C. macrophyllum</u> (A.Rich.) Bak., <u>C. toqoense</u> Engl., and <u>Chlorophytum</u> X (very close to <u>C. elatum</u> R.Br.).

#### COLLECTION TRIPS

Several local and two long field trips were undertaken. On each occasion, at least one experienced field worker from either the Forest Research Institute or the Nursery/Botanical Garden of the Department of Botany, was taken along on the trip.

#### a. Local Trips:

Local collection trips were made to the neighbouring areas including the Botanical Garden, University of Ibadan, Olokemeji Forest Reserve, Federal Forest Reserve, Eleyele, Ibadan, Oke-Ogun Forest Reserve, Iseyin and Federal Forest Nursery, Igbetti. Most of the Chlorophytum specimens brought from these local trips were checked by comparing them with cited herbarium specimens for identification. Further confirmation was made by using Flora of West Tropical Africa, Volume III Part I. Species identified from the Chlorophytum specimens were C. togoense, C. macrophyllum, C. inornatum and C. alismifolium. Chlorophytum caulescens (A. caulescens) was also collected. Some of the plants were preserved as herbarium specimens while others were planted in plastic pots in the Nursery of Botany Department, University of Ibadan. Periodic visits were made to the habitats of C. caulescens, C. inornatum and C. macrophyllum in the Botanical Garden, University of Ibadan, Ibadan. Those at Onigambari, Igbetti and Iseyin were

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visited every three months. The investigations of these plants in their natural habitat were to find out if any of their characters were due to seasonal variations and also to compare them with those found in cultivation.

## b. Long Trips:

Two long collection trips were made during this study. The long trips were necessary so as to have a wider sample of each species and to particularly look for species of <u>Afithericum</u> which are found mainly in the Northern parts of the country. Possibility of collecting new taxa. of <u>Chlorophytum</u> was also anticipated.

In August, 1977, a field trip was made to northern Nigeria Fig. 1). During this trip, <u>A. nubicum, <u>G. caulescens</u>, . <u>A. subpetiolatum</u>, <u>A. limosum</u>, <u>A. pubir.hachis and <u>A. pterocaulon</u> I were collected. Other specimens collected were <u>C. blepharophyllum</u>, <u>C. stenopesulum</u> I and <u>C. geophilum</u>. Most of these plants did not have seeds while some, especially the <u>Anthericum</u> species, were not flowering at the time of collection. They were therefore planted in pots in the Nursery for observation. Three of the <u>Anthericum</u> species - <u>A. pubirhachis</u>, <u>A. subpetiolatum</u> and <u>A. nubicum</u> - and <u>C. geophilum</u> withered and did not come up again during the following growing season.</u></u>

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In order to get more species of <u>Anthericum</u> and possibly collect them in flowers, another field trip was undertaken towards the end of June and early July in 1979. During this trip many specimens of <u>Anthericum</u> and <u>Chlorophytum</u> were again collected. Many populations of <u>C. stenopetalum</u>, <u>C. macrophyllum</u>, <u>C. inornatum</u>, <u>C. blepharophyllum</u> and <u>C. laxum</u> were sampled. <u>Anthericum pubirhachis</u>, <u>A. limosum</u>, <u>A. pterocaulon</u> and <u>A.</u> <u>uyuiense</u> were also sampled from different populations.

The following is a list of the taxa collected and on which ecological, morphological, anatomical, cytological and genetical studies were carried out:

> Chlorophytum alismifolium Bak. Anthericum limosum Bak. Anthericum pterocaulon Welw. ex. Bak. Anthericum pubirhachis Bak. Chlorophytum caulescens (Bak.) Marais & Reilly Chlorophytum X (close to C. elatum R.Br.) Chlorophytum blepharophyllum Schweinf ex. Bak. Chlorophytum inormatum Ker- Gawl. Chlorophytum laxum R. Br. Chlorophytum macrophyllum (A.Rich.) Aschers Chlorophytum stenopetalum Bak. Chlorophytum togoense Engl.

In addition, some data were also accumulated on a variety of each of <u>A</u>. <u>nubicum</u> Bak., <u>A</u>. ?<u>pterocaulon</u>, <u>A</u>. <u>uyuiense</u> Rendle and <u>C</u>. <u>geophilum</u> Peter ex v. Poelln. which were collected

Voucher specimens of all the plants collected are lodged in the University of Ibadan Herbarium (UIH) and Forest Research Institute Herbarium, Ibadan (FHI).

#### B. ECOLOGICAL STUDY

Anthericum and Chlorophytum species have been reported to be greatly influenced by environmental factors (Hanid, 1974). The confused state of their taxonomy is attributed to their quick morphological responses to ecological changes (Ngwa, 1979). Thus the study of their ecology is necessary in order to be in a better position to assess objectively the influences of various ecological factors on their distribution and morphology.

#### 1. METHODS:

## (i) Environmental Studies:

Each collected sample from every sampled population was given an index number immediately after collection and labelled accordingly. Records of their vegetative peculiarities, area of collection, and floristic features of the location of each population were all kept in a field notebook for each sample. Samples of each specimen, labelled with the specimen's index number were planted in plastic pots after each flatd trip. The specimens were then identified by comparison with authenticated herbarium specimens.

Representatives of each sample were put in the green house, some under the Nursery shed, while a set was kept outside under shade in the Nursery. Another set of the samples was kept at Ijebu-Ode, 70km Southwest of Ibadan, for observation and investigation.

## (ii) Soil Analysis:

Soil sample was taken from the habitat of every sampled population just from the spot where specimens were collected. The soil was labelled with the same index number as the plant samples. The soil sample was weighed with a spring balance and the weight was recorded accordingly in the field note book (See (b) below).

## a. Mechanical Analysis:

Soil samples collected during 1977 field trip to the Northern States were analysed roughly. Each soil sample was suspended in 50ml. of distilled water in a 100ml. graduated glass cylinder. The suspension was thoroughly shaken and allowed to stand for twenty-four hours to settle down properly. Each soil layer was then worked as a percentage of the whole column of the soil (Oyewole, 1971).

The above method, very simple in operation, was found very helpful for materials occurring in widely varied ecological niches. Since species of <u>Anthericum</u> and <u>Chlorophytum</u> occur in very similar ecological niches, this method became inadequate. To forestall the limitation of this method, hydrometer method of soil mechanical analysis, modified by Bouyoucos (1936) was used for the soil samples collected during 1979 field trip.

## b. Determination of Field Water Holding Capacity.

After the field trip, the soil samples were dried at room temperature and weighed. The difference was recorded as the weight of water contained in thet given weight of the soil. The water holding capacity was expressed as the ratio of weight of water per unit weight of soil.

### c. Determination of Soil Water.

The air dried soil sample was weiged. It was then oven dried for twelve hours. The soil was allowd to cool down and weighed. The difference in the weight of air dried soil and oven dried soil was recorded as the weight of soil water. It was found as a percentage of the air dried soil.

## d. Determination of Total Nitrogen:

For the total nitrogen content of each soil sample, the method described by Yuen and Pollard (1953) and Machenie & Wallace (1954), modified by Department of Agronomy, University of Ibadan, Nigeria was used.

The total percentage of Nitrogen in the soil was calculated and recorded accordingly.

## e. pH. Measurement:

10gm. of air dried soil sample was weighed into a pH cup. 25ml, of distilled water was added to the soil and stirred for five minutes. The pH meter was set at pH 4, pH 7, and pH 9. The suspension was then stirred just prior to taking of the pH reading. The pH for each soil sample was recorded.

f. Exhangeable Cations:

Exchangeable cations determined for this work were potassium (K<sup>+</sup>), Sodium (Na<sup>+</sup>), Calcium (Ca<sup>++</sup>) and Magnesium (Mg<sup>++</sup>). SandelWs (1944) methods for determination of K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>++</sup> and Mg<sup>++</sup> were used. The amount of Na<sup>+</sup>, K<sup>+</sup> and Ca<sup>++</sup> were read from each soil filtrate on Flame photometer. The amount of Mg<sup>++</sup> present was read on the Atomic Absorption spectrophotometer.

g. Determination of Phosphorus in the Soil:

The method of Jackson (1962) modified by the Agronomy Department, University of Ibadan, Nigeria, was used for the determination of the average P in each soil sample.

The amount of average P was read on Spectromic 70 at 882nm. The reading was recorded for each soil sample.

(iii) ECOLOGICAL DATA:

The ecological . data of <u>Anthericum</u> and <u>Chlorophytum</u> representatives in Nigeria were collected from the Herbarium of the Forest: Research Institute of Nigeria, Ibadan. Some data were also collected from the Department of Botany, University of Ibadan, Department of Botany, University of Ife, Department of Botany, University of Jos, Department of Biological Sciences, Ahmadu Bello University, Zaria and that of University of Ilorin. The herbaria of the Universities of Calabar, Port-Harcourt, Nigeria, Nsukka; Ado Bayero, Kano; and Lagos were visited in June/ July, 1979. They all had little or no records of <u>Anthericum</u> and <u>Chlorophytum</u>. The geographical locations of the populations sampled and the area of collection mentioned by Hepper in the Flora of West Tropical Africa were also extracted for record purposes.

From all the above sources, geographical locations of the habitats of <u>Anthericum</u> and <u>Chlorophytum</u> species, so far collected in Nigeria were compiled (See Map II).

#### C. MORPHOLOGICAL AND ECOLOGICAL OBSERVATIONS

The ecology of three <u>Anthericum</u> species and eight species of <u>Chlorophytum</u> was studied. A comparative study between the collected specimens and those cultivated revealed some degree of observable changes in their morphological characters. A further observation between specimens of the same species put at different locations in the same locality - Nursery of Botany Department - and those at Ijebu-Ode, revealed some morphological changes.

Chlorophytum caulescens (Bak.) Marais & Reilly (A. caulescens)

Populations of <u>C</u>. <u>caulescent</u> were observed and sampled from many habitats including Botanical Garden, University of Ibadan, the outskirts of Kagara, a small village 70 km. south of Kaduna, and at a <u>Eucalyptus</u> plantation just twenty-four kilometres to Kaduna from Birnin Gwari. Those collected from the Kaduna district and those at the Botanical Garden were the same morphologically. However, those from Kaduna had narrower leaves and were taller than those at Ibadan, which were collected among crasses (e.g. <u>Andropogon tectorum</u>). The specimens had "pseudostem" made up of sheathed leaf bases. The leaf base had no reddish mottling.

Those collected from about 200 metres south of Kagara town, (along the side of a hill lying about seventy metres

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to the right of the main road), an open area, were different from those of the other populations. They had soft hairy structures which covered the leaf lamina and the sheath. In cultivation they had more leaves. The hairiness of leaf and sheath surfaces became reduced and was eventually lost.

### Anthericum limosum, Bak.

Populations were sampled along Zaria - Samaru road and around the railway crossing in Jos, where they grew with species of Andropogon, Jardinea and Imperate.

The species forms short 'pseudostems' which are compressed or flattened. Their leaf bases have no reddish mottling although they are pinkish.

There were no observable morphological differences attributable to environmental factors. The species flowered very well in cultivation, but produced no matured fruits. Even those preserved in Forestry Research Institute of Nigeria, Ibadan and those at Universities herbaria visited seldomly had more than two developed fruits in a whole inflorescence of more than ten flowers.

#### Anthericum pterocaulon Welw. ex. Bak.

Two varieties of this species were encountered in this work. The first variety grew luxuriantly in the Nursery but did not flower for two years. After the root-tips had been harvested for mitotic studies, the specimen withered. Unfortunately the specimen did not sprout during the following growing season. The area of collection and the collector were not known. Efforts made to get new specimens on field trips failed. This variety was hairy. It had short compressed 'pseudostem'.

The second variety of <u>A</u>. <u>pterocaulon</u> was sampled from two populations during field trips. The first population was sampled on the outskirts of Jos. The habitat was a well weathered rock just before entering the town from Zaria. The area was shaded by some species of <u>Combretum</u>. The second population samped was located a few metres from Bukuru on Gimi Road. They were found among species of <u>Andropogon</u>, <u>Imperata</u>, <u>Perotis</u> and <u>Combretum</u>.

The specimen had short compressed pseudostem formed by the leaf base sheaths. The leaf base had no reddish mottling. It was pinkish. The specimen did not grow well in cultivation, Most of the cultivated samples died after the first growing season.

## Anthericum pubirhachis Bak.

Only one population of <u>A.pubirhachis</u> was sampled about one kilometre to Jengre on Zaria-Jos Road. The population was found just a few metres from the main road. The soil was dry and cakey. The specimen was stunied and found among short grasses like <u>Heteropogon</u> and <u>Setaria</u> species. The mottling of the leaf base was not observable in the field.

They grew well in cultivation. The lear mases had reddish mottling or reddish stripes. A short compressed 'pseudostem' was formed. The specimen representatives flowered in cultivation. The reddish mottling started to fade away as from mid-June in mature leaves.

#### Chlorophytum alismifolium Bak.

Populations were sampled from two habitats. The first location was along a foot path to Yemoji Niver, about one kilometre to the shrine in Ilese, five kilometres outside 1jebu-Ode. The second site was the outskirts of Ore from Benin end of Ore - Benin Road. These areas are in the forest zone, and the samples were collected from shaded areas.

<u>C. alignifolium</u> has short compressed 'pseudostem'. They have long sheathing petioles and lanceolate leaves.

plant and needed high relative humidity. The specimens planted in pots and placed in the green house were stunted with small leaves. On the other hand, those potted and placed under tree **shade** in the nursery grew well and were bigger. The specimens had shorter petioles in cultivation.

### Chlorophytum blepharophyllum

Populations were sampled from different areas in the northern parts of the country. A population of this species was found on the left side of the road, just about ten kilometres from Jebba bridge on Jebba - Mokwa Road. Others were found in Ahradu Bello University Research Station site, Mokwa, along Bukura - Wamba Road about two kilometre to Gimi, and on the outskirts of Fadan Ayu. The vegetation of these areas has been disturbed in one form or the other, but it could be referred to as a derived Savanna or woodland Savanna. The common trees are species of <u>Ficus, Parkia, Gussonia</u> and <u>Combretum</u> while <u>Andropogon</u>, <u>Pennisetum</u> and <u>Hyperrhenia</u> species are the counton grasses.

The leaves of the species were lanceolate and crect; the lamina was hairy and with ciliate margin. The apex was sharply acuminate, the leaf base was not mottled, but ensheathing. The species had roundish 'pseudostea' formed by leaf sheaths. Dry hairy structures arose from the short stem.

Under cultivation, it was observed that the extent of hairings of the leaf surface reduced and the dry hairy structures from the stem disappeared. Some of the specimens lost the formation of 'pseudostems' especially after collecting their root-tips for mitotic study. However, the leaves were still erect. The root tubers were less developed during rainy season and many roots had none whereas almost all roots had root tubers in the dry season in their natural habitats.

## Chlorophytum larum R. Br.

Specimens were collected along Zaria - Jos New Road about 2 kilometres to Soba. They were growing among grasses of <u>Heteropogon</u>, <u>Paspalum</u> and <u>Perotis</u> species. Some scanty <u>Combretum</u> shrub species were also in the neighbourhood. The leaf was leathery with short petiole. There was not much difference in the species after cultivation. However, the leathery leaf became **fleshy and succulent** and definite petioles were developed. A compressed short 'pseudostem' developed in cultivation while the leaves just spread out in its natural habitat.

## Chlorophytum inornatum. Ker - Gawl.

Two populations were sampled. A population in the Botanical Garden, Ibadan grew on marshy soil among banana trees. The other population was sampled along a foot path on the outskirts of Jebba from Ilorin. The population was growing on marshy bank of a stream. The leaves, which are petiolate, spread out just above the soil level. The inner lower part of the leaf petiole was pinkish. In cultivation, no morphological variation was observed. However, by regular watering, the flowerin period was Prolonged in cultivation than in their natural habitat. Chlorophytum macrophyllum. (A. Rich.) Ascheps.

Cultivated specimens were collected by me from a population along Oyo - Iseyin Road and at the Onigambari, opposite the Federal Forestry Reserve along Ijebu-Ode - Ibadan Road. These areas are secondary forests showing past disturbances. Other samples were collected from the North around Wushishi on Tegina - Bida Road, and between Wamba and Akwanja, about fifteen kilometres to Akwanga. One <u>Chlorophytum</u> stand wes collected on the outskirts of Naraguta from Jos, on Jos - Bauchi Hoad. It was found among grasses.

In cultivation, there were no observable morphological changes. However, the specimen collected at Naraguta did not flower in time though it grew luxuriantly and propagated vegetatively. This specimen had been confirmed as <u>C. macrophyllum</u>. When some of its representatives were subjected to dry condition, (that is, by wetting them once in a week) they flowered and formed few fruits. Chlorophytum stenopetalum. Bak.

Populations were sampled from Eucalyptus plantation twentyfour kilometres to Kaduna from the South, around Mokwa, Samaru,

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Zuntu, Zungeru and Bukuru. They were usually found in cool, shaded areas especially in the Eucalyptus plantation.

The specimens were petiolate; the leaves spread out from the base and the lamina was arching. The leaves per plant ranged from five to eight, but usually six or seven. The roots usually had one median root tuber each.

A variety of this species, confirmed to key out to . <u>C. stenopetalum</u> Bak. , has ten to twelve leaves. It was collected about one hundred metres away from another population of <u>Chlorophytum</u> in Kaduna area, in an open habitat. The roots were long with rows of alternating and oppositely arranged small root tubers. This taxon is here referred to as <u>C. stenopetalum</u> II, for the purpose of reference.

The root morphology of the two taxa (<u>C. stenopetalum</u> I and II) is of interest. With a view to eliminating any possible microclimatic and/or soil chemical influence, representatives of the two taxa were planted in the same big plastic pots.

There were no observable changes in the morphological characters in <u>C. stenopetalum</u> I. However, the leaves of <u>C. stenopetalum</u> II became fresher, more succulent and more developed. The root morphology in both taxa was maintained, thus it was not environmentally induced.

#### Chlorophytum togense. Engl.

<u>C. togoense</u> is the most commonly distributed species in Nigeria according to data collected from herbarium specimens. However, few stands were collected on Mokwa - Kainji Road in 1977. Populations were also sampled from Iseyin area.

There was no observable ecological adaptation. The species grew very well in cultivation. It flowers almost throughout the year if watered regularly.

#### Chlorophytum X

A <u>Chlorophytum</u> species was collected from cultivation. The collector is not known. Even though the present worker was conversant with this plant in its wild form, the known habitat had been destroyed.

This taxon was formerly taken to be <u>C</u>. etatum R. Br., a native of South Africa. The photograph of <u>C</u>. elatum presented by Storey (1968) is different worphologically from the present material. Moreover, <u>C</u> elatum occasionally flowers in South Africa (Storey, 1968) while the present material flowers throughout the year and occasionally forms fruits with seeds. <u>C</u>. elatum is a tetraploid with 2n = 28, 2n = 32 and 2n = 24 (Deshpande, 1955b; Storey, 1968; and Koul, 1970). The material studied is a diploid with 2n = 16. The possibility of a tetraploid becoming a diploid in cultivation is very doubtful.

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However, <u>A. variegatum</u>, Hort. <u>A. vittatum</u>, Hort. <u>A. picturatum</u> Hort. <u>A. williamsii</u> Hort. are synonyms to <u>C. elatum</u> R. Br.. Hence the present material could be one of these synonyms.

On the other hand, the material shows some features of <u>A. bichetii</u>, Hort., a West Tropical African species (Bailey, 1947). Since the description is very brief, one is not sure whether it should be <u>A. bichetii</u>. The two taxa have flexible, varigated leaves with white and elegant habit.

Because of the unsettled taxonomy of this taxon, it is referred to as <u>Chlorophytum</u> X until its identity is confirmed 194. Kew.

#### RESULTS

i. Ecological Survey:

Figure 1 shows map of Nigeria and the routes followed during field collections and ecological survey trips. Figure II shows the ecological distribution of <u>Anthericum</u> and Chlorophytum in Nigeria,

The distribution table for <u>Anthericum</u> and <u>Chlorophytum</u> species in Nigeria is presented as <u>Appendix</u> I.

#### ii. Soil Analysis:

Data from soil mechanical and chemical analysis are presented as Appendices 2 to 4.

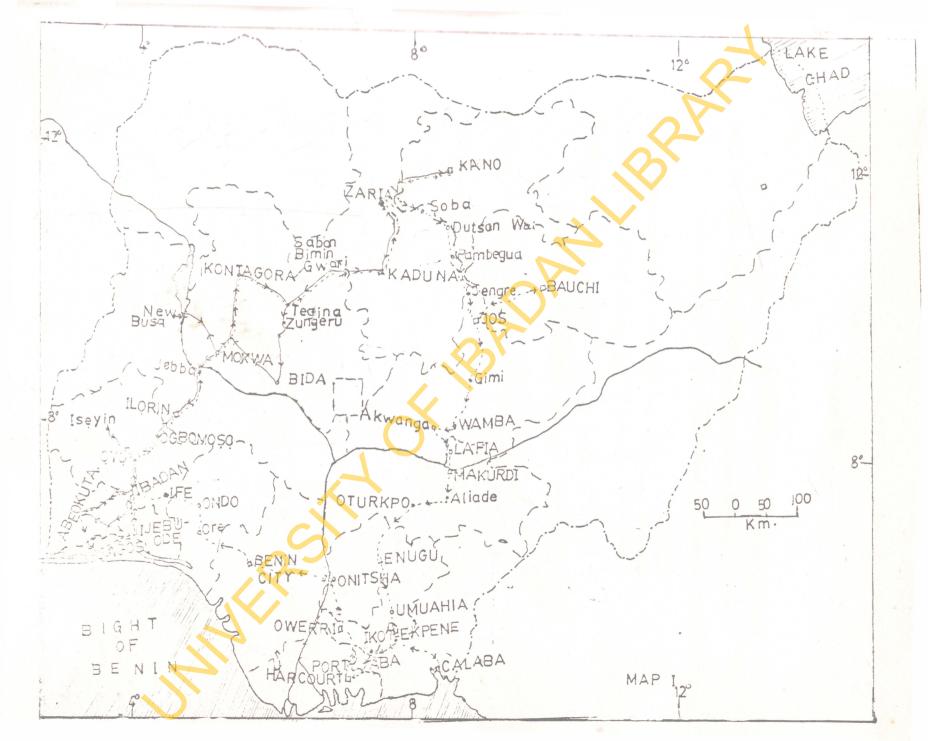
# FIGURE I

NERSI

Map of Nigeria showing areas covered during the field surveys and collection trips in sixteen states of Nigeria.

shows the routes followed during local trips and first long trip to the Northern part of Nigeria. ----- shows the route followed during the 1979 long trip. or > shows the direction of movement.

and > shows that the same route was taken to and fro.



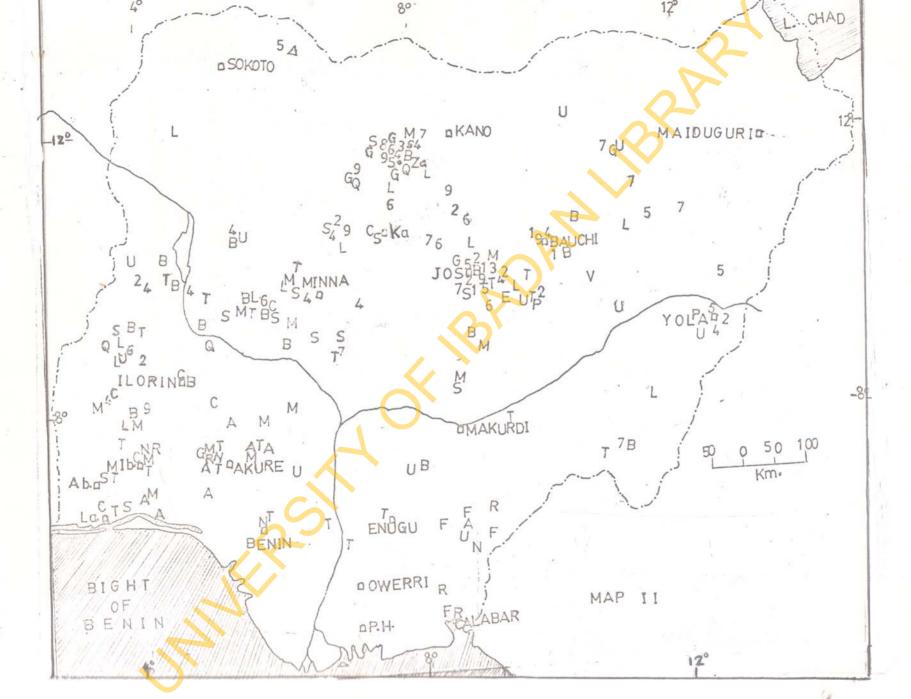
#### FIGURE II

Map of Nigeria showing the ecological distribution of <u>Anthericum</u> and <u>Chlorophytum</u> species in Nigeria. Arabic numbers and  $\triangle$  sign are used for Anthericum species. Letters of the alphabet are used for <u>Chlorophytum</u> species.

The Anthericum species are:

- 1. <u>A. dalzielii;</u> 2. <u>A. limosum;</u> 3. <u>A. nigericum;</u>
- 4. A. nubicum; 5. A. pterocaulon; 6. A. pubirhachis;
- A. <u>speciosum</u>; 8. <u>A</u>. <u>subpetiolatum</u>; 9. <u>A</u>. <u>uyuiense</u>; and △ A.zenker!
   The species of Chlorophytum are
- A. <u>C. alismifolium</u>; B. <u>C. blepharophyllum</u>; C. <u>C. caulescens</u> (<u>A. caulescens</u>); D. <u>bequaertii</u>; F. <u>C. sparsiflorum</u>,
- G. C. geophilum E. C. aureum; L. C. laxum; M. C. macrophyllum
- N. C. inornatum; P. C. pusillum; Q. C. gallabatense
- R. C. orchidastrum; S. C. stenopetalum; T. C. togoense;
- U. C. tuberosum; V. C. phystachys

The towns are Ab. - ABEOKUTA; Ib. - IBADAN; La. - LAGOS; Ka. - KADUNA; Za. - ZANLA: and P.H. - PORT HARCOURT.



#### E. DISCUSSION:

The ecological influences on <u>Anthericum</u> and <u>Chlorophytum</u> species has been reported by many authors (Obermeyer, 1962; Hanid, 1974; Marais and Reilly, 1978). Thus a comparative ecological study **has** enabledone to find out which of the ecological factors **readily** affect these plants. It has been observed that the ecological influences on these plants can be considered from two points of view:

- i. the environmental factors and
- ii. the influence of soil chemical composition, which may affect the ecological distributions of these plants.

#### Environmental Influences:

Species of both genera show a morphological response to changes in the environment. It is observed that shade, temperature and relative humidity affect the morphology of these plants.

The response of <u>C</u>. <u>alismifolium</u> put under tree shade, a few metres from the greenhouse and those in the greenhouse gives a good example. Those in the shade grew very well with relatively longer petioles and larger laminae. Those in the greenhouse had short petioles and smaller laminae. This can be due to the fact that those in the shade had lower rate of transpiration because the area was cooler than the greenhouse. It therefore means that those could afford to expose larger leaf surface area since the transpiration would be less. Those in the greenhouse needed to reduce their leaf surface area to cut down excess transpiration. Larger and more green lamina would result in higher production of carbohydrates through photosynthesis; thus more energy would be made available to plants in the shaded area. On the other hand, small leaf surface area of those in the greenhouse would result in comparatively low photosynthetic products and lower energy for vegetative growth. Thus they were stanted in growth. This phenomenon was also observed in <u>C. macrophyllum, C. laxum, C. inornatum</u> and <u>C. togoense</u>. The openness or shadiness of habitats had little or no influence on <u>Anthericum</u> species. This could be closely related to their habitats which were usually open fields in the savanna zone.

The specimens in the shade developed longer leaf petioles while those in the greenhouse developed shorter leaf petioles. <u>C. laxum</u> collected from the open field in the North had no definite and distinctive petiole in nature. In cultivation under the shade, the same specimen developed well defined and distinct leaf petioles. The possible reason could be that the production of longer petiole in the shaded area helped the specimens to display their leaves. This will enable them to maximize the use of the light penetrating through the shade for photosynthesis. Since the specimens in the open get even more than enough sunlight, the production of long petiole is not necessary. <u>C. alismifolium, C. macrophyllum</u> <u>C. stenopetalum</u> and <u>C. inornatum</u> show this ecological adaptation.also.

The development of hairy structures or shining leaf surface is a device to check excessive transpiration. This is also an environmental adaptation. <u>C. caulescens</u> and <u>C. blepharophyllum</u> collected from the North had hairy leaf surfaces. When these specimens were cultivated in the Nursery, the hairy structures reduced greatly and eventually disappeared under shade. Thus the possession of hairy leaf surface is a means of checking excess transpiration in an area where the supply of water is scarcely sufficient. This adaptation was therefore found unnecessary when the water supply was sufficient.

It has been observed that environmental ecological factors generally affect the size of leaves and the presence of hairs on the leaf surfaces. Presence or absence of petiole or relationship of petiole to heaf blade is also ecologically influenced. Thus the use of any of these characters for taxonomic purpose will eventually create taxonomic problem which is now a feature of this complex. A specimen of <u>C</u>. inornatum or <u>C</u>. alismifolium with little or no petiole may not be considered as the same species with one which has long, well developed petioles. In the same way, a <u>C</u>. blepharophyllum without hairy leaf surface may be considered as another species different from those with hairy leaf surface. There is evidence in this complex where two or more taxonomic names have been given to representative specimens of the same taxon. Hence Obermeyer, (1962) reduced five species of <u>Chlorophytum</u> to synonyme. Hanid (1974) enlarged the representatives of <u>C. affine</u> by adding nine species of <u>Anthericum</u> and two species of <u>Chlorophytum</u>. Hencer (1968) also reduced twenty-four species of <u>Anthericum</u> in Nest Tropical Africa to fourteen synonyms and fifty species of <u>Chlorophytum</u> to twentyone species.

It must be noted that the marginal ciliation in <u>C</u>. <u>blepharophyllum</u> is not ecologically induced. The cultivated specimens continued to exhibit the marginal ciliation of their leaves. <u>Soil Mechanical and Chemical Influence</u>:

Soil physical composition and its chemical contents have great influence on plants growing in it. <u>Anthericum</u> and <u>Chlorophytum</u> taxa show physiological responses to soil chemical composition. Since the soil composition gives no clue to the problem at hand, the data are only presented in the Appendix.

However, soil magnesium concentration seems to influence the distribution of these plants. <u>A. limosum, A. pterocaulon</u> II and <u>A. pubirhachis</u> have their soil  $Mg^{++}$  concentration ranging from 0.66me/100g to 0.82me/100g. On the other hand, <u>Chlorophytum</u> species

are found in soil with relatively higher Mg<sup>++</sup> concentration ranging from 1.24me/100g to 3.51me/100g. It is evident that <u>Anthericum</u> thrives well in soils with less than 1.0me/100g Mg<sup>++</sup> while low soil Mg<sup>++</sup> concentration (i.e. below 1.0me/100g) limits, at least in part, the growth of <u>Chlorophytum</u>.

Mg<sup>++</sup> is an essential constituent of chlorophyll. It is also involved in enzymatic reactions (Truog, et. e1, 1947) •

#### CHAPTER 3

# MORPHOLOGICAL STUDIES

Morphological or phenotypic characters are the attributes of any organism which are commonly used in identifying the organism. They are manifestations of the interplay of genes. cytogenes. and the factors. Hence no meaningful relationship can environmental be drawn between two species or genera without considering their phenotypic characters. Thus the morphological studies will enable one to determine distinguishing characters of each species which can be found useful for hybridization and cytogenetic studies of the two genera under consideration. This study can also be found useful in solving the taxonomic problems of the group of related genera in which Anthericum and Chlorophytum are included. To this end, the morphological characters of the leaves, roots, floral parts and pollen grains were studied and analysed statistically. The qualitative morphological characters were also investigated so as to find out whether such character(s) could be useful as diagnostic character(s) for each species and/or genus. Morphological characters were defined from the leaf apex, leaf . shape, leaf margin, type of leaf base, colour of leaf base, type of shoot, shape of 'pseudoster:,' location of the root tuber(s) location and shape of peduncle, type of inflorescence, colour of tepals and anthers. Other characters investigated were the colour and shape of fruits. disposition of tepals at pollination and number of tepals.

A. METHODS:

The morphological characters of cultivated representatives of each species, planted under the same experimental condition, were studied. This was done in order to reduce the possible environmental influences on the morphology of the plants.

Leaf morphological characters, inflorescence and root lengths were measured with one-metre rule and recorded in centimetres. Floral parts were measured with transparent six-centimetre rule and recorded in millimetres. The circumference of root-tubers was measured with already graduated strip of paper wrapped round it and recorded in cm.

(a) Fresh Leaf Morphology

The adult leaves of mature plants were used for this study because quantitative morphological characters of leaves change with the age of the plants. This also gave the same basis for comparison of the data collected. Hence, from each leaf handled were recorded the length, width, index, number of veins, and the vein spacing intervals. Records were taken for fifty loaves of each species, from not less than twenty different plants, except A. pubirhachis.

Leaf length was measured from the end of the leaf sheath or petiole to the apex of the leaf, and recorded in cm. The mean value was calculated.

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The widest part of each leaf blade, usually around the middle, was measured and recorded, in cm. against the length of the leaf. The mean value was determined.

The leaf index was calculated as the ratio of length to the width

# i.e. Length Width

The number of veins was counted across the widest part of the leaf blade with the aid of X10 hand lens. The mid-rib was not counted. The number was recorded as usual.

The spacing of veins in each leaf was calculated as follows:

# Leaf Width x 10 mm.

where the added 2 represented the two marginal spaces. The vein spacing interval was recorded in millimetres. The mean value was calculated.

(b) Morphology of Preserved Leaf.

Measurements from herbarium specimens were taken across different habitats, stages of developments and varieties of each species. Counting of voins on leaves was equally randomly sampled. Specimens at the herbaria of the University of Ibadan, University of Ife and those at the Forest Research Institute of Nigeria. Ibadan were all investigated. However, data for <u>A. moniliforme</u> was collected from only three individual representatives.

#### C Root Morphology

i. Root length, tuber length and tuber circumference were measured, as previously outlined, and recorded for each root in each specimen studied. Five to ten roots were handled in each specimen and a total of 100 roots was handled in each species. The mean values were calculated and recorded. The number of tuber(s) was recorded for each root.

# ii. Ratio of Root Tuber: Root Length

The ratio of root tuber to the whole length of each root was calculated by dividing the root tuber length by the length of the root. The quotient was multiplied by 100 and thus recorded as percentage. The mean for each species was calculated and recorded.

## D Floral Morphology

The length of at least twenty peduncles was taken for each species after the fruits had been developed or after flowering in such cases where fruits were not formed. This was because the peduncle should have reached its maximum elongation after the fruits had been formed or all flowers aborted. It was measured from the base to its peak. The range and mean were found and recorded in centimetres.

The floral parts were measured at flower anthesis. The tepals, stamens and pistils were removed carefully and their full lengths were measured. The ranges and means of each character for each species were determined and recorded in millimetres.

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<u>Pollen Grains.</u> The pollen grains were dusted on clean glass slides from dehisced anthers. A drop of 2% acetocarmine stain was dropped and then carefully covered with clean coverslip. The pollen grains were then measured at x40 objective of a light microscope. A unit of the micrometre eyepiece graticule equalled 2.5 microns at this magnification. The micrometre units were then converted into microns. The range and mean for the length and width (or diameters) were determined and recorded in microns.

The index of each pollen was found by dividing the length by the width. The mean index and range were found and recorded.

## E QUALITATIVE MORPHOLOGICAL CHARACTERS

Unlike the quantitative characters which were studied at plant maturity, qualitative characters were studied right from the germination of seed or sprouting of new shoot to the end of flowering or fruiting. This was done so as to cover the whole life cycle of each species in order to investigate whether any character(s) were due to stages of development and/or seasonal variations. Records were kept for at least twenty representatives of each species (except <u>A. pubirhachis</u>). These records were then used in compiling the qualitative characters as stated above.

The root structures for the plants were examined at different stages of development. The location of the root tuber(s) and number per root were noted.

# General Observations

The plants in this complex have six tepals; three large while the other three are relatively smaller in width. A careful observation of the tepal arrangement reveals that most <u>Chlorophytum</u> species have the larger tepals in the outer whorl while the smaller ones are inside. In the species of <u>Anthericum</u> and <u>Chlorophytum</u> X, the smaller tepals are outside while the larger ones are in the inner whorl. These arrangements could have been a decisive separating factor for the two genera, but <u>C. togoense</u> has the two types of arrangement.

The tepal disposition at the time of pollination is very interesting. In <u>A. limosum</u>, <u>A. pterocaulon</u>, <u>A. pubirhachis</u>, <u>Chlorophytum</u> X and <u>C. alismifolium</u>, the tepals spread out to the base and later close up after pollination. In the other species of <u>Chrorophytum</u>, the flowers open and the tepals bend backwards to expose the whole pistil and stamens during pollination. After pollination, the tepals close up again. That is, the open flower is rotate in <u>Inthericum</u> but companulate in <u>Chlorophytum</u>.

#### RESULTS

The vegetative morphology of representatives of the species of Anthericum and Chlorophytum investigated are presented in Plates I and II

Table 1 shows the leaf morphological data for ten Chlorophytum and three Anthericum taxa.

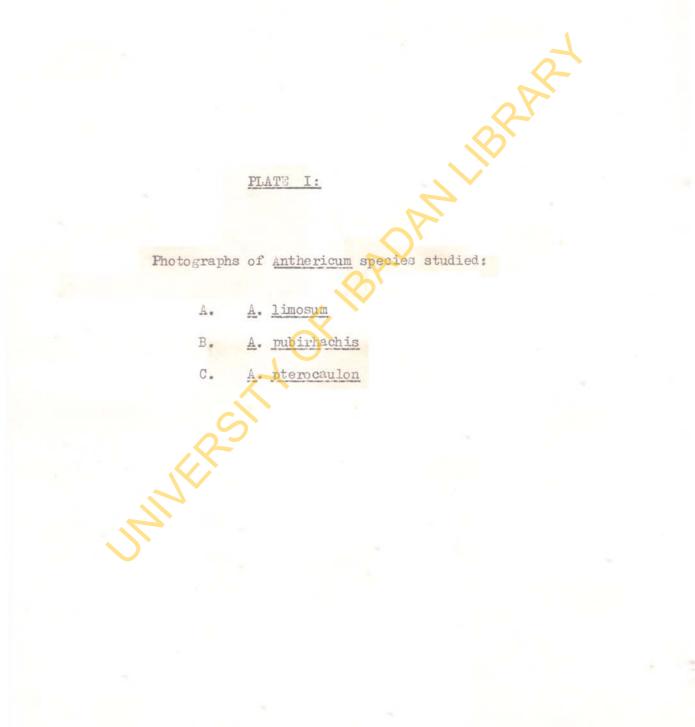
Table 2 contains some of the data collected from preserved herbarium specimens of all known species of the two genera in Nigeria, so as to highlight the possible usefulness of leaf index and vein spacing intervals in these plants.

Table 3 contains the root morphological data of ten <u>Chlorophytum</u> and three <u>Anthericum</u> taxa.

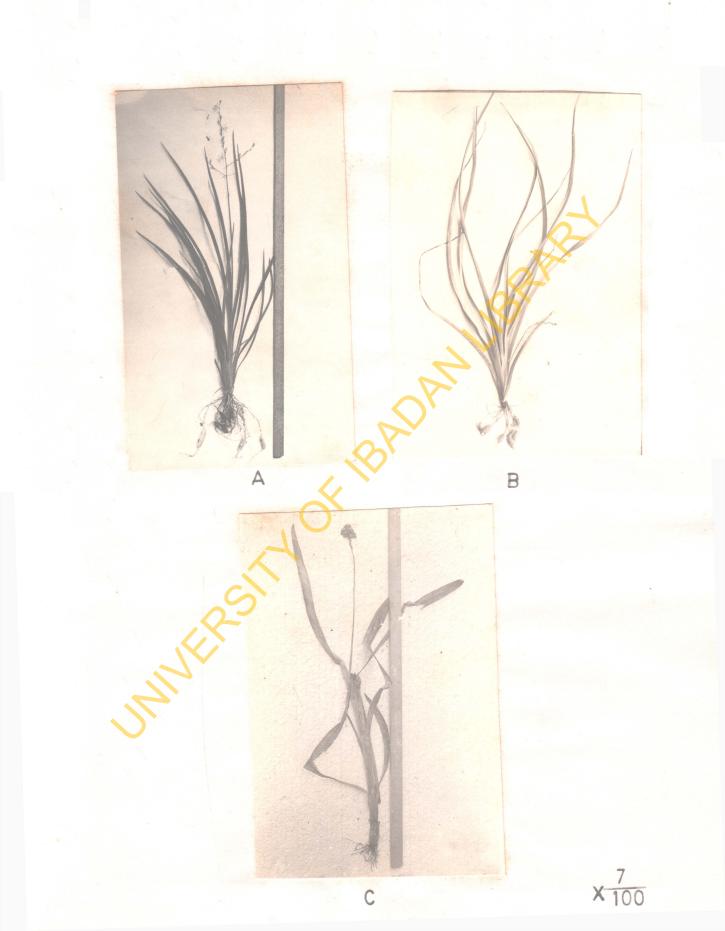
Table 4 shows data for the reproductive shoot of some representa-

Table 5 gives the measurements of the pollen grains in the plants handled.

Table 6 shows the summary of the qualitative characters for some species of the two remera found in Nigeria.



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# PLATE II

Photographys of Chlorophytum species studied:

A.	Chlorophy tum	blepharophyllum
в.	11	mac rophyl lum
C.	n	<u>inornatum</u>
D.	"	laxum
E.	) <b>"</b>	stenopetalum I
F.	н	<u>stenopetalum</u> II
G.	11	togoense
H.	n	alismifolium
Ro	"	caulescens (A. caulescens)
r.	A	Chlorophytum X

Note the root systems in <u>Chlorophytum stenopetalum</u> I and II, their inflorescence and number of leaves.







0 D		HIDIT AST	(cm.)		THE REAL PROPERTY IN THE REAL PROPERTY INTO THE REAL PR	(cm)		LINE DIDEC			NO. OF VEINS/	1 Sec.		VEIN SPACING	BO. OF LEAF/		HO. OF LEAF/		
O HARACTER		NO. OBS.	RANCE	MEAN	No. 023.	RANCE	NUX	NO. OBS.	RANGE	TAEX	NO. 033.	RANGE	MEAN	VEIN SPACING INVERVAL (mm.)	NO. OB3.	PAT CP	MEAN		
<u>blepharephyllu</u>	<u>c</u>	50	30.6-48.0	37.2	50	2.5-4.7	3.9	5	8.14-10.71	9.56	50	26-30	27.5	1.32	8	6-8	7.3		
macrophyllum	er.	8	34-51	43.83	- 50	5.4-8.0	6.88	8	5.0-7.91	6.41	50	26-42	31.5	2.05	20	7 - 11	8.8 -		
inornatum	<u>c</u>	50	22.1-25.8	24.19	50	4.4-5.7	5.17	50	4.24-5.05	4.69	50	22-28	25	1.91	20	7 - 9	- 8		
<u>laxum</u>	<u>c</u>	50	12.5-18.3	15.27	50	1.25-1.8	1.34	50	8.63-11.54	9.93	50	22-28	25.8	0.48	20	7 - 9	8.3		
. <u>stenopetalum</u> I	; <u>c</u>	50	22.4-30.4	26.99	50	3.8-4.9	4.32	50	5.0-6.67	6.25	50	24-36	31.2	1.30	20	01 1 05	6.7		
. <u>stenopetalum</u> I		2 2 2 2	21.5-31.5	26,32	50	3.7-5.3	4.61	50	5.11-6.1	5.7	50	40-45	43	1.02	20	10 - 16	14.3		
. <u>torcense</u>	<u>c</u>	50	18.0-35.6	25.46	50	2.8-4.9	3.69 -	50	6.03-9.37	6.88	50	16- 22	20.5	1.64	20	5 1 8	6.6		
. <u>aliamifolium</u>	c	r 4 0 7	5.4-8.3	5.6	50	2.4-3.7	3.05	50	1.77-2.52	2.18	50	20-22 .	20,88	1.33	20	7 9	8.4		
hlorophytum X	3 0	50	12.2-19	15.62	50	0.8-1.3	1.18	50	11.65-15.25 7.18-12.13	13-24	50	20-24	21.6	0.50	20	8 - 10	9		
. <u>caulescens</u>	3		Ú.	34.05	50	2.1-4.0	3.48	50	7.18-12.1	9,73	50	24-28	25.8	1-35	20	13 - 15	13.8		
. <u>limomm</u>	A		30.5-56.1	41.69	50	0.5-1.1	0.85	55		50.86	50	20-24	12	0.37	8	10 - 13	11.7		
. <u>pterocaulon</u>	-		0	47.29	50	1.1-2.8	1.77	50	33.89-68.82 21.85-31.05 58.3-79	26.76	50	48-60	54	0.32	20	0 1 0	6.5		
. Inbirhachia	Δ	50	18.0-42.0	8.3	50	0.3-0.6	0.42	50	5 58.3-79	66.32"	50	8 - 12	19.5	0.37	10	10 - 13	 		

TABLE 1. LEAF MORPHOLOGY

#### LEAF MORPHOLOGY OF HERBARIUM SPECIMENS OF ANTHERICUM AND CHLOROPHYTUM SPACIES IN NIGERIA

	LENGTH	OF LEAF		WIDTH	OF LEAF		NO.	OF VEINS	3	7	
SPECIES	NO. OBSERVED	RANGE (cm.)	MEAN (cm.)	NC OBSERVED	RANGE (cm.)	MEAN (cm.)	NO. OBSERVED	RANGE	HEAN	INDX	VEINS SPACING INTERVAI (mm.)
C. aureum	50	19-24	20.2	50	2.8-3.0	2.9	50	24-32	28	6.97	0.97
C. alismifolium	50	6-15	11.5	50	1.4-3.6	2.8	50	18-24	18	4.11	1.40
<u>V. bequaertii</u>	50	16-20	18.8	50	1.5-2.1	1.7	50	14-18	16	11.06	0.94 .
C. blepharophyllum	50	30-41	38.5	50	1.5-3.2	3.05	50	20-28	26	12.62	1.08
C. gallabatense	50	4.5-5	4.8	50	2.6-3.2	2.9	50	26-30	28	1.66	0.97
C. geovhilum	50	10-24	17	50	2.1-4.3	3.2	50	24-30	26	5.31	1.14
. inornatum	50	11-37	19.6	50	2.6-4.8	3.5	50	16-26	20	5.6	1.59
C. laxum	50	7-16	12.1	50	0.6-1.4	-1.1/	50	16-30	24	11.0	0.42
C. macrophyllum	50	10-62	32.2	50	2.1-8.4	6.8	50	26.44	32	4.73	2.00
. orchidastrum	50	9-21	13.2	50	2-2.6	2.2	50	16-20	18	6.0	1.10
. sparsiflorum	50	10-34	19.5	50	1.6-8.8	6.1	50	16-26	24	3.2	2.35
. stenopetalum	50	12-31	19.5	50	1.6-2.4	2.3	50	16-20	19	8.48	1.10
. togoense	50	20-30	23.6	50	3.2-4.6	3.9	50	20-30	26	6.05	1.39
. tuberosum	50	7-16	12.8	50	1.3-2.8	1.8	50	12-18	16	7.11	1.0
C. caulescens	50	14-36	30.1	50	1.0-3.4	3.05	50	24-30	28	9.87	1.02
. subpetiolatum	50	24-36	25.2	50	1.2-3.5	2.6	50	16-20	18	9.69	1.30
A. <u>dalzielii</u>	50	10-15	13.7	50	0.1-0.2	0.12	50	8-12	9	114.2	0.11
. limosum	50	16-33	27.2	50	1.0-1.5	1.1	50	26-34	30	24.73	0.34
. <u>nigericum</u>	50	10.20	1,6	50	0.1-0.2	0.11	50	6-8	7	145.45	0.12 .
. nubicum	50	30-69	35.2	50	1.1-1.3	1.2	50	28.34	32	29.33	0.35
. moniliforme	20	38-51	44.5	20	1.4-2.2	1.83	20	38-52	47	24.32	0.37
1. pterocaulon	50	20-40	35.2	50	1.1-1.6	1.3	50	44-54	46	27.08	0.29
. pubirhachis	50	29-42	40.1	50	0.6-0.8	0.72	50	2026	24	55.69	0,28
. specious	50	28-44	40.1	50	0.4-0.6	0.54	50	20-28	26	74.26	0.19
. <u>uyuiense</u>	50	47-52	49	50	0.5-0.8	0.6	50	10-26	16	81.67	0.33

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BATIO OF INSTR ROOT: ROOT LEVIT	✓ XEGNI 12ELL	(vin+)	ROOT/TUBER	an south	fame burness of the second	AUDINE AD	SINCE MUDELD	the strated on .	Tanala and the		-	2007 (cm.)	TENSIR OF	GHARACTERS
<u>T.T.</u> 3.L.	L.	MEAN	RANGE	NC. OB.	MEAN .	PANCE	NO. OB.	MEAN	RAN JE	NO. CB.	NEAN	PANCE	NO. OB.	13
16	52		ı	100	2.78	2.4-3.0	100	5.32	4.1-11.0	100	33.5	27-39	100	2. blephic- phr/lum
vi	96	1.3	1 - 2	100	3.4	2.4-5.0	100	3.56	2.5-4.8	100	69.47	27-5-110	100	2- C. macro-
19	97		i	100 .	3.95	3.4-4.5	oot	4.08	3.8-4.2	100	21.25	17.5-24	100	C. inor- natum
13	104	-	ı	100	2.24	1.6-3.4	100	2,15	1.3-2.8	100	16.34	7=21.5	100	C. laxum
vi,	197	-	ı	100	2.3	1.1-3.6	100	1.17	0.7-1.5	100	21.75	13:7-27-3	100	C. steno- pstalum I
7	70	.26	17-34	100	1.92	1.0-2.5	100	2.74	0.8-3.5	100	41	35-47	100	C. steno- petalum II
16	54	1	1	100	2.23	2.1-2.5	100	4.12	3.6-52	100	26.21	20.5-32	100	C. togo-
26	70	ij.	1 1 2	100	1.71	1.2-2.0	100	2.45	1.6-2.7	100	9.28	7- 11.8	100	C. alismi- folium
16	106	1.1	1 - 2	100	1.77	1.5-2.2	100	1.67	1.2-2.3	100	10.13	6.2-18.7	100	Chloro- phytum X
Ţ	1	1	t	1	1	j.	Į.	I.	t	1	37.48	28.1-45.2	100	C. caule-
15	84	. 14	1	100	2.39	1.8-3.1	100	2.84	1-4-4-1	100	19.22		100	A. lino-
12	128	4	ï	100	3.23	3.0-3.5	100	2.53	1.8-3.0	100	20.31	13.4-24.5 15.3-25.2	100	A. ptero- caulon
29	103	0.6	0 - 1	:00	2.51	1.7-3.4	100	2.43	1-3-3-1	100	8.35	1.0-12	100	A. mbirh- schiz

CHLOROPHYTUM AND ANTHERICUM ROOTS MORPHOLOGY

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#### REPRODUCTIVE SHOOT OF SOME CHLOROPHYTUM AND ANTHE ICUM SPECIES

CHARAC	OTERS	C. blepharo phyllum	- <u>C. macro-</u> <u>phyllum</u>	<u>C. inor-</u> natum	<u>C. laxum</u>	<u>C. steno-</u> <u>petalum</u> I	<u>C. steno-</u> <u>petalum</u> II	<u>C. toko-</u> ense	<u>C: alis-</u> mifolium	Chloro- phytom X	C. <u>caule-</u> scens	A. Minosun	<u>A. storo-</u> caulon	A. <u>subir</u>
ENGTH OF	NO. OBS.	20	20	20	20	20	20	20	20	20	20	20	20	10
FEDUNCLE (cm)	RANGS	27.5-41.2	15.0-20.0	21.0-31.5	16.0-33.0	6.0-8.5	7.2-13.0	30.5-76.0	9.8-11.0	10.1-12.5	56.0-69.5	60.7-76.0	96.2-110	50 - 80
	MEAN	31.5	17.8	26.5	19.9	7.5	10.9	61.7	10.3	11.75	61.3	67.8	108.3	72.5
20 Percent	NO. CES.	100	100	100	100	100	100	100	100	100	100	100	100	100
LENGTH OF TEPAL (mm) 3 MIDTH OF 3 TEPAL (mm) 4 TEPAL (mm) 4 LENGTH OF 5 FILAMENT(mm) 8 LENGTH OF 5 LENGTH OF 5	RATOR	6.0-7.0	7.0-8.0	6.5-7.5	5.0-6.0	5.5-6.0	5.5-6.0	11.0-12.0	5.5-6.5	7.5-8.0	12.0-12.5	11.0-12.0	10.0-11.0	8.0-9.0
	MEAU	6.8	7.5	7.0	5.5	5.75	5.75	11.5	6.0	7.67	12.2	11.5	10.5	8.5
	NO. OBS.	100	100	100	100	100	100	100	100	100	100	100	100	1.00
	RANCE	2.0-3.0	1.5-2.0	1.5-2.0	1.5-2.0	1.0-2.0	1.0-1.5	3.0-5.0	1.5-2.0	2.5-3.0	1.8-2.0	3.0-5.0	2.0-2.5	3.5-5.0
California ( more )	KEAT	2.25	1.75	1.75	1.75	1.5	1.25	4.0	1.75	2.75	1.93	4.0	2.25	3.83
TEFAL	IIDEX <u>V</u>	33	23	25	32	26	22	35	29	36	15.82	35	21	45
	NO. OBS.	100	100	100	100	100	100	100	100	100	100	100	100	100
	RANGE .	5.0-5.5	5.5-6.0	3.0-4.5	4.0-4.5	4.5-5.0	6.0-7.0	4.5-5.0	4.5-5.0	3 - 5	7.0-9.0	4.5-5.0	5.0-5.5	4.0-4.5
Thomas I amon	MEAN	5.25	5.75	3.86	4.15	4.75	6.21	4.75	4.75	4.05	8.0	4.75	5.25	4.25
THOTH OF	NO. OBS.	100	100	100 .	100	100	100	100	100	100	100	1.00	100	100
ANTINER (mm.)	RANGE	2.0-2.5	2.0-2.5	1.0-1.5	1.0-2.0	1.5-2.0	1.5-2.0	1.5-2.0	1.5-2.0 .	2.5-3.0	1.5-2.5	4.0-5.0	4.0.6.0	2.5-3.5
	YZAN	2.25	2.25	1.43	1.6	1.9	1.65	1.65	1.55	2.64	2.24	4.5	5.0	3.0
MEAN LENGTH O	P STANEN	6.38	6,88	4.48	4.95	5.7.	7.04	5.58	5.53	5.37	9.12	7,00	7.75	5.75
LENGTH OF	NC. 035.	50	50	50	50	50	50	50 .	50	50	50	50	50	20
STYLE (mm.)	RANG	4.0-5.0	7 - 9	3.0-4.0	3.0-3.5	5.0-5.5	6.0-7.5	4.0-5.5	4.0-4.5	5.0-6.0	7 - 9	11 - 13	8 - 11	6.5 - 8.
	NRATI	4.6	8.2	3.63	3.21	4.75	6.5	5.0	4.12	5.8	7.89	12.5	9.0	7.0
LENGTH OF	NC. CB3.	50	50	50	50	50	50	50	50	50	50	50	50	20
OVARY (mm.)	RANCE	1.5-2.0	2.0-3	1.5-2.0	1.0-1.5	1.0-2.0	1.5 - 2.0	2.0-3.0	1.0-1.5	1.5-2.0	1.5-3.0	2.0-3.5	2.0-2.5	2.0-2.5
	MRAT	1.96	2.25	1.85	1.15	1.55	1.8	2.45	1.1	1.62	2.4	2.5	2.15	2,1
FISTIL MEAN LE	51 225	6.56	10.45	5.51	4.36	6.3	8.3	7.45	5.22	7,42	10,29	15	11.15	9.1
≶ 07 STMED:	FISTIL	97.26	65.84	81.31	113.53	90.48	84.82	74.90	105.94	72.37	88.63	46.67	69.51	63.19
े दे रह स्वरसंखः	FILANENT	42,86	39.13	37.05	38.55	40.0	26.57	34.74	32.63	65.19	28.0	94.74	95.24	70.59
S OF OVERY:	SIVLE	42.61	27.44	51.79	35.83	32.63	27.69	49.0	26,70	27.93	30.42	20.0	23.69	30.0
	and the second se													

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	INDEX OF POLLEN TRAIN		FOLLEN TRAIN	SIDTE 03		FOLLET GRAIN	+ 07	LENGTH	CH ARACTERS
MEAN	RANGE	NO. OES	YZAN	RANCE	NO. OBS.	MEAN	RANGE	NO. OBS.	11873
1.38	1-16-1-4	100	34.87	31.0-38.8	100	43.18	45-52	100	C. blerharonhyllum
1.11	1.04-1.9	100	54.91	42.5-65	100	61.11	56.0-75	100	C. macrophyllum
1.14	1.0-1.7	100	36.74	32.5-41.5	100	41.88	32.5-45.5	100	0. inornatum
1.26	1.19-1.44	100	38.64	33.3-40	100	48.47	45-51-3	100	<u>C. laxum</u>
1.15 .	1.03-1.23 1.08-1.23	100	36.72	32-40	100	42.29	36.0-48	100	<u>C</u> . <u>stenopetalum</u> I
1.19	1.08-1.23	100	27.67	25-30	100	32.91	30-38	100	<u>C. stenopetalum</u> II
51-1	01	100	34.01	31-5-40	100	30.76	35 -42-5	100	C. togoense
1-14	1.05-1.24	100	36.83	32.5-45	100	- 42.06	35.5-50	100	<u>C. alismifolium</u>
1.25	1.08-1.44	, 100	25.71	20.5-34	100	31.71	25-38	100	Chlorophytum X
1.18	1.06-1.3	100	37.04	32-42.5	100	43.51	33-50	100	C. <u>caulescons</u>
1.26	1.21-1.42	100	34-37	32-5-35	100	40.06	37.5-45	100	A. limosum
1.21	1.01-1.31	100	36.42	32.5-40	100	445	42.5-47.5	100	A. <u>sterocaulon</u>
1.42	1.32 -1.6	100	35.06	34-35.5	100	50.35	49-55	100	A. mubirhachig

TABLE 5 FOLLEN GRAIN MORPHOLOGY

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QUALITATIVE MORPHOLOGY OF SOME NICERIAN SPECIES OF CHLOROTHYLUM AND ANTHONY

CELRACISES	G. hlevharovhyllum	C. macrophyllum	C. Inornatum	<u>c</u> . <u>1axun</u>	C. atenopetalum I	C. stemopetalum II	C. toroonse	G. alistifolium	Chlorophytum Z	C. caulencena	A. lineaum	A. Herosulon	A. Jubirhachis
LEAP APER	Acuminate	Acuminate	Acuminate	Acuminate	Acuminate	Acuminate	Acuminate		Acuminate	Acuminate	Acuminate	Acuminate	Acuminate
LEAP SHAPE			Lanceolate									Linear	Linear
LUAF MARGIN	Ciliate	Entire	Rntire	Entire	Entire	Entire	Entire	Sntire	Entire A White	Entire	Entire	Entire	Entire
TYPE OF LEAF BASE	Petiolate	Petiolate	Petiolate	Peticlate	Peticlate	Fetiolate	Petiolate	Sheathed	Sheathed	Sheathed	Sheathed	Sheathed	Sheathed
LOCATION OF INFLORESCENCE	Central	Central	Aril Central	Central	Central	Central	Central	Central	Central	Central	Central	Central	Central
COLOUR OF TEPAL	Brown	White	Green	White	White	White	Green	White	White	Brown	White	White	White
COLOUR OF ANTHER	Yellow	Yellow	Yellow	Yellow	Yellow	Yellow	Yellow	White	White	Yellow	Yellow	Yellow	Brown
COLOUR OF LEAF BASE	Green	Green	Pinkish	Green	Green	Green	Green	Green	Green	Green	Pink	Fink	Red Kottled
LOCATION OF BOOT TUBER(S)	Median	Alternate	Distal _	Proximal	Median	Alternate	Distal	Distal	Proximal	None	Median	Distal	Distal
TYPE OF INFLORESCENCE	Raceme	Raceme	Racene	Raceme	Raceme	Raceme	Panicle	Raceme	Raceme	Panicle	Panicle	Panicle	Paricle
TOTAL DISPOSICION A7 POLLINATION	Bent Backward	Bent Backward	Bent Backward	Spread out	Bent Backward	Bent Backward	Bent Backward	Spread out	Spread out	Bent Backward	Sproud out	Spread out	Spread out
TUPAL ARRANGEMENT	Large Petals in Outer part	Large Petals in Cuter part			"Large Petals in Outer part					Large Petals in Outer part			
FEULT SHAPE	Deeply Lobed	Deeply .Lobed	Deeply Lobed	Shallowly Lobed	Deeply Lobed	Deeply Lobed	Deeply Lobed	Deeply Lobed	Deeply Lobed	Shallowly Lobed	Shallowly Lobed	Shalloyly Lobed	Shallowly Lobed
SEAPE OF PEDDNELE	Circular	Circular!	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Flattened	Flattened	Flattened
SEAFE OF PSEUDOSTEN	Circular		-	Flattened	-	-	Circular	Flattened	Flattened	Circular	Flattened	Flattened	Flattened

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The morphological studies on the three taxa of <u>Anthericum</u> and ten taxa of <u>Chlorophytum</u> reveal much overlappings and similarities. Data collected from ten species of <u>Anthericum</u> and fifteen species of <u>Chlorophytum</u> from herbarium specimens equally confirm the similarities of the two genera.

Data in Table 1 reveal the overlapping of the leaf lengths, leaf widths, number of veins per leaf and number of leaves per plant. These overlapping characters can not be very useful as distinguishing characters for genetic studies. The type of leaf margin is common to the two genera except <u>C</u>. <u>blepharophyllum</u> which has ciliated margin and <u>Chlorophytum X</u> with hyaline margin. The vein spacing intervals across the leaf blade and even the leaf index can be used to separate the two genera.

The leaf index of the taxa divide them into three groups. The first group is made up of taxa with their leaf indices above 20 (i.e. Group A). The intermediate group (i.e. Group B) is made up of plants having their leaf indices between 10 and 20. Whereas, the third group (Group C) consists of taxa having leaf indices below 10. Using Tables 1 and 2 (i.e. data from both fresh and herbarium specimens), the taxa in this complex can be separated as follows:

- 50 -

Group A plants: <u>A. dalzielli; A. limosum; A. nigericum; A. nubicum;</u> <u>A. pterocaulon, A. pubirhachis, A. specious;</u>

A. uyuiense, and A. moniliforme.

Group B plants are: <u>Chlorophytum X, C. bequaertii, C. laxum</u>, and C. blepharophyllum.

Group C plants are: C. alismifolium, C. gallabatense, C. reophilum,

C. mornatum, C. macrophyllum C. orchidastrum,

C. aureum, C. tuberosum, C. caulescens (A. caulescens);

A. subpetiolatum, C. Jaxum and C. blepharophyllum.

The double grouping of <u>G</u>. <u>laxem</u> and <u>C</u>. <u>blepharophyllum</u> can be considered from two points of view. Data from fresh specimens place the two species in group C. while data from preserved specimens place them in group B (see Tables 1 and 2 respectively). The varieties dealt with in the three herbaria were invarious stages of development and sizes, while the cultivated specimens were almost of the same variety and stage of development. It is therefore reasonable that the grouping should vary in the two tables. The second possible reason is that the herbarium specimens shrink more breadth-wise than lengthwise because the mid-ribs and veins normally check: longitudinal reduction in size as a result of drying. This then results in increased indices in the spocies since the widths are relatively more reduced.

These plants are all affected by one or both reasons given above. However, it is interesting to note that the grouping in 88% is the same. This thus means that the leaf index is a reliable dividing character for this group of plants. Using this criterion places all the <u>Anthericum</u> species in Group A except <u>A. subpetiolatum</u>. On the other hand, all the <u>Chlorophytum</u> species with <u>G. caulescens</u> (i.e. <u>A. caulescens</u>) and <u>A. subpetiolatum</u> are placed in Group C. (except <u>Chlorophytum</u> X and <u>C. bequaettii</u>). Though, this criterion can not be taken as sufficient at the moment for transferring species from one genus to the other, it could meinforce other criteria.

A careful examination of vein spacing intervals (VSI) presented in the 13th column of Table 1 and in the 11th vertical column of Table 2 shows two groupings of the plants in the complex. Group A is made up of plants having their VSI less than 0.5 mm., and Group B plants have their VSI from 0.5 and above.

Group A plants are <u>A</u>. <u>dalzielii</u>, <u>A</u>. <u>limosum</u>, <u>A</u>. <u>nubicum</u>, <u>A</u>. <u>nigericum</u>, <u>A</u>. <u>pterocaulon</u> <u>A</u>. <u>pubirhachis</u>, <u>A</u>. <u>specious</u>, <u>A</u>. <u>uyuiense</u>, <u>A</u>. <u>moniliforme</u>, <u>Chlorophytum</u> X. and <u>C</u>. <u>laxum</u>

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Group B plants are <u>C</u>. <u>aureum</u>, <u>C</u>. <u>bequartii</u>, <u>C</u>. <u>alismifolium</u>,

<u>C. blepharophyllum, C. geophilum C. gallabatense,</u> <u>C. inornatum, C. togoense, C. macrophyllum,</u> <u>C. orchidastrum C. sparsiflorum, C. stenopetalum I & II,</u> <u>C. tuberosum, C. caulescens (A. caulescens)</u> and A. subpetiolatum.

The grouping by the vein spacing intervals agrees with that of the leaf mindex. Thus the use of these criteria is somehow complementary and objective. The positions of the intermediates and the species of <u>Anthericum</u> grouped along with the <u>Chlorophytum</u> species still need further investigations.

The root quantitative morphology presented in Table 3 simply are shows that <u>Anthericum</u> and <u>Chlorophytum</u> genera/close relatives. There are many overlappings. Hence the usefulness of these root morphological characters for cytogenetic study and taxonomic separation is limited. However, qualitative properties of the root types (see Table 6) in the two genera, reveal three main root types. They are: Type I plants have alternating - oppositely arranged root-tubers. Taxa in this group are <u>C. stenopetalum</u> II and <u>C. macrophyllum</u> Type II) are plants with one or two root tubers per root. This type can further be divided into three sections:

- b: Those with root tuber(s) in the median area e.g.
  <u>C. stenopetalum I, C. blepharophyllum</u>, and
  A. limosum.
- c: Plants with root tuber(s) at the distal end of the root: <u>C. inormatum, C. alismifolium</u>,

<u>C. tokoense, A. pterocaulon and A. pubirhachis</u> make up this section.

Type III: Plant with thickened fibrous root system without most tuber. The only taxon with this type is C. caulescens.

It is worth noting that the three groupings found in the root types do not agree or complement the previous groupings under leaf index and vein spacing intervals. However, the qualitative root characters can be found very useful in genetical and cytogenetical studies.

Table 4 demonstrates the close relationship between the two genera. However, the mean length of anthers divides the plants in this complex into two groups:

Group I is made up of taxa having large anthers, whose anthers are 2.5 millimetres and above. <u>A. limosum, A. pterocaulon</u>, <u>A.</u> <u>pubirhachis</u> and <u>Chlorophytum</u> X are in this group. Group II plants have their mean anther lengths less than

2.5 millimetres. Taxa in this group are C. alismifolium,

C. blepharophyllum, C. inornatum, C. laxum, C. macrophyllum,

C. stenopetalum I and II, C. togoense and C. caulescens.

The percentage of anther as a ratio of the filament (ATRP) also divides the plants into two groups:

- Group I is made up of plants having their anther: filament ratio percentage above 50%. Plants in this group are <u>Chlorophytum X</u> and the Anthericum species.
- Group II plants have their anther: filament ratio percentage below 50%. These are the remaining Chlorophytum species.

The use of the anther: filament ratio percentage as a criterion A helps to solve the intermediate position of <u>Chlorophytum</u> X under the mean anther length grouping. In the latter, its value, which is 2.5 mm. forms the upper limit of <u>Chlorophytum</u> and the lower limit of <u>Anthericum</u> species. The use of AFRP decisively places <u>Chlorophytum</u> X along with the <u>Anthericum</u> species. Its AFRP value of 63% is above 50%, the lower limit of <u>Anthericum</u>.

It may be necessary to point out here that the grouping, resulting from the use of the mean anther lengen and AFRP values agree with that of leaf index and VSI groupings stated earlier. The pollen grain morphological data as presented in Table 5 only confirm the closeness of the two genera. The index for each taxon, which shows the shape of the pollen grains, overlaps. Hence this has limited use for the moment. It may be necessary to place it on record that some oily droplets were observed in <u>C. laxum</u> pollen grains. This is not found in other species. However, more detailed investigations and studies are hoped for in future research work.

The qualitative characters for the thirteen taxa of Anthericum and Chlorophytum genera presented in Table 7 give further confirmation of the relationships existing between the two genera. Most of the characters show overlappings, thus indicating their closeness. However, the anther colour varies in intensity from yellow to brown in both genera, but Chlorophytum X and C. nlismifolium have white anthers. Colour of leaf base is usually green in Chlorophytum species but those of Anthericum species tre pinlish. A. mubirhachis even has reddish mottling. The pseudostem in Anthericum species, Chlorophytum X, C. alismifolium and C. laxum is flattened or compressed. The pseudostem is circular in C. caulescens and other species of Chlorophytum when present. The shape of the peduacle show that A. limosum, A.pterocasion and A. pubirhachis have flattened and angled peduncles while those of C. caulescens and other taxa of Chlorophytum are circular. The shape of leaf for Anthericum is linear while that of Chlorophytum species is lanceolate.

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The vegetative morphological data analysed and presented above show that <u>Anthericum</u> and <u>Chlorophytum</u> have much in common. It has also been suggested that the two genera can be separated tentatively by using the leaf index, vein spacing intervals (USI), mean lengths of anthers, anther: filament ratio percentage (AFRP), shapes of leaf, shapes of peduncle, shapes of pseudostem and the tepal dispositions during pollination. Using these criteria, <u>Chlorophytum X</u> agrees more with the <u>Anthericum</u> species in many respects. However, <u>C. laxum</u> and <u>C. alismifolium</u> share their characters within the two genera, thus maintaining intermediate position between the two genera.

The locations of root tuber(s) on the roots, shapes of peduncle, the size and colour of anthers, tepal arrangements and tepal disposition during pollination have been noted as useful 'marker' characters for genetical and cytogenetical studies.

#### CHAPTER 4

#### ANATOMICAL STUDIES

Leaf surface patterns, anatomical features and structures have been demonstrated to be genetically controlled (Cutler, 1972; Cutler and Brandham, 1977). Hence, a serious cytogenetic consideration should include the anatomical features of the organisms that are being studied. Thus the anatomy of representative species of <u>Anthericum</u> and <u>Chlorophytum</u> was studied with a view to indirectly assessing the cytogenetic relationships of the two genera. It was hoped that this would help to confirm the separation of the complex into two distinct genera, merging them into one single genus or subgenera of the same genus. It is also hoped that the anatomical study might throw light on the probable mode of evolution of the complex.

#### METHODS:

A. LEAF EPIDERMIS:

The leaf surfaces were cleaned with lens tissue scaked in ' actone. This was to stimulate the proper opening of the stomata. The epidermis was removed mechanically from about the middle of the leaf blade. Good strips from the upper and lower surfaces were mounted in 50% glycerine for microscopic examinations.

i. The stomatal apparatus and the neighbouring cells were measured at X 10 and X 40 objectives of a light microscope with A previously graduated micrometer eyepiece graticule. One unit of graticule equalled 10.0 microns at X 10 and 2.5 microns at X 40 objectives. The measurements were recorded in microns. At least fifty stomata and their neighbouring cells were randomly selected from at least five specimens of each species and so treated. Mean values of the length and width of stomatal apparatus as well as those of the neighbouring cells were calculated.

ii. Index for each stoma and neighbouring ce'l was calculated individually.

iii. The number of stomata, using X 10 objective, was counted from at least five specimens of each species. Twenty such counts were made for each species. The mean per field of view of the microscope at X 10 objective at which one unit of micrometer graticule equits 10 Jan. was calculated as follows:

Area of a circle 
$$iir^2$$
.  
Diameter of field of view  $= \frac{10.0 \times 100}{1000}$  rm.  
the radius is  $\frac{1}{2}$   $(\frac{10 \times 100}{1000})$  mm.  
 $= \frac{1}{2}$  am.

Thus area of field view

at X 10 objective =  $\left(\frac{1}{2}^2 \times 11\right)$  sq. mm. = 0.785 mm<sup>2</sup>. The distribution of number of stomata per square millimetre was calculated thus:

 $(x \ge 1)$  stomata / sq. mm. Where x is the mean number of stomata per field of view y is the calculated area of the field of view

The product was approximated to the nearest whole number. It was then recorded as the number of stomata/sq. mm. for each species. iv. The mean length of the stomata was found as the ratio of the mean length of the neighbouring cells and the mean width. The mean index of stomata was also determined as a ratio of the mean index of the neighbouring cells.

If the stamata : Neighbouring cells ratio = A:B

\*\* % of stomatal ratio =  $\left(\frac{L}{A+B} \times 100\right)\%$ 

The product is then put in brackets above the ratio. This is done in order to have the same basis of comparison.

B. LEAF ANATOMY:

Transverse sections were cut from fresh leaf by using Reichert sliding microtome. Leaves from seedlings, relatively mature plants, flowering plants and old fruiting plants were all sampled and examined in each species. This was to ascertain whether any anatomical feature(s) or structure(s) were due to the leaf developmental variations.

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#### ii. Leaf Anatomy of Dry Specimens:

Leaf samples were taken from wide range of preserved specimens. The dry leaves were gently boiled and preserved for forty-eight hours in F.A.A. (made up with 10mls of 40% Formaldehye; 5mls of 95% ethyl alchol; 5mls of glacial Acetic acid and 35mls of distilled water (Ayensu, 1972). Part of each treated leaf was onhedded in paraffin wax and cut by using a Beck's Rotary Microtome. The transverse sections crumbled. The freezing microtome und even the Reichert sliding microtome were used, but there was no better sectioning.

Other pretreatments of the preserved leaves were tried. Some leaves were soaked in warm water for twenty-four hours before sectioning. Some leaves were only boiled, allowed to cool and sectioned without preservation. The sections cut crumbled. It is therefore not possible 'o present transverse sections from preserved specimens.

C. ROOT ANATOMY:

Transverse sections from fresh roots were tried on the freezing microtome as well as the Reichert sliding microtome but the sections erumbled. However, hand sectioning was done with sharp razor. Fairly thin transverse sections were cut in 70% ethyl alcohol; stained with alcoholic safranin, dehydrated in absolute ethyl alcohol and mounted in Canada Balsam for permanent preparation.

### D. PHOTOGRAPHY:

Leaf epidermal cells, transverse sections of leaves and root were photographed on the Carl Zeiss photonicroscope at X4 and X10 objectives. Kodak Panatomic -X film PX135 - 36 was used sequential record of each photographed specimen was kept as it was taken.

Films were developed and printed on Agfa 6N1 (5) papers. The films and pictures were then labelled and stored.

# RESULTS

After all examinations, it was found that anatomical structures were constant when the plants were at the flowering stages. This also formed a common basis of comparison.

The statistical analysis of the leaf epidermal data is presented in Table 7.

Plate III shows representative photographs of the epiderual layers while others are shown in the appendix 5.

The photographs shown in Plate: IV are the representatives of the leaf anatomy emphasizing the patterns of the epidernal cells at the leaf margin; while others are shown in the Appendix 6. Representative transverse sections of roots in both genora are presented in Plate V.

							NGI CHIBOURING CELLS								I VELIA ALPARATU I								0.0			
15 OF 3107474/m <sup>2</sup>	STORA IN N. CELL	I: N.				STRA US. STL		Tecal			WIDTS (microns)		(aisrons)		a cut stat		The second			(literons)	-		(storons)	1-17-2	5	ELECTED
1.2	LL I RATIO	3.BC	SELL 4 RATIO	R	PATIO	1 P. M.	RYCH	21122	NO. 083.	ZZAT	EVI02	NC. 023.	ATT A	EC LYEE	NC. 083.	NEW.	221722	TC. CB3.	NCV.	2.0 272	10. 255.	NEX	20.02	YO DES.		
65	10:13 -	(43.5)	10:12	(45.5)	10:12	(45.5)	1.4	1.0-1.34	250	92.63	82.5-102.5	250	135,821	100-160	250	1.22	1.12-1.38	250	79.25	65-82.5	250	96.5	90-100	350	C play	rophy) be
103	10:24	(29.4)	10:15	(40)	10:35	(22.2)	2.49	1.91-3.11	250	50.82	52.5-75	250	150_07	110-195	250	1-05	1.0 -1.13	250	40.63	37-5-42-5	250	42.15	40-45	250	Ç. macroj	4yllum
128	10:19	(34.5)	10:17	(37.0)	10:32	(23.8)	2.37	1.74-3.21	250	55.75.	47.5-72.5	250	- 1 30,28	115-192.5	250	1.24	1.15-1.33	250	32.88	30-35	250	40.75	37.5-45.0	250	C. inorna	itum
71	10:26	(27.8)	10:18	(38.7)	10:45	(18.2)	3.26	2.0-4.91	250	52.64	46.64.5	250	170.83	92.5-249	250	1-27	1.08-1.45	250	29,88	27.5-32.5	250	37.75	35-40	250	<u>C. 1400</u>	
100	10:24	(29.4)	10:19	(34.5)	- 10:47	(17.5)	2.7	2.11-3.19	250	64.93	60-70	250	176.88	137.5-215	250	1.12	1.0-1.25	250	34	30-37.5	250	打*82	35-40	250	<u>C. stere</u>	petalum I
56	10:21	(32.3)	10:22	(31.3)	10:45	(18.2)	2,3	1.82-3.0	250	78,88	67.5-87.5	250	179.63	145-225	250	1.12	1.03-1.21	250	35.82	35.0-37.5	250	40.25	37-5-45	250	G. stene	<u>p⊹talum</u> I
144	10:25	(28,6)	10:20	(33.3)	10:50	(16.7)	2+96	2.32-3.79	250	57-39	40-67.5	250	167	135-215	250	1.2	1.09-1.33	250	28.25	25-72.5	250	33.75	30-40	250	<u>c</u> . topo	ense.
202	10:9	(52.6)	10:19	(34.5)	10:17	(37.0)	1.1	1.0-1.21	250	86.75	80.0-92.5	250	101.5 .	100-120	250	1.25	1.0-1.45	250	57.38	47.5-65	250	71	65-82.5	250	<u>C. Alia</u>	aifolium
35	10:31	(24-4)	10:11	(47.6)	10:35	(22.2)	5.34	2.5 4.8	250	36.17	25-42.5	250	118.49	82.5-135	250	1.08	0.91-1-18	250	31.93	27-5-37-5	250	34 .3	30-39	250	Chlorop	nytum X
107	10:21	(32.5)	10:11	(47.6)	10:25	(30.3)	2.46	2.11-5.0	250	51.95	41.2-57.0	250	127.84	97-5-167-5	021	1.06	0.93-1.18	250	40.91	32.5-55	250	43-15	35-57.5	250	<u>c. ma</u> )	acena
173	10:54	(15.5)	10:9	(52.6)	10:47	(17.5)	6.8S	6.1-10.38	250	22.7	17.5-27:5	250	155.88	120-207-5	250	1+28	0.95-1.18 1.18-1.56	250	25.13	22.5-30	622	33.25	30-37.5	250	Δ. 100	1125
58	10:59	(14-5)	10:9	(52.6)	10:25	(15.4)	7.3	5.07-9.44	250	26.19	20 - 37.5	050	186-45	150-210	250	1.23	1.15-1-5	250	28.07	22.5-32.5	C52	34.13	27-5-37-5	250	A. Liter	cau <u>lon</u>
74	10:79 -	(11.2)	6.U.	(55.6)	10:65	(13.3)	9.43	7.89-12.45	250	34.03	18.5-37.5	250	321.73	184-401-5	250	1. 2	1.05-1.33	250	40.38	37-5-45	050	49.75	45-55	250	A. mahl	r <u>hachta</u>

LEAP BELDERVAL ANATONY IN SOME NIPERIAN SPECIES OF CELOSOPEVER AND AUDITATION.

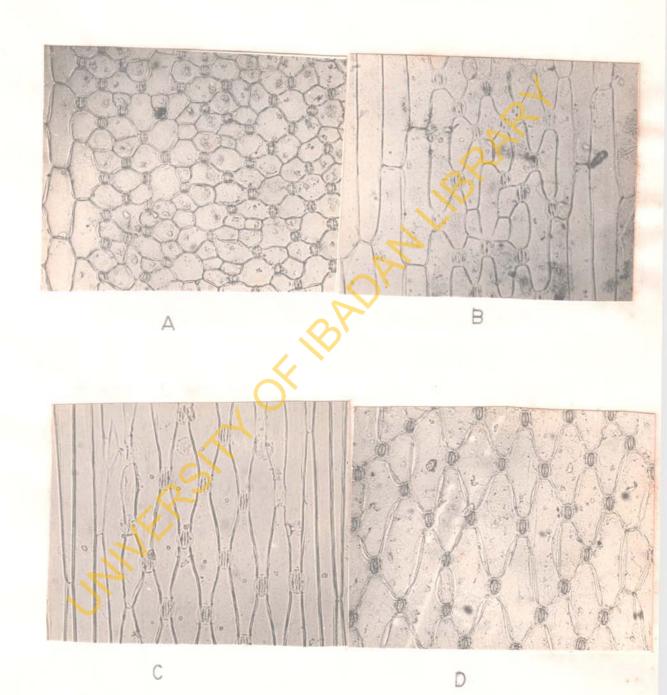
TABLE 7 - 65 - .

## PLATE III

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Photographs of leaf surface patterns in Anthericum and Chlorophytum complex.

- A. Primitive leaf surface pattern as in <u>C</u>. alignifolium.
  B. <u>Chlorophytum</u> X leaf surface pattern showing a primitive pattern.
- C. Typical leaf surface pattern in <u>Anthericum</u> species. (A. pubirhachis).
- D. Typical leaf surface pattern in <u>Chlerophy</u>tum species
   (C. stenopetalum I ).



X 200

### PLATE IV

fer.

Photographs of leaf margin sections; showing typical leaf margin anatomy in species of <u>Anthericum</u> and <u>Chlorophytum</u>: A & B show typical leaf margin anatomy in <u>Anthericum</u> species (A. <u>A. Limosum</u> B. <u>A. pubirhachis</u>)

C & D shows typical leaf margin anatomy in <u>Chlorophytum</u> species (C. <u>C. blepharophyllum</u> D. <u>C. stenopetalum</u> II)



# PLATE V

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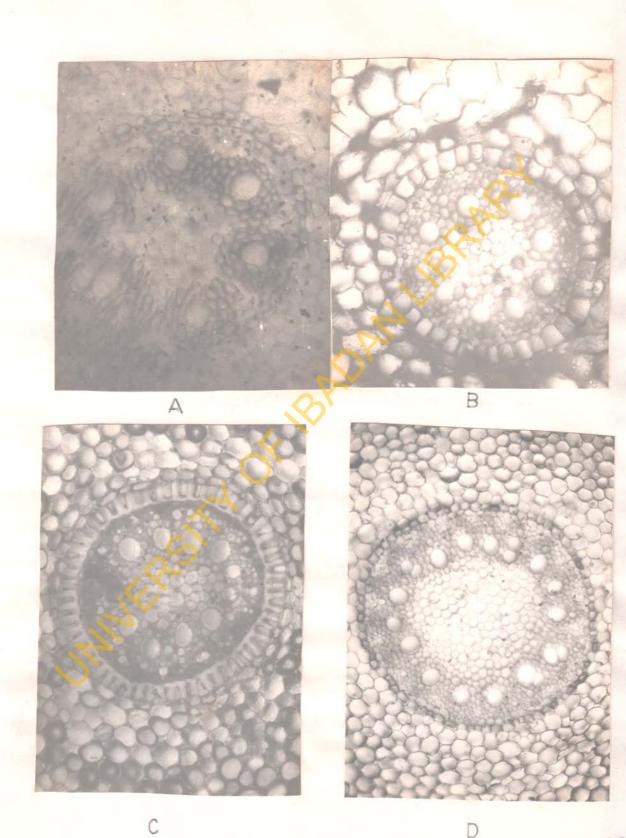
NERSIN OF BROMMUS Photographs of root sections:

Δ.

в.

C.

D.



### DISCUSSION

#### Leaf Epidermal Structures:

i. Number of stomata per square millimetre varies within the specimens of the same species. The stomatal size varies even in the same specimen depending on the position of the stomata in the leaf. These characters are not reliable for taxonomic purpose because they are influenced by environmental factors (Meidner and Mansfield, 1968; Kramer, 1969).

The characters of stomatal length, width and index are not found useful taxonomically as both taxa overlap in each of them. As can be seen in Table 7 the mean stomatal length ranges from 33.25 um to 49.75 um for <u>Anthericum</u>, while that of <u>Chlorophytum</u> ranges from 33.75 um to 96.5 um. The mean width ranges are brow 26.13 um to 40.9 um. (<u>Anthericum</u>) and from 29.88 um to 79.25 um. (<u>Chlorophytum</u>). Also, the index ranges from 1.06 to 1.28 for <u>Anthericum</u> species and from 1.05 to 1.24 for <u>Chlorophytum</u> species.

The stomatal differentiation is under genetic control. The similarrities of stomatal structure in this complex, seems to be an indication of the close genetical relationship along the component taxa. ii. The Neighbouring Cells:

The mean length of the neighbouring colls in this complex overlaps; however, the mean width and mean index divide the taxa in the complex into two. The mean width of the neighbouring cells in <u>Anthericum</u> species is from 22.7 µm to 34.0 µm. The range for <u>Chlorophytum</u> species is from 52.64 µm to 110.75 µm (excluding <u>Chlorophytum</u> X having 36.17 µm). Using the width of the neighbouring cells as a criterion, the complex can be divided into two broad groups: Group A, being made up of taxa having the width of their neighbouring cells less than 40µm while Group B plants have theirs above 40µm.

The index of the neighbouring cells as presented in Table 7, shows a range of 6.88 to 9.48 for <u>Anthericum</u> and 1.1 to 3.34 for <u>Chlorophytum</u>. Using index above 5.0 for taxa in Group A and from 5.0 and below for those in Group B, the grouping falls in line with the one suggested above. The only exception, however, is that <u>Chlorophytum</u> X is transferred from Group A to Group B. The double grouping of <u>Chlorophytum</u> X may probably be due to its possible evolutionary position in the complex.

## iii. The Ratio of Stomatal Apparatus: Neighbouring Cells:

The percentage of mean length of stomata : mean length of neighbouring cells, in Table 7, only shows continuity of the taxa in the complex. The percentage of width: width ratio as can be seen in column xxi of Table 7, separates the taxa into two groups. Group A plants are  $\geq 50\%$  (all <u>Anthericum</u> species)while

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that of Group B is  $\swarrow$  50% (all <u>Chlorophytum</u> species). Column xxii of Table 7 showing the percentage of index: index ratio also divides the taxa into two groups. The grouping correlates with the above grouping under width: width ratio. In this case, Group A has  $\lt$  25% while Group B has 7 25%.

Even though Atchley et. al (1976) stated, f... that not only have biologists been misled ... to what ratios actually show, but ratios greatly confuse and, in many cases invalidate critical statistical or biological analyses of the original data." However, the use of ratios in this work does not confuse or invalidate but rather complements and strengthens other biological analyses. It is interesting to note that the groupin; resulting from the use of ratios here totally agrees with and supports those of leaf index, vein spacing intervals, anther: filament ratio percentage and other morphological criteria.

The general surface pattern of the loaf epidernal cells supports the groupings based on the width and index of the neighbouring cells and those of ratios, as suggested above. (See Plate III). In addition, while the shapes of the neighbouring cells in the Group A taxa are elongated parallelograms, those of Group B taxa are roughly rhombic in shape.

The importance of the leaf surface anatomy in solving taxonokic problems is now gaining more credibility because it is known to be genetically controlled. Cutler (1972), working on leaf surface patterns of Aloe, Gasteria and Haworthia, concluded that the various leaf surface patterns are under genetic control and that environment plays very little, if any, part in determining the appearence of the leaf surface of members of the tribe Aloineae, to which his materials belong. The genetical control of the leaf surface pattern was further confirmed by the work of Cutler and Brandham (1977) by showing that the hybrid leaf surface anatomy was intermediate between the two parents, as evidenced in the cross Aloe rauhii X Aloe dawei. It may be suggested at this point that the leaf surface anatomy of C. alismifolium and Chlorophytum X probably are the most primitive patterns.

## Leaf Anatomy:

There are intermediates in the vascular bundles of the plants in this complex. Hence no clear-cut division can be formed on these structures. A look at the anatomy of the leaf margin of the plants in this complex as presented in Plate IV, shows two clear-cut divisions. <u>A. limosum, A.</u> <u>pterocaulon, A. pubirhachis, A. uyuiense, Chlorophytum X</u> and <u>C. alismifolium</u> form Group A. Their epidermal cells surround the mesophyll cells at the leaf margin. Taxa in

- 70 -

#### their

Group B have/leaf margin formed by tapering and reduction in number of epidermal cell layers, terminating in one or two epidermal cells. All the <u>Chlorophytum</u> species except <u>C. alismifolium</u> and <u>Chlorophytum</u> X form this group B.

Herat and Theobald (1977) employed vegetative anatomy and morphology in resolving the tribe and genera complexity in Gesneriaceae. Karlstrom (1978) employed observations on differences of epidermal leaf structures in the species of Strobilantheae and Petalidineae (Acanthaceae) to justify a separate subtribe Petalidinae. In the same way, Pant and Basu (1977) employed leaf anatomy to confirm the separation of Keteeria and Cathoya genera. In view of the above facts given about taxa in Group A and B, it is hereby suggested that Anthericum and Chlorophytum should remain as two separate genera of Liliaceae. Group A taxa should thus be regarded as Anthericum species while Group B taxa are Chlorophytum spcies. The position of C. alismifolium, C. laxum and Chlorophytum X will be further examined later along with other criteria in Chapter 7

Root Anatomy:

The root steles do not give clear and useful information as regards the separation of the two genera. They are structurally similar. The identical anatomical features in roots of <u>Anthericum</u> and <u>Chlorophytum</u> reported by Deshpande (1955a) and Coetzee and Schijff (1969) is hereby supported (see Plate V).

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The anatomical studies reveal similarities in stomatal apparatus, root steles and overlappings of other characters. This suggests that the two genera have a common ancestor or one common parent whose characters they both retain and perpetuate. The leaf epidermal structures of Calismifolium and Chlorophytum X suggest that taxa in this complex might have arisen by natural hybridization. The epidermal anatomy structures in other species seem to be intermediate between those of C. alismifolium and Chlorophytum X. For example, the leaf epidermal surface pattern in C. blepharophyllum is intermediate between those of C. Alismifolium and Chlorophytum X with C. alismifolium possibly being the maternal plant. It is my belief that the evolutonary line in this complex diverged after the initial parental crosses to give rise to the present separate genera. Thus the two genera might have followed parallel lines of evolution. Their similarities thus confirm that they both retain most of their ancestrial characters.

#### CHAPTER 5

#### CYTOLOGICAL STUDIES

The aims of the cytological studies were to establish the basic chromosome number, confirm or reject chromosome numbers reported for some species and report new chromosome numbers (if found) for some Nigerian species of the complex. It was also aimed to study the Karyomorphology, mitotic and meiotic chromosome behaviours with a view to helping incritical examination of the possible relationship and mode of evolution of the Karyotypes in the genera.

#### A MATERIALS AND METHODS

Mitotic studies were carried out on all the species investigated in this work, while meastic studies were carried out on all but <u>C. geophilum, A. nubicum</u> and <u>A. pterocaulon</u> I.

#### MITOTIC STUDIES

Young root tips were used for the mitotic studies. Samples of root tips were taken from ten individual plants of each species except in <u>A. mubicum</u> where samples were taken from the only two individual plants available.

#### (i) Collection of Material for Karyotype study

Root tips collected, pre-treated, fixed and preserved in the field during the 1979 field trips as well as from cultivated samples were investigated.

Collected specimens were planted in plastic pots and on experimental beds in the Botany Department, University of Ibadan. They were subjected to uniform experimental conditions. It was found difficult to get good root tips from the specimens cultivated on experimental beds, hence root tips were collected from plants in plastic pots. The root tips were harvested by gently rocking the plastic pots and pouring out the soil and the plants. The roots were then carefully washed free of soil particles in running tap water, immediately cut and put in pre-treatment drug. Collection of root tips was done at different times to ascertain the best time for active cell division. Mitotic cell division was found most active between 6.30 a.m. and 8.00 a.m. for specimens of both taxa.

## (ii) Pre-treatment of Root Tips

Root tips (1 cm to 2 cm. in length) were removed into specimen tubes containing saturated aqueous solution of p-dichlorobenzene or other pre-treatment drugs. They were kept for three hours in the dark. When kept for a lesser period, the chromosomes were long and thin while at three hours, they were short, easily spread out and definitive in structure. The dark period treatment was found to be necessary for normal growth.

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The roots did not show evidence of normal growth when left in the light. Apart from aqueous solution of p-dichlorobenzene, other pre-treatment drugs like C-monobromonaphthalene, 1% colchicine and 8-hydroxyquinoline were also tried. Even though all the pre-treatment drugs gave some measure of success, p-dichlorobenzene gave a more satisfactory result and was easy to prepare. Hence it was used for this work so as to give a common basis for comparison.

(iii) Fixation and Preservation

After three hours, specimen tubes were taken out from the dark. The pre-treated roots were then washed in running tap water for two minutes so as to wash off the pre-treatment drug. Carnoy's solution was prepared fresh (Darlington & La Cour, 1962), mixed just a few seconds before the roots were kept in the solution. The tubes were labelled and put in the deep freeze of the refrigerator for at least twenty-four hours. The fixation was necessary in order to kill the cells and arrest the nuclear activities while storage in the refrigerator encouraged proper staining of the chromosomes.

## (iv) Hydrolysis

The roots were taken out from the preserving solution and washed thoroughly for two minutes in running tap water. Hydrolysis was necessary to soften the root tips which aids in spreading out of the cells and their chromosomes. Two methods of hydrolysis were tried which were (a) simultaneous hydrolysis and staining and (b) hydrolysis at room temperature by using low concentration of HC1.

(a) Simultaneous hydrolysis and staining method

This method formally referred to as Acetic Orcein stain technique (Darlington and La Cour, 1962) was modified by Marenah and Holden (1967). Root tips were put in a mixture of nine parts of 2% acetic orcein and one part of IN HCI (by volume) and kept in an oven at 60°C for 8 - 10 minutes. Squashing followed in a drop of 2% acetic orcein on a clean slide.

(b) Hydrolysis at Room Temperature

Oyewole (1975) used 37.5% HCI for hydrolysing root tips at room temperature while Olorode (1973b) used 15% HCl for his own hydrolysis. In this study, 35% HCl was used as hydrolysing solution at room temperature. The period of hydrolysis was between 6 and 10 minutes depending on the thickness or size of the root. Hydrolysed roots were washed for two minutes in running tap water.

This method saved time and it worked for the material under investigation. The root caps readily fell off on their own during washing. Another point for this method was that it did not depend on electric power supply, which was usually irregular as hydrolysis was at room temperature.

### (v) Squashing

Washed and hydrolysed root tip (i.e. the meristematic part) was squashed in a drop of stain on a clean slide. It was gently covered with a coverslip smeared with a tiny speck of glycerine albumen and dried over a flame of the spirit lamp. The cells readily spread out. The coverslip was tapped gently with pointed needle to allow for even spreading of the cells and their chromosomes. The slide was then examined under the microscope to find out whether the cells and their chromosomes were spread enough. When the chromosomes were not spread enough, further gentle tapping of the slide was done. Stains used for this work were 1% Aceto orcein (Marenah and Holden, 1967) and 2% Acetocarmine solutions. Slides meant for further studies were sealed up at the edges with nail varnish and kept in the refrigerator. Slides

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with this treatment usually lasted for five to eight days without being spoilt. Moreover, the chromosomes stained better under this condition.

#### (vi) Permanent Preparations

Good preparations were made into permanent slides as follows: Two or three corners of the coverslip were marked with a diamond pencil. Slide was placed in 45% acetic acid in a slide tray with the coverslip downwards. After the coverslip had fallen off, the slide was transferred into 1:1 acetic acid: butanol.(by volume) for 10 to 15 seconds. The slide was then removed and dehydrated in absolute butanol for two minutes. The slide and coverslip were placed on clean filter paper with their faces upwards to allow them dry. A drop of Canada Balsam in xylene (a mountant) was put on the dry coverslip. The slide was then placed properly, setting the marking on the coverslip and then pressed slightly. It was then turned face up and allowed to dry. The slides were labelled and kept in plastic slide trays for future use.

## (vii) Chromosome counts:

Chromosomes were counted in at least ten representatives of each species (except in <u>A. nubicum</u>, <u>C. geophilum</u> and <u>A. pterocaulon</u> I).

In each representative, chromosomes in 10-20 intact cells, were counted, thus chromosomes in 100-200 cells were counted for each species. The metaphase chromosomes were counted at x10, and x40 or x100 (with oil immersion) objectives.

(viii) Karyotype

Karyomorphology of each species was studied, taking note of the centromeric positions, the shape of the chromosomes, presence or absence of secondary constrictions, and satellites. Absolute length and r-value of each chromosome were also determined.

- (a) <u>Measurements of chromosome size</u>
  - The measurement of each chromosome was made at x100 objective (with oil immersion) of a light microscope in which a unit of the micrometer eyepiece graticle equalled one micron (um). The long arm (l.), the short arm (s) and the absolute length (c) of each chromosome were recorded. The position of centromere was determined as the position of a major construction between the two arms of each chromosome. Due to the different positions of cells in a root tissue and different stages of arrest of the mitotic stages even at the metaphase, it became necessary to standardise the thickness of the chromosomes to about two microns (-2um). Using this thickness approximation for each measured chromosome, the

variation of homologous chromosomes in different cells of the same species, in different representatives was negligible. However, an average was calculated for each homologous pair of chromosomes (approximated to the nearest 0.5 µm) in at least 20 chromosome complements.

(b) The r-value (1) was calculated for each homologous pair of chromosomes for the determination of the centromeric position. The chromosome morphology was analysed by the method of Levan, et al (1964) as follows:

Position of centromere	r-value	symbol
Terminal point	12.1 - 00	Т
Terminal region	7.1 - 12.0	t
Subterminal region	3.1 - 7.0	st.
Submedian region	1.71- 3.0	sm.
Median region	1.1 - 1.7	m.
Median point	1.0	M

The chromosomes were further classified as long when they were above 6.0 µm Chromosomes having their total length from 6.0 µm - 4.0 µm were regarded as medium in size while those less than 4.0 µm were referred to as short chromosomes.

## (ix) Photography

Well spread chromosomes of intact cells at metaphase stage were photographed immediately with the Carl Zeiss photomicroscope at x40 objective. Kodak Panatomic -x Px 32 ASA/17 Din 35 mm.films were used. The best time of exposure was found to be between 5 seconds and 10 seconds depending on the light intensity and how deeply the chromosomes were stained. It was found that preparations left overnight gave better pictures at No IV on the light control board of the photomicroscope for 8 seconds exposure. The films were developed in D196 developer. Printing was done on Ernst Leitz GMBH Wetzlar Focomat 11<sup>°</sup> Enlarger. The films and photographs were labelled and stored.

(x) Idiograms

B.

The mean chromosome lengths were used for drawing the idiograms.

Meiotic atudies were made so as to confirm the chromosome numbers established by mitotic studies and to investigate the homology and pairing patterns of the chromosomes. These studies could be found useful in elucidating the evolution of the chromosome complements in the complex. An attempt was also maded to assess the possible incidence of chromosomal irregularities; inversions, translocations, duplications and/or deletions in the complex.

## (i) Choice of material

Anthers of young flower buds were used because they could be ohtained easily. The abundant pollen mother cells (PMC.) in the six anthers found in each flower made the anthers better material for meiotic studies than the ovules which were difficult to reach and relatively very few in number. Good meiotic stages were usually found in young flower buds when they were still green. However, random trials of different stages of flower buds usually gave good result, since the meiotic activity depends on species and time of collection.

## (ii) Collection of material

(iii)

Flower buds were collected from at least ten representatives for each species. Collections were made throughout the day. However, it was found that materials collected between 8.00 a.m. and 12.00 noon usually gave good meiotic division stages. Specimens collected in the afternoon and evening usually had clumped chromosomes. Fixation and Preservation

Flower buds of different stages of development were harvested and fixed immediately in freshly prepared Carnoy solution and preserved in a refrigerator for at least twenty-four hours, This is necessary, so as to allow for proper fixing of the cells and the removal of mucilage from the anthers.

#### (iv) Preparation of Meiotic Materials.

Fixed and preserved flower buds were washed in running tap water. Flower buds were hydrolysed in 10% HCL for 5 minutes so that the pollen mother cells (PMC) would be softened to allow for easy spreading of the chromosomes. The flower bud was washed in running tap w.ter. The anthers were picked out from the flower bud with clean pins and forceps. Two or three anthers from each flower bud were squashed in a drop of 2% aceto-carmine on clean slide and covered with a clean coverslip. The preparation was slightly tapped and scanned under the microscope. Further tapping and irrigation (with staining sol".) of preparations were done when found necessary. The process of permanent preparation given under mitosis was followed.

(v) Studies of Meiotic Behaviours

Each meiotic preparation was exhaustively scanned under a light microscope at x40 objective. The behaviour of the chromosomes at the prophase stages; metaphases I and II, anaphases I and II and telophases I and II were carefully studied. The prophase I stages atudied were Pachytene, Zygotene, Diplotene and Diakinesis. These atages were studied to find out the pattern of pairing, whether any of the apparent forms were results of breakage and/or reunion of non-sister chromatids. The metaphase I chromosomes were counted so as to confirm the chromosome numbers established by mitotic studies. Metaphase II was examined to find out any unequal separation that might have occurred during the first anaphase. Anaphases I and II were studied to find out whether any of the sister chromatids might be forming laggards, non-disjunctions and/or dicentric bridges. Early telophases I and II were investigated to note the probable 'left out' chromosomes outside the daughter nuclei. The late telophases I and II were also examined to investigate the presence of any 'micro-nuclei', number of daughter nuclei and their separation.

## (vi) Photography

Well spread diplotene, diakinesis or metaphase I chromosomes with definite cell walls were photographed as detailed for mitotic study.

## (vii) <u>Magnification</u>

Marked specific chromosomes and cells were measured at x100 objective of a light microscope in which one unit of micrometer graticle equalled one micron (um). The measurement was recorded in microns. The cells and/or specific chromosomes were photographed on Carl Zeiss Phase Contrast photomicroscope. The film was developed and printed at different elevations of the Focomat 11<sup>°</sup> Enlarger. Measurements from photogrphs of the cells and specifically marked chromosomes were taken and recorded in millimetres (mm.). The magnification was calculated by dividing the photographic measurement by the actual measurement thus:

Magnification =  $\frac{\text{Photographic measurement}}{\text{Actual measurement}} \times \frac{1000*}{1}$ 

\*(1000 microns make 1 millimetre)

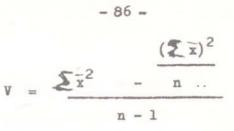
The magnification of any cell or chromosome photographed at x40 objective of the Carl Zeiss Phase Contrast photomicroscope, developed and printed at 11th level of elevation on the Ernst Leitz GMBH Wetzlar Focomat 11° Enlarger was found to be x1000. Thus all the mitotic and meiotic cells were photographed at x40 objective of Carl Zeiss photomicroscope and printed at the 11th level of elevation of the Focomat 11° Enlarger.

### C STATISTICAL ANALYSES OF THE KARYOTYPES

The chromosome measurements were subjected to some simple statistical analyses.

(i) The variance

The variance of each chromosome complement and the mean total length of chromsome for each species were calculated by using the formula:



where  $\sum_{\mathbf{x}}$  means sum  $\mathbf{x}$  " the value of variables. n " the number of variables.

2. Mean Variance was determined as

$$\overline{\mathbf{v}} = \frac{1}{n} \begin{bmatrix} \overline{\mathbf{x}}^2 - (\overline{\mathbf{x}})^2 \\ \frac{n}{n-1} \end{bmatrix}$$

## 3. Standard Deviation

This was calculated by employing the formula:

$$S = \frac{\left(\sum_{x}\right)^{2}}{n}$$

$$n - 1$$

Student t test

Popham and Sintnils's (1973) separate variance t model formula was used for calculating the t value.

i.e. 
$$\bar{x}_1 - \bar{x}_2$$
  
 $t = \sqrt{\frac{S_1^2}{n_1} + \frac{S_2^2}{n_2}}$   
where  $\bar{x}$  is the mean of the variables  
 $S^2$  is the variance found by squaring the standard deviation)  
n is the number of variables.

### Homogeneity of Variances. F-test

To test the hypothesis that  $\sigma_1^2 = \sigma_2^2$ , a simple statistical test, the F ratio, according to Popham and Sirotnik (1973), was employed.

$$r = \frac{s_g^2}{s_s^2}$$

where

F = the value by which variance homegeneity
will be tested

 $S_g^2$  = the greater (larger) simple variance

 $S_1^2$  = the lesser (smaller) simple variance (degree of freedom is n - 1 for each of the variances, that is, two values were obtained).

### REGULTS

A MITOTIC STUDIES

The mitotic divisions were normal in almost all the species of both genera. However, endomitosis was observed in few cells of  $\underline{C}$ . stenopetalum I and in many cells of  $\underline{A}$ . pubirhachis.

#### Karyotype Measurements

Chromosome measurements are presented in Tables8 and 9. The haploid chromosome set is presented for diploid and tetraploid

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species. The somatic chromosome numbers are also presented. Summary of similar chromosome sets in the complex is presented in Table 10. They are grouped according to their r - values and total lengths. Table 11 shows the statistical analysis of the C. stenopelalum complex.

#### Idiograms

Idiograms constructed with the mean chromosome lengths are presented in Figure III for <u>Chlorophytum</u> and <u>Anthericum</u> species. <u>Photographs</u>

Somatic chromosomes are presented in Plates VI and VII for <u>Chlorophytum</u> and <u>Anthericum</u> species respectively.

#### MEIOTIC STUDIES

Except in <u>Chlorophytum X, C. alismifolium</u> and few cells of <u>C. caulescens</u> and <u>A. pterocaulon</u> II, meiotic division and chromosomal behaviours were normal.

The frequency of chiasma formation at diakinesis is presented in Table 12. Type of chiasma formation in <u>C. stenopetalum</u> complex is presented in Table 13.

#### Photographs:

Photographs of meiotic cells are presented in Plates VIII and IX. Irregularities in meiotic divisions and chromosomal behaviours at meiosis are presented in Plate X.

B

KARYOMORPHOLOGY OF TEN SPECIES OF CHLOROPHYTUM

SI	PECIES		I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	MEAN JJM
<u>C</u> .	blepharo-	C	10	9.5	8	8	8	7	7	7	6.5	6	6	5	5	5	
	ph:llum	1	6	7	6	5	4	5	4	4	4	3	5	3	2.5	4	7.0
		s	4	2.5	2	3	4	2	3	3	2.5	3	1	2	2.5	1	
		r-v	1.5	2.8	3.0	1.7	1.0	2.5	1.3	1.3	1.8	1.0	5.0	1.5	1.0	4.0	
3M	4m 5sm 2st	loc	m	sm	Sm	<u>Iu</u>	M	sm	m	70	SIL	M	st	m	M	st	
<u>C</u> .	geophilum	C	0.5	9	9	8.5	8	8	7.5	7	7	7	7	6.5	6	3	7.43
		1	6	6	5	6	5	4	5	4.5	4	4	4.5	4	3	1.5	
		s	4.5	3	4	2.5	3	4	2.5	2.5	3	3	2.5	2.5	3	1.5	
		r-v	1.3	2.0	1.3	2.4	1.7	1.0	2.0	1.8	1.3	1.3	1.8	1.6	1.0	1.0	
3M	5m 6sm -st	loc	m	sm	m	sm	m	M	sn	sm	m	m	sm	m	M	M	
<u>c</u> .	macrophy-	C	10	9	9	8	8	7	7	7	7	6.5	6	6	5.5	4	7.14
	llum	1	6	7	5	6	5	6	4	4	5	5	4	3	3	2	
		s	4	2	4	2	3	1	3	3	2	1.5	2	3	2.5	2	
		r-v	1.5	3.5	1.3	3.0	1.7	6.0	1.3	1.3	2.5	3.3	2.0	1.0	1.2	1.0	
2M	5m 4sm 38t	loc	m	st	m	sm	m	st	m	m	sm	st	sm	M	m	M	
<u>C</u> .	inornatum	C	11	9.5	9	9	9	8	7						-		8.93
		ı	7	7	7	5	5	4	5								
		s	4	2.5	2	4	4	4	2								-
		r-v	1.8	2.8	3.5	1.3	1.3	1.0	2.5								
1M	2m 3sm 1st	loc	sm	sm	st	m	m	M	sm								

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TABLE 8 (CONTD.)

	_	I	п	III	IV	V	VI	VII	MEAN
C. inornatum	C	11	9.5	9	9	9	8	7	8.93
	1	7	7	7	5	5	4	5	
	s	4	2.5	2	4	4	4	2	
	r-v	1.8	2.8	3.5	1.3	1.3	1.0	2.5	
1M 2m 3sm 1st	loc	sm	sm	st	m	m	M	sm	
C. laxum	đ	11	9	8	7	7	6	6	7.71
	1	7	9	5	5.5	4	4	3	
	S	4	4	3	1.5	3	2	3	
	r-v	1.8	1.3	1.7	3.7	1.3	2.0	1.0	
1M 3m 2sm 1st	loc	sm	m	m	st .	m	sm	M	
C. stemopetalum I	C	8	8	8	. 7	7	6	6	7.14
	1	6	5	4	5	4	5	3	
	gs	2	3	4	2	3	1	3	
	r-v	3.0	1.7	1.0	2.5	1.3	5.0	1.0	
2M 2m 2sm 1st	loc	sm	m	м	sm	'n	st	M	1
C. stenopetalum II	C	10	9	9	8.5	7.5	6	6	8.0
	1	7	5	4.5	5	5.5	5	3	1
	s	3	4	4.5	3.5	2	1	3	
	r-v	2.3	1.3	1.0	1.4	2.8	5.0	1.0	
2M 2m 3sm -st	loc	sm	m	M	m	sm	st	M	

		I	II	III	IV	V	VI	VII	VIII	MEAN
C. togoense	C	9	8	8	8	8	7	5		7.57
(2n = 14)	1	5	6	6	5	4	5	2.5		
	s	4	2	2	3	4	2	2.5		
	r-v	1.3	2.0	3.0	1.7	1.0	2.5	1.0		
2M 1m 4sm -st	loc	m	sm	sm	sm	M	sm	M		
C. alismifolium	C	8	7	7	6.5	6	6.0	5.5	5	6.38
(2n = 16)	1	5	5	4	4	5	3	3	3	
	s	3	2	3	2.5	1	3	2.5	2	
	r-v	1.7	2.5	1.3	1.6	5.0	1.0	1.2	1.5	
1M 4m 2sm 1st	loc	sm	sm	'n	m	st	M	m	m	
Chlorophytum X	C	8	8	8	7	7	6	6	5	6.88
(2n = 16)	1	5	5	4	6	4	4	4	2.5	
	S	3	3	4	1	3	2	2	2,5	
	r-v	1.7	1.7	1.0	6.0	1.3	2.0	2.0	1.0	
2M 1m 4sm 1st	loc	sm	sm	M	st	m	sm	sm	M	
C. caulescens	C	10	10	10	9	7	7	7	6	8.25
(2n = 16)	1	8	7	6	6	5	5	4	3	
	s	2	3	4	3	2	2	3	3	
	r-v	4.0	2.3	1.5	2.0	2.5	2.5	1.3	1.0	
1M 2m 4sm 1st	loc	st	sm	m	sm	sm	sm	m	M	

	KERYOMORPHOLOGY	IN SIX	SPECIES	OF	ANTHERICUM
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		I	П	III	IV	v	VI	VII	VIII	MEAN
A. limosum	C	8	8	7	7	7	6	6	4	6.63
(2n = 16)	1	6	. 5	6	5	4	4	3	3	
(211 = 10)	s	2	3	1	2	3	2	3	1	
	r-v	3.0	1.7	6.0	2.5	1.3	2.0	1.0	3.0	
1M 1m 5sm 1st	loc	sm	sm	st	SM	m	SM	M	sm	
A. nubicum	C	8	8	7	T	7	6.5	5.	4	6.56
(2n = 16)	1	6	5	6	5	4	4.5	2.5	3	
(	s	2	3	1	2	3	2	2.5	1	
	r-v	3.0	1.7	6.0	2.5	1.3	2.3	1.0	3.0	
1M 2m 4sm 1st	100	sm	m	st	sm	m	SM	М	sm	
A. pterocaulon I	C	9	8	7	6	6	5	5	5	6.38
(2n = 24)	1	5	5	5	5	4	4	3	2.5	
	S	4	3	. 2	1	2	Ť	2	2:5	
	r-v	1.3	1.7	2.5	5.0	2.0	4.0	1.5	1.0	
11 3m 28m 28t	loc	m	m	sm	st	sm	st	m	M	
A. pterocaulon II	C	8	7	7	6	6	6	5	5	6.25
(2n = 16)	1	5	5	4	4	3	3.5	3	4	
(211 - 10)	s	3	2	3	2	3	2.5	2	1	
	r-v	1.7	2.5	1.3	2.0	1.0	1.4	1.5	4.0	
1M 4m 2sm 1st	loc	m	sm	m	SIA	M	ш	m	st	
A. pubirhachis	C	7	6	5.5	5	5	4.5	4	4	5.13
	1	5	4	4	3	2.5	2.5	3	3	
	s	2	2	1.5	2	2.5	2	1	1	
	r-v	2.5	2.0	2.7	1.5	1.0	1.7	3.0	3.0	
IM 2m 5sm -st	loc	500	sm	sm	m	M	m	sm	sm	

#### SUMMARY OF SIMILAR CHROMOSOME SETS IN THE ANTHERICUM - CHLOROPHYTUM COMPLEX

	-						·		•			
		A			В				C			D
	C	8 6 5		. 7	7 10 5	8	6.5		7	9.5 8		5 6
C. blepharophyllum	r-v	1.0 1.0 1.0		1.3 1	.3 1.5 1.5	1.7	1.8		2.5	2.8 3.0		4.0 5.0
	100	M M M		m	n m m	m	sm		sm	sm sm		st st
	C	8 6 3	10.	5 9 7	7.	5.5 8	7 7	7.5 9	8.5			
C. geophilum	r-v	1.0 1.0 1.0	1.	3 1.3 1.3 1	3	1.6 1.7	1.8.1.	8 2.0 2.0	2.4			
	Loc		······································		n	m m	sm sm	sm sm	Sm			
C. macrophyllum	C		5.5	9 7		. 8		6	7	8	6.5 9	
Mill Million Striker of the Striker of Strikerow of Striker of Striker of Striker of Striker of Striker of Str	r-v Loc	1.0 1.0 M M	1.2° m	1.31.31 m m		. 1.7 m		2.0 sm	2.5 sm	3.0 sm	3.3 3.5 st st	
					ц ш-	ш		300	•			
. inornatum	6	8	-	°9			11		, 7	9.5	9	
	r-v Loc			.31.3 m			1.8 sm		2.5	2.8 sm	3.5 st	
											. 7	
C. lazum	C	6 1.0		9 7-		8	11	6			7.3.1	
	r-v Loc	M		1.31.3 m m		1.7 m	sm	2.0 sm			J. st	
	C	8 6		7		8		Sm	7	8		6
<u>C. stenopetalum</u> I	r-v Loc	1.0 1.0 M M		1.3 m		1.7 m			2.5 sm	3.0 sm		5.0 st
<u>C. stenopetalum</u> II	C r-v Loc			9 1.3 m	10 1.5 m	$\odot$				7.5 2.8 sm	8.5 3.5 st	6 5.0 st
C. topoense	C r-v Loc	8 5 1.0 1.0 M M		9 1.3 m		8 1.7 m			7 2.5 sm	8 8 3.0 3.0 sm sm		
<u>C. alismifolium</u>	C r-v Loc	6 1.0 M	5.5 1.2 m	7 1.3 m	1.5	5.58 1,61.7 mm,			7 2.5 sm			6 5.0 st
Chlorothytum X		1.0 1.0		7 1.3	-	8 8	7	6 6 ·2.0 2.0				
	Loc C.	M M		7	10	mm		sm sm 6 7	7	7	8	
C. <u>caulescens</u>	r-v Loc			1.3 m	1.5 m		Ti.	2.0 2. sm sm	3 2.5 2	5	4. st	0
A. limosum	C r-v	6		7		8 1.7		6 2.0	7 2.5	8 4 3.0 3.0		
	Loc	M		m		m	1.1	sm	sm	sm sm		
A. nubicum	r-v Loc			7 1.3 m		8 1.7 m	1	5. 2. sm	3 2.5	8 4 3.0 3.0 sm sm		
A. pterocaulon I	C r-v	5		9	5 1.5	8		6 2.0	- 2.5			5 6 4.0 5.0
	Loc	.M.		m	m	m		sm	Sm			st st
<u>A. pterocaulon</u> II	C r-V Loc			7 1.3 m	1.5	6 8 1.4 1.7 m m		6 2.0 sm	7 2.5 sm			5 4.0 st
A. pubirhachis	C r-v	5			5 1.5	4.5		6 2.0	7 2.5	5.5 4 4 2.7 3.0 3.0	1.1	
	Loc		1		m	m	1 1 1	sm	sm	sm sm sm		

STATISTICAL ANALYSES OF CHROMOSOME COMPLEMENTS OF TWO FORMS OF C. STENOPETALUM

CHARACTER	FORM	I	II	III	IV	V	VI	VII	MEAN	VARIANCE		CALCU- LATED F-RATIO VALVE	PROBABILI
TOTAL LENGTH OF	I	8	8	8	7	7	6	6	7.143	0.810	.1.262	-2,984	>5%
CHROMOSOME	II	10	9	9	8,5	7.5	6	6	8.0	2.417			
r-VALUE	I	3.0	1.7	1.0	2.5	1.3	5.0	1.0	2.214	2.085	0.872	4.335	> 5%
OF CHROMOSOME	II	2.3	1.3	1.0	1.4	2.8	2.0	1.0	1.688	0.481			

CRITICAL t - VALUE 1.782 TABLED VALUES OF F 4.38 8.47

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# CHIASMATA FREQUENCY IN THE POLLEN MOTHER CELLS OF SOME NIGERIAN SPECIES OF CHLOROPHYTUM AND ANTHERICUM

SPECIES	n value	NO OBS.	TOTAL CHIASMATA	MEAN CHIASMATA/ P.M.C.	MEAN CHLASMATA/ BIVALENT
C. blepharophyllum	14	10	213	21.3	1.52
C. macrophyllum	14	7	166	23.71	1.69
<u>C. inornatum</u>	7	15	204	13.6	1.94
<u>C. laxum</u>	7	10	37	13.7	1.96
<u>C. stenopetalum</u> I	7	10	1 38	13.8	1.97
C. stenopetalum II	7	10	141	14.1	2.01
C. togoense	7	8	110	13.75	1.96
C. alismifolium	8	20	303	15.15	1.89
<u>C. caulescens</u>	8	10	161	16.1	2.01
<u>A. limosum</u>	8	10	158	15.8	1.98
A. pterocaulon II	8	10	159	15.9	1.99
A. pubirhachis	8	10	154	15.4	1.93

TABLE . 13

# CHIASMA FORMATION AND TYPES IN TWO FORMS OF C. STENOPETALUM

TAXON		I	II	III	VI	V	VI	VII
C. stenopetalum I	USUAL NO. OF CHIAMATA	2	2	2	2	2	2	2
	TYPE	Т	sub	. Т	с <b>П</b> .	T	T.	T
	111 12	J	sub	. I	sub.	I	sub.	I
C. stenopetalum II	USUAL NO. OF CHIAMATA	2	2	2	2	2	2	2
	TYPE	sub.	Т	T	T	I	т	Т
		I	T	I	T	I	T	T

N.B. Т. means Terminal chiasma

sub. \*\*

I.

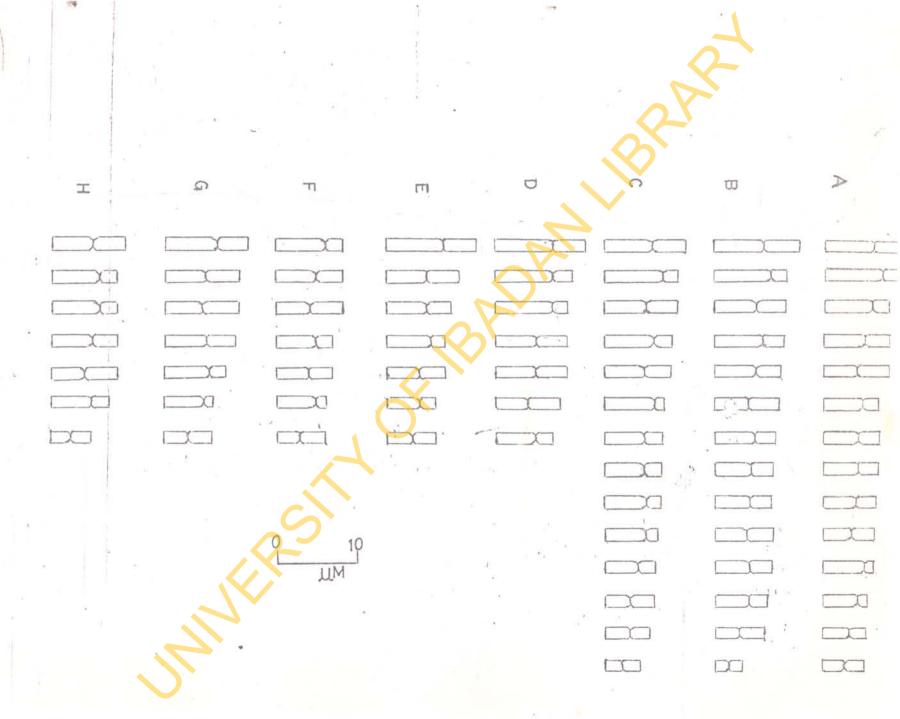
Subterminal chiasma

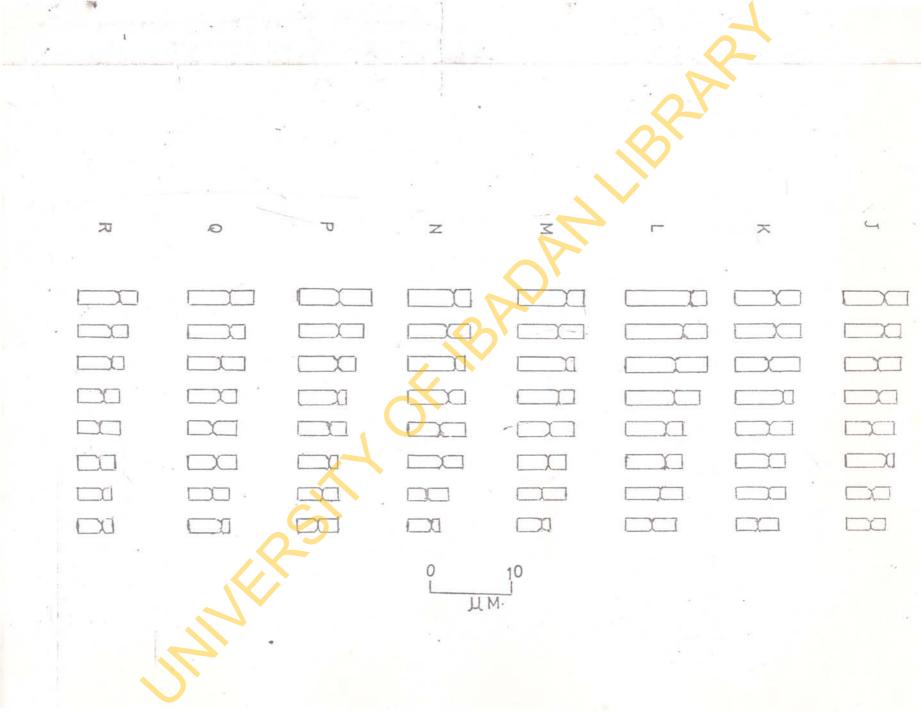
11 Interstitial or median chiasma.

# FIGURE III

Idiograms of some species of <u>Chlorophytum</u> and <u>Anthericum</u> in Nigeria:

Α.	C. blepharophyllum
Β.	C. geophilum
C.	C. macrophyllum
D.	<u>C. inornatum</u>
E.	C. laxum
F.	<u>C. stenopetalum</u> I
G.	<u>C. stenopetalum</u> II
H.	<u>C.</u> togoense
J.	<u>C. alismifolium</u>
ĸ.	Chlorophytum X
L.	C. caulescens (A. caulescens)
M	A. limosum
N.	A. nubicum
P.	A. pterocaulon I
Q.	A. pterocaulon II
R.	A. pubirhachis



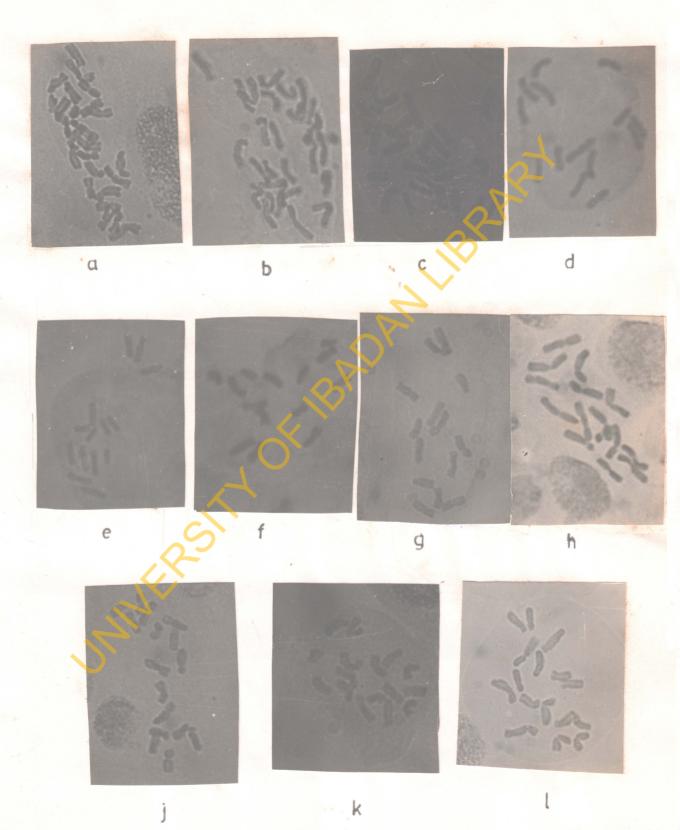


# PLATE VI

Photographs showing metaphase somatic chromosomes in some species of <u>Chlorophytum</u>:

8.	<u>c</u> .	blepharophyllum	(2n = 28)	
Ъ	<u>c</u> .	geophilum	(2n = 28)	
c	<u>C</u> .	marcophytum	(2n = 28)	
d	<u>C</u> .	inornatum	(2n = 14)	
е	<u>C</u> .	lazum	(2n = 14)	
f	<u>c</u> .	stenopetalum I	(2n = 14)	
g	<u>C</u> .	stenopetalum II	(2n = 14)	
h	c.	togoense	(2n = 14)	
j	<u>C</u> .	alismifolium	(2n = 16)	
k	Ch	lorophy tum X	(2n = 16)	
1	<u>C</u> .	caulescens (A. ca	ulescens)	(2n = 16)

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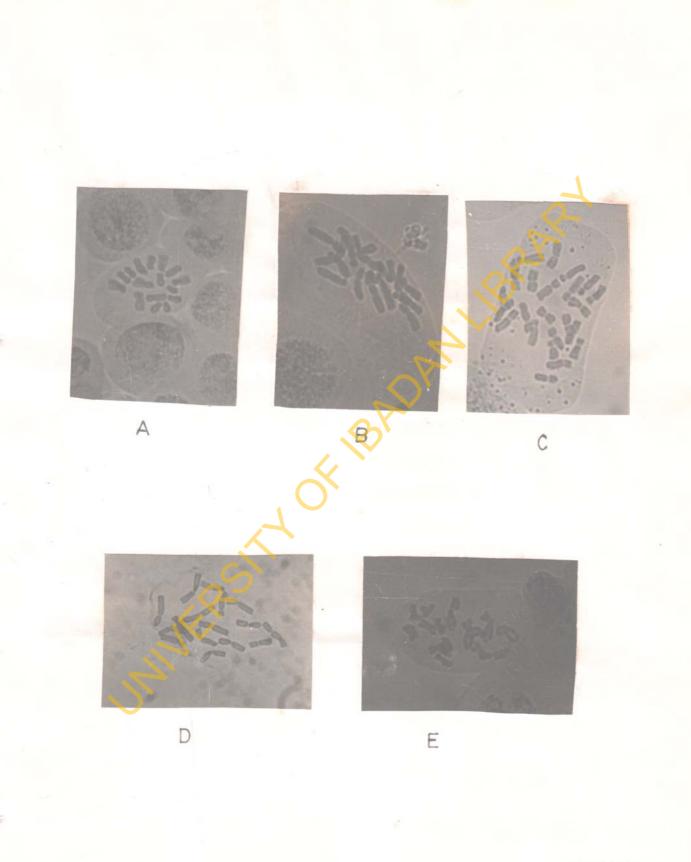


# PLATE VII

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Photographs showing metaphase somatic chromosomes in some species of Anthericum:

Α.	<u>A. lir</u>	mosum	(2n = 16)
·B •	A. nul	bicum	(2n = 16)
C .	A. pte	erocaulon I	(21 = 24)
D'.	A. pte	erocaulon I	(2n = 16)
E• ·	A. pul	birhachis	(2n = 16)



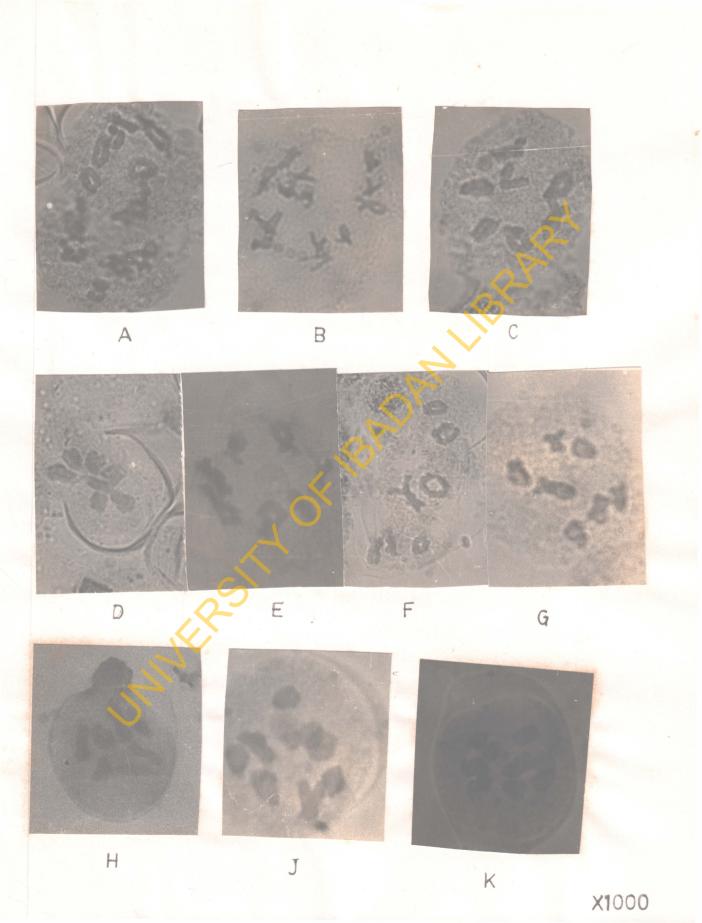
X1000

### PLATE VIII

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Photographs showing meiotic bivalent formations in some species of <u>Chlorophytum</u>:

A. <u>C. blepharophyllum</u>
B. <u>C. macrophyllum</u>
B. <u>C. macrophyllum</u>
C. <u>inornatum</u>
C. <u>inornatum</u>
D. <u>C. lazum</u>
D. <u>C. lazum</u>
E. <u>C. stenopetalum</u> I
F. <u>C. stenopetalum</u> II
G. <u>C. togoense</u>
H. <u>C. alismifolium</u>
J. <u>Chlorophytum</u> X
K. <u>C. caulescens</u> (<u>A. caulescens</u>)



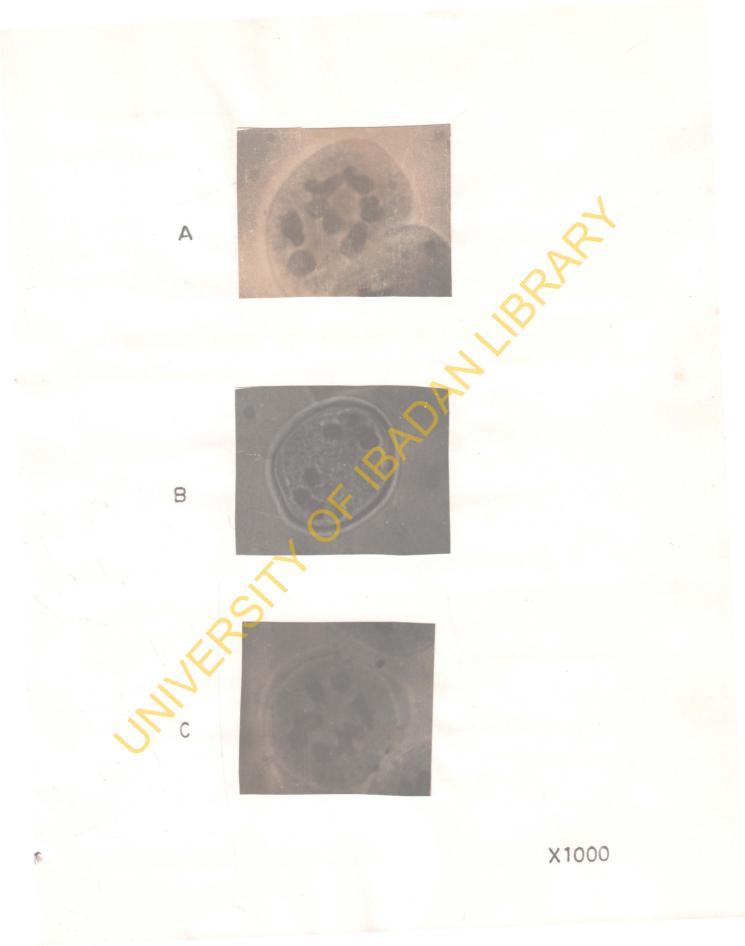
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### PLATE IX

Photographs showing meiotic bivalent formations in these species of Anthericum:

A. <u>A. limosum</u> B. <u>A. pterocaulon</u> I

C. A. pubir hachis



#### PLATE X

Photographs showing some irregularities in reproductive cells of some species of <u>Chlorophytum</u> and <u>Anthericum</u>:

- A. C. alismifolium showing two trivalents indicated with ( -> ) mark
- B. C. <u>alismifolium</u> showing the pairing of two unequal chromosomes. One of each bevalent chromosome has a satellite.

> indicates the bivalents.

- C. <u>C. alimnifolium</u> showing a pair of lagging chromosomes along the equatorial plane in (i) while the bivalent is excluded in (ii)
- D. <u>Chlorophytum</u> X showing non-formation of middle lamella initial (MLI) plates in some pollen mother cells.
- E. <u>Chlorophytum</u> X showing four groupings (2n = 16) of chromosomes; two hexavalents and two bivalents.
- F. <u>C. inornatum</u> showing four groupings (2n = 14) of chromosomes; three quadrivalents and one bivalent.
- G. <u>C. stenopetalum</u> II showing anaphase bridge at anaphase I.
- H. <u>A. pterocaulon</u> II showing association of two bivalents in the middle.

C. caulescens showing anaphase bridge at anaphase I.

J.



#### KARYOMORPHOLOGY

In <u>Anthericum</u> genus, two ploid levels were found; diploids with 2n = 16 and a triploid, 2n = 24. In <u>Chlorophytum</u> species, three types of chromosome complements were observed; diploid levels with 2n = 14 and 2n = 16 and a tetraploid level with 2n = 28.

There were no observable differences in numbers and behaviours of chromosomes of specimens fixed during field trips and those cultivated.

i. <u>C. blepharophyllum</u> (Plate VIa), is a tetraploid, 2n = 28. This complement is composed of eight long and six medium pairs. Four of the long pairs have centromere in the submedian region, at the median point in one and in the median region in the remaining three pairs. Two of the six medium pairs have centromere in the submedian region, two are subterminal pairs while the remaining two pairs are metacentric. The mean chromosome length is 7.0 microns. The meiotic cell complement is composed of six bivalents, two groups of associations each consisting of three bivalents and four univalents (Plate VIIIa)

ii. <u>C. macrophyllum</u> (Plate VIC) is a tetraploid, 2n = 28. The complement is composed of ten long and four medium pairs. Four of the long pairs have the centromere in the medium region, three pairs are submedian while the remaining three long pairs are subterminal chromosomes. Two of the medium pairs are, metacentric one pair is submedian while the fourth pair has the centromere in

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the median region. The mean chromosome length is 7.14 microns.

The meiotic complement is composed of fourteen bivalents (Plate VIIIb). Eleven of the fourteen bivalents usually have two chiasmata each while each of the remaining three bivalents usually has one chiasma. Meiotic division is normal.

**111.** <u>C. geophilum</u> (Plate VIb) is a tetraploid, 2n = 28. The complement is composed of twelve long and two medium pairs. Six out of the twelve long pairs have the centromere in the submedian region, in the median region in five pairs while the twelfth has its own at the median point. The two medium-sized pairs have the centromere at the median point. The mean length of chromosome is 7.43 microns.

The plants did not flower before they withered and died, hence meiotic behaviour could not be investigated.

iv. <u>C. inornatum</u>, is a diploid, 2n = 14 (Plate VId) composed of seven long pairs. The centromere in three pairs, is in the submedian region, in median region in two pairs while the sixth and seventh pairs are metacentric and subterminal respectively. The mean chromosome length is 8.93 microns.

The meiotic complement is composed of seven bivalents (Plate VIIIc). Two bivalents form rings by having their chiasmata at their two distal ends. One bivalent has one distal chiasma. In some cells, three quadrivalent associations and a bivalent are usually observed (Plate Xf) in approximately 10% of the meiotic cells observed. v. <u>C. laxum</u> (Plate VIe) is a diploid, 2n = 14. The somatic complement is composed of five long and two medium pairs. Two of the five long pairs have the centromere in the median region, while the remaining three pairs have it in the submedian region. One of the two medium pairs has its centromere at the median point while the second pair has it in the submedian region. The mean chromosome length is 7.71 microns.

The meistic complement is made up of seven bivalents. Each bivalent usually has two chiasmata. Meiotic division is normal (Plate VIIId).

vi. <u>C. stenopetalum</u> I (Plate VIf) is a diploid, 2n = 14. The complement is composed of five long and two medium pairs. Three long pairs have the centromere in the submedian region, one is metacentric while the fifth has the median region. One medium-sized pair has the centromere in the subterminal region while the second pair is metacentric. The mean chromosome length is 7.14 microns.

The meiotic complement (Plate VIIIe) is composed of seven bivalents. The mean frequency of chiasma. formation is 1.97 as presented in Table 12: Meiotic division is normal

vii. <u>C. stenopetalum</u> II (Plate VIg) is also a diploid, 2n = 14. The complement is composed of five long and two medium pairs. Three of the long pairs have the centromere in the median region, one pair is submedian, while the fifth pair has its own at the median point. One of the medium-sized pairs has the centromere in the subterminal region while the second pair is metacentric. The mean chromosome length is 8.0 microns.

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The meiotic complement (Plate VIIIf) is composed of seven bivalents with usually two chiasmata each. The mean frequency of chiasma formation is 2.01. Four of the bivalents have their chiasmata at their distal ends. The meiotic division is normal. However, anaphase bridges were observed at the anaphase I in 2% of the meiotically dividing cells (Plate  $\mathbf{x}$ g).

viii. <u>C. togoense</u> (Plate VIh) is a diploid, 2n = 14; composed of six long and one medium-sized pairs. The centromere is in the submedian region in four long pairs, in the median region in one and at the median point in the sixth pair. The only medium-sized pair is metacentric. The mean chromosome length is 7.57 microns.

Meiotic complement (Plate VIIIg) is composed of seven bivalents with a mean chiasma frequency of 1.96 (Table 12). The meiotic division is normal.

ix. <u>c. alismifolium</u> (Plate VIj) is a diploid 2n = 16, composed of four long and four median-sized pairs. Two of the four long pairs: have the centromere in the submedian region, while it is in the median region in the other two. The centromere is at the median point in one of the medium-sized pairs, in the median region in two and in the subterminal region in the fourth pair. The mean chromosome length is 6.38 microns. The meiotic complement (Plate VIIIh) is composed of eight bivalents. One of the bivalents has two distal chiasmata and thus forms a 'ring'. In over 70% of the pollen mother cells investigated, five bivalents and two trivalent associations were commonly found (Plate Xa). The association is between one bivalent and one univalent of a bivalent which does not pair. It is specifically between a 'ring' bivalent (i.e. chromosome v) and a 'rod' bivalent (i.e. chromosome vii). Two excluded chromosomes (Plate Xc) were found along the equatorial plane or by the sides in 60% of the pollen mother cells investigated. It may be that the associated univalent chromosomes were those that were excluded. It is possible that the associated univalent chromosomes were not properly attached to the spindle during the late metaphase stage, hence they were excluded at the anaphase stage of division.

Two bivalents are of unequal sizes (Plate Xb). This might have resulted from the loss of a chromosome segment in one of the pairs. The other possibility is that there was a spontaneous translocation between one chromosome of the two bivalents which had now stabilised. This is possibly what had happened, since each longer chromosome in both bivalents was having satellite. x. <u>Chlorophytum</u> X(Plate VIk) is a diploid, 2n = 16, composed of five long and three medium - sized pairs. One of the five long pairs is metacentric, two have the centromere in the submedian region, in the median region in one and in the sub-terminal region in the fifth pair. One medium-sized pair is metacentric while the other two have centromere in the submedian region. The mean chromosome length is 6.88 microns.

There are irregularities in the meiotic divisions. Nonformation of middle lamella initial(MLI) plates at telophase I and II is a common occurrence (Plate Xd). There are eight bivalents (Plate VIIIj). In 23% of the pollon mother cells observed, four groupings of chromosomes were found. This is made up of two hexavalents and two bivalents (Plate Ke). The anaphase stages were rarely observed.

xi. <u>C. caulescens</u> (Bak.) Marais & Reily (<u>A. caulescens</u>) Plate VI is a diploid, 2n = 16; composed of seven long and one medium-sized pairs. Three of the long pairs have the centromere in the submedian region, in the median region in another three pairs while the seventh pair has its centromere in the subterminal region. The medium-sized pair is metacentric. There is a secondary constriction in chromosome II. The mean chromosome length is 8.25 microns.

Meiotic division is normal in most cases (Plate VIIIc), showing eight bivalents. However, few cases of anaphase bridges were found. (Plate Xj). -109 -

xii. <u>A. limosum</u> (Plate VIIb) a diploid with 2n = 16, composed of five long and three medium-sized pairs. The centromere is in the submedian region in three pairs, in the median region in one and subterminal in the fifth long pair. Two of the medium-sized pairs have the centromere in the submedian region, while the third pair is metacentric. The mean chromosome length is 6.63 microns.

The meiotic division was normal, composed of eight bivalents (Plate IX2). The frequency of chiasma formation (Table 12) is 1.98. xiii. A. nubicum (Plate VIIb) is a diploid with 2n = 16, with six long and two medium-sized pairs. Four of the six long pairs have their centromere in the submedian region, one in the median region, while the sixth pair has its own in the subterminal region. One of the two medium pairs is metacentric, while the other has its own in the subterminal region. One of the two medium pairs is metacentric, while the other has its own in the submedian region. The mean chromosome length is 6.56 microns.

The plant did not flower before it died, hence meiotic investigation was not done.

xiv. A. <u>pterocaulon</u> I (Plate VIIC) a triploid, 2n = 24 is composed of four long and four medium-sized trivalents. Two of the long trivalents have the centromere in submedian region, in median region in one and in the subterminal region in the fourth. .... One of the medium-sized triads has the centromere at the median point, in the median region in another, submedian in the third while the fourth has its own in the subterminal region. The mean chromosome length is 6.56 microns.

The plant did not flower before it withered and died. xv. A. pterocaulon II (Plate VIId) is a diploid, 2n = 16. The somatic complement is composed of three long and five medium pairs. Two of the long pairs have their contromere in the submedian region while the third has its own in the median region. One of the five medium-sized pairs is metacentric, two have their centromere in the median region, submedian in another while the fifth has its own in the subterminal region. The mean chromosome length is 6.25 microns.

The meiotic division is normal. The complement is composed of eight bivalents (Plate IXb). However, association between two bivalents is observed (Plate Xh) in13.4% of the pollen mother cells observed. The frequency of chiasma. formation is 1.99.

xvi. A. <u>pubirhachis</u> (Plate VIIE) is a diploid, 2n = 16. The complement is composed of one long and seven medium-sized pairs. The long pair has the centromere in the submedian region. One of the medium-sized pairs has the centromere at the median point, it is in the median region in two, and in the submedian region in the remaining three pairs. Endomitosis occurs in 81% of the cells investigated. The mean chromosome length is 5.13 microns.

The meiotic division is usually normal, The complement is composed of six bivalents and one quadrivalent.

#### DISCUSSION

Cytological results have been found useful in solving taxonomic problems. The usefulness of cytology in analysing the possible relationships between Anthericum and Chlorophytum complex cannot therefore be ignored. The chromosome is a stable and definite species character as it has been found in other plants. Within the same sportes, indivisuals usually have somatic and meiotic chromosomes of reasonably constant morphology (Marenah and Holden, 1967). As can be seen in Tables 8 and 9, Plates VI, VII., VIII and IX, each species has its own specific karyotype morphology, even though similarities exist in the whole complex. The summary of similarities and specificity of the chromosomes in the complex (See Table 10) reveals that no two species have the same complement. The tetraploid complements do not show any evidence of direct doubling of their diploid relatives. Thus the tetraploids might have been allopolyploids.

The method used by measuring only chromosomes that were about 2 um in thickness helps to have standardised measurements of the chromosomes. The means for indivisual chromosome lengths in each species and for each species, as presented in Tables 8 and 9, show that the <u>Chlorophytum</u> species have larger chromosomes than <u>Anthericum</u>. Taking <u>C</u>. <u>alismifolium</u> and <u>Chlorophytum</u> X as intermediates, as suggested earlier, the mean chromosome length for <u>Chlorophytum</u> ranges from 7.0 um to 8.93 um., while those of <u>Anthericum</u> range from 5.13 um to 6.56 um. The mean value for

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<u>C. alismifolium</u> and <u>Chlorophytum</u> X are 6.38 umand 6.88 um respectively. The total chromosome length for <u>Chlorophytum</u> ranges from 50 um to 66 um <sup>•</sup> while those of <u>Anthericum</u> are from 41 um to 53 um. Those of <u>C. alismifolium</u> and <u>Chlorophytum</u> X are 51.0 umand 55 um respectively. It therefore seems that species having their basic chromosome number as n = 8, except <u>C. caulescens</u>, have smaller chromosomes than those in which the basic number is 7.

A basic cause for differences in chromosome size among related species is the genotype (Thomas, 1936; Rees, 1961; Fernades, 1961; and Jackson, 1971). Other sources of differences in chromosome size among related taxa are not completely explicit and they can only be hypothesized (Tobgy, 1943: Stebbins, 1950 and Brown, 1966). Navashin (1934) reported more varied chromosome sizes in parent cells of Crepis than in hybrid cells. Tobgy (1943) could not observe such differences in the chromosomes of Crepis neglecta and C. fulignosa and their hybrid. He therefore postulated that the size of chromosomes is independently controlled by genes located close to the centromere. Olorode (1973) working on Emilia, went further to postulate that a single gene producing a genome-specific and genomewide effect with respect to contraction of chromosomes in hybrid cell appears more plausible, if the difference in size is considered to be of genetic origin. Thus the differences in the size of chromosomes in the Anthericum and Chlorophytum complex is possibly due to genetic constitution of the complex.

Three chromosomal levels are found in <u>Chlorophytum</u>: diploid levels with 2n = 14 and 2n = 16, and a tetraploid level with 2n = 28. In <u>Anthericum</u>, two chromosomal levels are found; a diploid with 2n = 16 and a triploid level of 2n = 24. Thus it is not possible to separate the two genera on the basis of their basic chromosome numbers as suggested by Baldwin and Speese (1951) and Darlington and Wylie (1955).

<u>C</u>. elatum R.Br. has been reported to be 2n = 28 by many workers including Storey, (1968; Koul, (1970); and Zama & Matha, (1976) while Deshpande (1955a) recorded 2n = 32 for the same species. Also <u>C</u>. <u>laxum</u> (African species) has been reported to be 2n = 14 while the Indian material is 2n = 16. The two populations thus have their basic numbers as 7 and 8. **Ploid** levels reported for <u>C</u>. <u>orchidastrum</u> are diploid, 2n = 14(Boraiah and Fathima, 1971; Sheriff and Rao, 1971), tetraploid, 2n = 28, hexaploid, 2n = 42 (Jones & Smith, 1967; and Sheriff, 1967), while Larsen (1963) reported it to be octoploid 2n = 56. These differences in chromosome numbers may possibly be due to various degrees of endomitotic phenomena which the workers might not have taken into consideration. The other possibility is that some endomitotic cells might give rise to different polyploid levels since these plants propagate vegetatively very easily.

However, in this work, chromosome counts for Nigerian species of <u>C. inornatum</u>, 2n = 14; <u>C. macrophyllum</u>, 2n = 28; <u>C. blepharophyllum</u>, 2n = 28; <u>C. geophilum</u>, 2n = 28; <u>C. stenopetalum</u> I, 2n = 14; <u>C. stenopetalum</u> II,

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2n = 14; C. laxum, 2n = 14, and C. togoense, 2n = 14 agree with the basic number of 7. On the other hand, C. alismifolium, 2n = 16; Chlorophytum X, 2n = 16; C. caulescens, 2n = 16; A. limosum, 2n = 16; A. nubicum, 2n = 16; A. pterocaulon I, 2n = 24; A. pterocaulon II, 2n = 16; A. uyuiense, 2n = 16; A. pubirhachus 2n = 16 have their basic chromosome number as 8. The basic number 8 recorded for C. alismifolium and Chlorophytum X has been reported by some workers on these and other Chlorophytum species. Chlorophytum species with basic number of 8 are C. alismifolium 2n = 16 C. tuberosum, 2n = 16, C. acutale 2n = 16; C. glaucuum, 2n = 16 and C. bharuche, 2n = 16 (Kumar and Rao, 1958; Sheriff and Chennaveeraiah, 1972; Naik, 1976), C. comosum 2n = 24, C. sterubergianum 2n = 24, C. bracteatum 2n = 48, C. elatum 2n = 32 (Schnarf and Wunderlich, 1939; Deshpande, 1955a; and Jones & Smith 1967). The 2n = 16 reported for <u>Chlorophytum X in this work does not agree with the 2n = 28 and 2n = 32</u> reported for C. elatum var. variegatum (Deshpande, 1955a), hence this taxan may not be C. elatum. Storey (1968) reported varied chromosome numbers in the same root tip of C. elatum ranging from 2n = 28 to 56 and 84 and even 12, Deshpande (1955b) reported 2n = 24, 32 and even 64 for A. variegatum Hort ex Fl., a synomyn of C. elatum.

Other species in this complex having chromosome numbers with different basic numbers have been reported. For example <u>C</u>. <u>khasiamum</u> 2n = 22 and 2n = 30 (Jones and Smith, 1967; and Sarkar <u>et</u>. <u>al</u>, 1973), <u>L</u>. <u>longipedunculatum</u>, 2n = 46 (Riley, 1962), <u>A</u>. <u>liliago</u>, 2n = 64 and 2n = 60

and A. ramosum 2n = 32 and 2n = 30 (Elvers, 1932; Strandhede, 1963) have been recorded. These confusions might have been due to mistaken identity or as a result of hybridization between those having basic numbers of 7 and 8. The complement of such hybrid would be composed of two similar but not necessarily genetically identical chromosome sets. In order to ensure normal meiotic division to operate, there would be chromosome doubling which might have resulted in a tetraploid with 2n = 30 and octoploid with 2n = 60. The diploid C. khasiamum, 2n = 22might have been due to counting of chromosome fragment along with the other chromosomes (i.e. 21 chromosomes + 1 fragient chromosome = 22). A similar condition has been reported in Urginea altissima by Oyewole, (1980) in which two fragmental chromosomes were counted along with the normal 20 to give 2n = 20 + 2ff = 22. The 2n = 46 recorded for C. longipedunculatum by Riley (1962) might have been due to loss of a pair of chromosomes from a 2n = 48. In the same way, 2n = 30 and 2n = 60. might have been derived from 2n = 32 and 2n = 64 respectively. In the light of the above argument, one could fairly accept the basic chromosome numbers for the complex as 7 and 8 (Darlington & Wylie. 1955).

The chromosome morphology reveals a lot of similarities between the species of the same genus and between the two genera (Table 10). These similarities can not be accidental, for they might have been responsible, at least in part, for the high degree of vegetative morphological similarities in the genera. Though close similarities in chromosome morphology may not in itself indicate a genetic relationship as stated by Smith (1933), Dobzhansky (1935), Stebbins (1950) and Jones (1970) but a correlation between the vegetative morphological similarities and chromosome morphology in the two genera, presupposes a level of genetic relationship (Oyewole, 1972, 1975). These similarities in the karyotypes and phenotypes possibly suggest a common evolutionary ancestry for both genera.

Every species of both genera has at least one metacentric chromosome almost of the same size and a subterminal chromosome as can be seen from Tables 8, 9 and 10. A closer look and careful studies of the chromosome lengths and morphology have led to the identification of a basic chromosome number of 4. These are titled 'hypothetical original chromosomes A, B, C and D' in both genera. Each hypothetical chromosome is duggested as follows:

Chromosome		A	В	C	D
Absolute Length (um)	C	8 - 10	9 - 10	9 - 11	7 - 10
Long arm (um)	1	4	6	7	8
Short arm (µm)	S	4	4	3	2
r - value	r-v	1.0	1.5	2.3	4.0
Location of centromere	loc	М	m	Sm	st.

## HYPOTHEFICAL ANCESTRAL CHROMOSOMES OF AN EHENICUM AND CHLOROPHYFUM COMPLEX

These hypothetical ancestral chromosomes, due to translocations, inversions, loss of chromosome segments, breaks and bridges and loss or fusion of chromosomes may have resulted in their present forms as presented in Table 10.

In spite of many structural changes that might have occurred in these 'hypothetical chromosomes', their morphologies are still somehow retained. The total haploid chromosomes found in the taxa investigated is 141. Out of this, 17.73% are metacentric (i.e. chromosome A) while 36.17% of the chromosomes have their centromere in the median region (i.e. chromosome B). Submetacentric chromosome C claims 34.04% while 12.06% are subterminal chromosome D type. A loss or gain in chromosome long or short arms will alter its r-value. It is therefore reasonable to suggest that some metacentric chromosomes and a few of submetacentric chromosomes contributed to the high percentage of chromosomes in B chromosome group. In like manner, some chromosomes originating from D chromosome might have been modified into submetacentric C chromosome. The alterations in the morphology of D chromosome is expected, since subterminal or acrocentric chromosomes are more inclined to structural and morphological changes (Stebbins, 1971).

The speculation here, is that the basic chromosome number for the two genera is 4; which possibly had the same morphology as the suggested hypothetical chromosomes. It is also speculated that there were two closely related individual taxa with 2n = 8 which were the ancestral

parents of these genera. There was possibly inter-breeding between the ancestral parent taxa. Since similar and/or identical chromosomes in related species, may not be 100% identical in gene arrangements, it is speculated that the genome of the cross or hybrid was followed by chromosome doubling. Plants in this complex propagate both vegetatively and sexually. Endomitosis is commonly reported in these plants. The other speculation is that a hybrid with 2n = 8 had some endomitotic cells which propagated vegetatively to give rise to 2n = 16 plants.

Some chromosomal aberrations and alterations in the cytoplasm arising from the initial hybridisation accumulated and resulted in different gene arrangements plus cytoplasmic genetic materials (e.g. cytogenes). These new gene arrangements eventually led to morphological changes and speciations.

Evidence. of chromosome alterations bas been reported in these genera. Thomas (1959) reported spontaneous reciprocal translocation in <u>C. hynei</u> involving satellites. Chromosome inversions and translocations have been observed in <u>C. comosum</u> by Sheriff (1967). The pairing of unequal chromosomes in two bivalents of <u>C. alismifolium</u> has been attributed to either loss of chromosome segments or reciprocal translocations (Plate Xb) by the present worker. This can not be a genetic mechanism to transmit the pair of univalents, because the trivalents are still clearly observable in the Plate. However, this is an evidence of chromosome alteration. Though translocation is not very significant in plant evolution, taxonomic relationship is usually correlated with similarity in chromosome morphology (Hollingshead and Babcock, 1930; Babcock and Cameron, 1934; and Levan, 1935). On the other hand, Ahmad et al (1977); have suggested that chromosome inversion plays some role in isolating two groups of soybeans growing in the same geographical area for centuries. It is therefore reasonable to deduce that cumulative translocations and inversions in the chromosome complements of <u>Antherioum</u> and <u>Chlorophytum</u> might result in re-arrangement of genes on the chromosomes which affected their phenotypic characters and effected their separation.

Tauber and Tauber (1977) had shown that two unlinked autosomal loci resulted in different photoperiodic responses in a cross of <u>Chrysopa</u> <u>carnea X Chrysopa downesi</u>. Harlan and De Wet (1977) observed unusual phenotypic characters in maize as a result of genetic transfers from <u>Tripsacum</u> to maize in maize X <u>Tripsacum</u> cross. Definitely any accidental cross between two related taxa will have more than two unlinked autosomal loci, when this is coupled with genetic transfers, it will lead to some physiological, morphological and anatomical differences, thus cumulating in phenotypic characters. The stabilisation of these differences will undoubtedly lead to speciation which may get as far as generic level.

# MEIOTIC IRRECULARI FIES

Zama and Matin (1976), analysing the meiotic irregularities in C. elatum (2n = 28) believed that C. elatum is an allotetraploid.

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Storey (1968) suggested that the formation of sesquihaploid and hemihaploid cells in pollen mother cells of C. elatum (2n = 28) is evidence of its being an autotetraploid. Meiotic abnormalities or irregularities have been suggested to occur through four causes, which are; by hybridity, unbalanced chromosome complements, unfavourable climatic conditions and mechanical injuries and changes in temperature (Stebbins, 1971). Olorode (1972) attributed meiotic irregularities in some Nigerian plants (e.g. multivalents and univalents, laggards and nondisjunctions) to structural heterozygosity or polyploidisation. Structural heterozygosity may originate from hybridisation of taxa that differ by a number of chromosome re-arrangements or from spontaneous chromosome re-arrangements (Star, 1970). The considerable amount of meiotic irregularities in Hyparrhenia involucrata and H. subplumosa made Olorode (1973a) suggest that they are polyploids which originated in the recent past.

The formation of multivalents and univalents, laggards and non-disjunctions in <u>Chlorophytum</u> X with 2n = 16, is evidence of its being a polyploid which originated in the recent past through structural heterozygosity. Meiotic irregularities in <u>C. elatum</u> have been reported from South Africa and Bangladesh and thus it may not be due to climatic conditions. The view that the irregularities in this taxon is due to polyploidy has been held by Storey (1968) and Zama and Matin (1976). Moreover, the non-formation of middle lamella initial in this taxon, and the four groupings of its meiotic chromosomes possibly suggest that <u>Chlorophytum X is a polyploid which has not stabilized.</u> If <u>Chlorophytum X</u> (2n = 16) is a polyploid, possibly an allotetraploid, then its basic chromosome number is 4. This view is further strengthened by the four groupings of the chromosome complement during meiosis (Plate Xe). Nai's (1977) working on Indian species of <u>Chlorophytum</u> also believed that the basic chromosome number for these plants is 4.

Non-formation of middle lamella initial to separate the four daughter cells in <u>Allium odorum</u> and <u>Crepis virens</u> has been attributed to low temperature (Yasui, 1931). Devise (1922) working on <u>Papaver sammiferum</u> X <u>P. orientale</u> concluded that the non-formation of middle lamella initial was due to simultaneous pollen dyad and tetrad formations. The nonformation of middle lamella initial in <u>Chlorophytum</u> X (Plate Xd) is possibly due to simultaneous formation of dyads and tetrads, for other stages of meiotic division were not clearly observable.

The formation of three quadrivalent associations and one bivalent in <u>C. inornatum</u> (Plate Xf) suggests that a bivalent might have been lost. Each quadrivalent seemingly has the same origin (Moore, 1976) while the bivalent might have lost its second bivalent, either through fusion and/or translocation to other chromosomes. However, the four groupings pre-suppose that the taxon arose from a lower chromosome basic number, probably 4.

Allopolyploidy is known to have played an important role in plant evolution. It is modified by the action of duplicate genes and by factors which were once identical, differentiated and govern the same character in different ways (Lawrence 1931). Environmental conditions have been suggested to influence pairing in chromosomes (Ahmad, <u>et. al</u>, 1977). However, the importance of environmental factors in influencing genetic manifestions rests in their independent changes to which specific genes respond differently (Kenneth, 1975). Thus allopolyploidy and environmental factors coupled with chromosome inversions, translocations, gene mutations and spontaneous re-arrangements in some particular chromosomes affect plant genotypes which manifest in phenotypic differences. Changes in the phenotypic characters will lead to speciation. These taxa, which have the same ancestry have now diverged in some way to reach generic levels with some intermediates.

The possible evolution of the 'secondary' basic chromosome numbers of n = 7 and n = 8 have been argued in both ways. Sheriff (1967) and Naik (1976, 1977) strongly believe that the n = 8 is more primitive and has less specialised karyotypes than n = 7. Thus, they suggested that n = 7 evolved from n = 8 by elmination of a pair of chromosomes. On the other hand, Baldwin and Speese (1951) argued that n = 8 evolved from n = 7by duplication of a chromosome.

Naik's (1977) argument was based on his study of <u>C</u>. <u>laxum</u>. He observed two lagging chromosomes in the Indian species with 2n = 16 which he believed had been lost in the African species by elimination. In support of his view a pair of lagging chromosomes is observed in <u>C</u>. <u>alismifolium</u>, 2n = 16. The elimination of this may result in n = 7. But this idea may not likely hold since this taxon is partially sterile. In most cases, only one or two fruits develop in an inflorescence having ten or more flowers. It therefore means that the lagging chromosome must be important in fruit development, thus their elimination will lead to sterility. But the <u>C</u>. <u>laxum</u>, 2n = 14, is fertile. Thus the lagging homologous chromosomes might not have been eliminated. Moreover, the total and mean chromosome lengths in <u>Chlorophytum</u> (n = 7) are greater than those with n = 8. It is therefore most unlikely for smaller chromosome to give rise to greater total chromosomes by elimination.

Baldwin and Speese (1951) based their own argument on <u>C</u>. <u>alismifolium</u>, 2n = 16. They observed a trivalent and a univalent in the metaphase I chromosome of the plant. They then believed that one of the chromosomes duplicated, resulting in n = 7. In support of his view two trivalents were observed in <u>C</u>. <u>alismifolium</u> by the present worker in over 80% of pollen mother cells observed (Plates Z a & b). There was no record of a univalent as claimed by earlier workers. I believe that the two propositions are possible. The excluded pairs of chromosomes in <u>C</u>. <u>laxum</u> (Maik, 1977) and in <u>C</u>. <u>alismifolium</u> (Plate Xc) were not actually eliminated. These chromosome(s) or chromosome fragments possibly fused with other chromosomes to give rise to n = 7, hence basic number 7 is derived from 8 by chromosome fusion and not by elimination as suggested by Naik (1977). This phenomenon has been reported in <u>Drosophila</u>.

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D. subobscure (n = 6) is believed to have given rise to D. pseudoobscura (n = 5) by a fusion of two chromosomes. In the same way D. <u>melanogaster</u>, (n = 4) evolved from D. <u>pseudoobscura</u> (n = 5) (Patterson, 1978). Tobgy (1943) has suggested that <u>Grepis fuliginosa</u>, n = 3 was derived from the closely related <u>C. neglecta</u> (n = 4). Sherman (1946) also believed that <u>Grepis kotschyana</u>, (n = 4) was derived from <u>C. foetide</u> (n = 5). It is my belief that one of the chromosomes which originated from hypothetical chromosome. D possibly fused with one of the submetacentric chromosomes.

Four of the eight taxa having their 'secondary' basic number as 8 (i.e. <u>Chlorophytum X, C. alismifolium, C. caulescens</u>, and <u>A. pterocaulon</u> II) show some meiotic irregularities. They have more primitive chromosomes. These also support the view that the n = 8 taxa are more primitive than those with n = 7 (Naik, 1977).

## CHLOROPHYTUM STENOPETALUM COMPLEX

Two <u>C</u>. <u>stenopetalum</u> populations were investigated. The variation in the chromosome lengths of both populations was statistically analysed and shown in Table 11. For record purposes, the populations are denoted as <u>C</u>. <u>stenopetalum</u> I and <u>C</u>. <u>stenopetalum</u> II. The separate variance t model test of the mean chromosome lengths and r-values show less t values than the critical t values, hence the variations are due to chance alone. The F ratio values for the homogeneity of variances of the characters are also less than the tabled F values. Since the tabled values exceed the computed F values, one may consider the variance in the two populations to be homogenous(**Popham** and Sirotnik 1973). This is expected since the whole genera arose from the same ancestral parents.

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The formation of chiasmata in the bivalents of the two populations is remarkable (Plates will e& f). As can be seen in Table 13, only chromosome III of both populations has the same type of chiasmata (i.e. terminal and interstitial chiasmata). Chiasma formation is genetically controlled. Hence differences in its formation indicate a degree of genetical differences (Rees 1961).

On the bases of mitotic and meiotic studies, and the morphological differences stated earlier in this work, it is strongly believed that the two populations should be separated as different species.

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### CHAPTER 6

#### HYBRIDIZATION TESTS

Inter-generic hybridization tests were made between <u>Anthericum</u> and <u>Chlorophytum</u> species, with a view to identifying the extent of their genetic affinities.

The studies of the morphology, cytology, flowering periods and fruiting in the plants were done in 1977. These studies were necessary to observe and select the "marker" characters of the plants to be used in crosses.

In 1978, crosses were made between <u>A</u>. <u>limosum</u>, <u>C</u>. <u>caulescens</u> and <u>C</u>. <u>alismifolium</u> on the basis of their chromosome numbers (2n = 16). <u>C</u>. <u>laxum</u> with 2n = 14, was also involved in the crosses on the basis of its seeming intermediate morphological position.

In 1979 and 1980, crosses were carried out on <u>A. linosum</u>, <u>A. pterocanlon</u>II, <u>A. pubirhachis</u>, <u>C. caulescens</u> and between <u>C. laxum</u>, <u>Chlorophytum X, C. alismifolium, C. togoense, C. inornatum</u> and <u>C.</u> <u>blepharophyllum</u>. Three sites were used for the hybridization tests: i. Nursery, Botany Department, University of Ibadan, ii. Ilese, in Ijebu-Ode District of Ogun State; iii. Botanical Garden University of Ibadan, Ibadan.

# A. METHOD

## (i) <u>CROSSES</u>:

Each of the three <u>Anthericum species was used as both recipient</u> and donor for every one of the eight <u>Chlorophytum</u> species. At least ten individual plants were used as female parents for each combination. At least ten flowers of each individual plant were involved in the forty-eight combinations, hence over ten thousand crosses were made. The flowering period in these plants overlaps between February and September. This ensured crosses for at least six months.

# (ii) <u>EMASCULATION</u>:

Plants meant for crosses were separated from others during flowering. The anthers of recipient plants were carefully removed in the morning when the flowers were just opening so as not to injure the stigmas. The removed anthers were put in alcohol so as to kill them and thus prefent accidental pollination from them by wind. The anthers were removed so as to prevent selfpollination. The donor plants were kept at a distance to prevent accidental pollinations.

(iii) ARTIFICIAL POLLINATION:

The anthers usually dehisced around 12.00 noon and the stigmas were usually ready to receive the pollen grains from the same time till 6.00 p.m. The stigma was usually identified as being ready when it contained some watery substance. Some good and well placed flowers on each recipient's inflorescence were selected for hybridization while others were destroyed. The whole donor flower was removed with a clean pair of forceps from the donor plant. The pollen grains were carefully dusted on the stigmas. The whole inflorescence of the recipient plant was then loosely covered with polythene bag which was then tied around the peduncle. The covering of the flowers was to prevent pollen grains from other plants or flowers being blown by wind.

(iv) POLLEN GRAINS VIABILITY TEST:

Ten flowers with dehisced anthers were collected from each species. The pollen grains from each flower were dusted on clean slides one after the other. They were then stained with 2% acetocarmine solution. Each slide was carefully covered with clean cover-slip and observed under light microscope. Properly-stained pollen grains containing nuclei were regarded as being viable. Those that were empty and/or slightly stained were regarded as being aborted. The aborted pollen grains were counted in each field of view of the light microscope. The whole number of pollen grains viewed at different fields of view were counted together. The total number of pollen grains and the number of aborted pollen grains were recorded for each species. The percentage viability was calculated by subtracting the number of aborted pollen grains from the total pollen grains recorded. The difference was then divided by the total and multiplied by 100.

100 /200

where T means the total number of pollen grains. a means the number of aborted pollen grains.

# (v) GERMINATION OF POLLEN GRAINS

The hanging drop method devised by Oyewole (1979) was used. Mature pollen grains from dehisced anthers were dusted on a clean slide. A drop of distilled water was added. The slide was then turned face down and hung over an opened cover-slip case in which distilled water was put. The coverslip case with the slide were placed inside labelled petri dish containing distilled water. It was then covered and set up in the Laboratory, Department of Botany, University of Ibadan, for 12 hours, 24 hours and 36 hours. The number of pollen grains and those that germinated were counted in five plants for each species after 24 hours and recorded. The average was found for each species and expressed as percentage. Germination of pollen grains on stigmas was also examined with hand lens. In some cases, the whole pistils were removed for examination under a light microscope.

# B. RESULTS:

The percentage of pollen grains viability and germination of pollen grains in the species involved in the cross are presented in Table 14. The results of hybridization tests in some Nigerian species of <u>Anthericum</u> and <u>Chlorophytum</u> are presented in Table 15.

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# TABLE 14

# POLLEN GRAIN VIABILITY AND GERMINATION OF SPECIES INVOLVED IN HYBRIDIZATION

	POLLEN	GRAIN VI	ABILITY	POLLEN GRAIN GERMINATION			
SPECIES			% OF VIABLE P.GRAINS	NO. OBSERVED		% of P.grain germin- ateon	
C. blepharophyllum	1000	927	92.7	156	82	52.6	
C. inornatum	1000	960	96.0	440	337	76.6	
C. laxum	1000	817	81.7	610	131	21.5	
C. togoense	1000	980	98.0	510	441	86.5	
C. alismifolium	1000	685	68.5	750	188	25.1	
Chlorophytum X	1000	368	36.8	252	4	1.6	
C. caulescens	1000	938	93.8	620	409	66.0	
A. limosum	1000	507	50.7	675	27	4.0	
A. pterocaulon II	1000	635	63.5	150	5	3.3	
A. pubirhachis	1000	534	53.4	120	49	40.8	

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#### TABLE 15

RESULTS OF HYBRIDIZATION IN SOME NIGERIAN SPECIES OF ANTHERIUM AND CHICROPHYTUM

SPECIES		<u>C. alismifolium</u> 2n = 16	<u>c.</u> X 2n = 16	$\frac{C}{2n} = 14$	<u>C. inornatum</u> 2n = 14	$\frac{c. togoense}{2n = 14}$	<u>C. blepharophyllum</u> 2n = 28
C. caulescens	δ	Pollen tubes produced. Zygote formed.	Pollen tube produced. No. syngamy	Syngamy occurred	Syngamy occurred	Syngamy occurred	Syngamy occurred
2n = 16	đ.	No syngamy ,	No pollen germination	Syngamy occurred	Syngamy occurred	Syngamy occurred	Syngamy occurred
A. linosum	02	Short pollen tubes formed.	Pollen grains germinate but very short pollen tubes	Pollen tubes produced	Pollen tubes produced	Pollen tube produced	Pollen tubes produced
2n = 16	9	Long pollen tubes but no syngamy.	No pollen germination	Syngamy occurred	Syngany occurred	Syngamy occurred	Syngamy occurred
A. pterocaulon II	3	Pállen tubes produced.	Pollen grains occasionally germinate	Long pollen tubes formed	Pollen tube formed	Short pollen tubes produced	Syngamy occurred
2n = 16	q	<ul> <li>Syngamy occurred in some cases;</li> </ul>	No pollen germination	Syngamy	Syngamy occurred	Syngamy occurred	Syngamy occurrod
A. pubirhachis	3	Short pollen tubes	Pollen usually germinate	Short pollen tubes formed		Short pollen tubes produced	Short pollen tubes formed
2n = 16	4	Very short pollen tubes produced	No pollen germination	Syngamy occurred	Syngamy occurred	Syngamy occurred	Syngamy occurred

The non-formation of mature fruits in <u>A</u>. <u>limosum</u>, <u>A</u>. <u>pterocaulon</u> II and <u>A</u>. <u>pubirbachia</u> in cultivation, may be one of the possible causes of non-formation of matured seeds in the crosses involving these plants. The fact that <u>A</u>. <u>Limosun</u>, <u>A</u>. <u>pterocaulon</u> II and <u>A</u>. <u>pubirbachin</u> propagate vegetatively in cultivation may be a means of selection to favour a few highly distinctive genotypes (Brandshaw, 1971). Geographical preferential selection of habitats by plants has been known to lead to genetic isolation. An extreme case is the two populations of <u>Agrostis strolonifera</u> which failed to hybriduze even though they were just about one metre apart along a shope (Maton and Brandshaw, 1966). Thus the geographical distribution of the two genera (Map II) may be another possibility. <u>Anthericum</u> species are usually collected from the savanna zone while <u>Chlorowhylam</u> species do well in the southerm forest zone.

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It has been experimentally proved that production of flowers and fruits in Pairie tall grasses reduced when the plants were protected from grazing, nowing, and/or burning (Weaver and Fitzpatrick, 1934; Weaver and Rowland, 1952; Kelting, 1954; Kucera and Koelling, 1964; Elroy and Parenti, 1978). Though mechanical removal of the Pairie grass litter by clipping or mowing usually resulted in increased numbers of flower culms and productivity (Curtis and Partch, 1950; Penfound, 1964; Hulbert, 1969; and Richards, 1969), mechanical removal of leaves from A. limosum, A. pterocaulon II and A. pubirhachis did not improve fruiting but flowering. Thus, the nonproduction of mature fruits in these species is probably due to their non-subjection to natural burning, since they were collected from savanna vegetation which is usually subjected to annual burning. Elroy and Parenti (1978) proved that the average soil temperature of burnt area in July was markedly higher than the area which was not burnt up till September. He recorded increased occurrence of anthesis and higher fruiting in the burnt area than the area which was not burnt. The non-successful crosses between Anthericum species and the <u>Chlorophytum</u> species may probably be due to the absence of burning of the Anthericum species. The burning may be necessary to increase their soil temperature and soil chemical redistribution which are possibly essential for their fruit developments. The suggestion supports the above observations in Pairie grasses and that of Oyewole (197%) in which he was able to induce flowering in a species of Urginea (i.e. U. gigantea) in Nigeria by burning. Burning of the plants was not done early enough, since one continuously expected fruit formation from the crosses and from the many flowers produced. However, some of the plants have been subjected to burning but their results are not yet available at this stage of writing.

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The failube of crosses between <u>C</u>: <u>caulescens</u>, <u>C</u>. <u>alismifolium</u>, <u>Chlorophytum</u> X (with 2n = 16) may be attributed to partial crossincompatibility as observed by Prentice (1978) in <u>Silene</u> section <u>Blisanthe</u> (Caryophyllaceae). <u>C</u>. <u>alismifolium</u> is partially sterile. Only one or two fruits usually develop on each of its inflorescences out of more than ten flowers. The partial sterility is possibly due to its meiotic irregularity. On the other hand, <u>Chlorophytum</u> X is sterile. There was only a few fruits formed in this plant throughout the four years of study. The non-fruiting is due to its meiotic irregularities as highlighted in Chapter 5. Thus the unsuccessful crosses involving <u>Chlorophytum</u> X and <u>C</u>. <u>alismifolium</u> is due to their meiotic irregularities and low level, germination of their pollen grains (Fable 14).

There are zygotes and partial fruit formation in <u>C</u>. <u>alismifolium</u> X <u>C</u>. <u>caulescens</u> crosses. The fruits failed to mature. Small aborted seeds were found inside the fruits. This may be due to genie incompatibility of the two genomes as a result of gene rearrangements on the chromosomes curver gene mutations. In the reciprocal crosses, no zygote formation was observed as can be seen in Table 15. This observation agrees with the results reported by Philippi (1961), Harmey and Chow (1971), Harmey (1976), and Coffin & Harmey (1978) in their work on <u>Pelargonium X hortorum X P. domesticum</u>, and <u>P. hortorum X</u> <u>P. peltatum</u>, in which they observed zygote formationsonly in reciprocal crosses.

#### POLLEN GRAINS STUDY

Pollen grain viability tests had been commonly used to assess fertility in plants. The present study showed that high percentage of pollen grain viability does not necessarily mean high germination rate of pollen grains. As presented in Table 14, C. caulescens has high pollen grain viability of 93.8% while only 66% germinates. On the other hand, C. togoense has 98% of its pollen grains viable and 86.5% germination. This means that a seemingly viable pollen grain may not contain all the necessary genes for pollen grain germination and consequent police, tube developments. The occurrence of non-disjunction in meiotic cells of C. caulescens possibly accounts for its relatively low pollen grain germination. It is also evident from the same Table 14, that low pollen grain viability means low pollen grain germination. C. alismifolium has 68.5% pollen grain viability and only recorded a 2.5% germination. Likewise Chlorophytum X with a 36.8% pollen grain viability recorded 1.6% pollen grain germination. The Low germination in these plants can be associated with their meiotic irregularities discussed in chapter 5.

From this study, it can be concluded that high pollen viability does not necessarily mean high pollen germination as can be seen in Table 14. However, low pollen grain viability means low germination of pollen grains. It has also been observed that meiotic irregularities affect pollen grain viability and germination.

The failure of hybridization in this complex has been associated with low fertility or partial sterility of <u>A</u>. <u>limosum</u>, <u>A</u>. <u>pubirhachis</u>, <u>A</u>. <u>pterocaulon</u> II (in cultivation), <u>C</u>. <u>alismifolium</u> and <u>Chlorophytum</u> X with unbalanced zygote chromosome complements. These conditions have been reported to reduce the chances of successful crosses in plants (Harney and Chow, 1977).

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#### CHAPTER 7

#### SYNTHESIS

#### A. TAXONOMY:

i. Emphasis is now shifting from vegetative morphology as criterion for taxonomic classification to anatomical characters coupled with cytological and genetical data. Many recent works, including those of Aldridge, (1977); Pant and Basu (1977); Brandham and Cutler (1978); Esipova (1978) and Karlstrom (1978) employed anatomical approach in the solution of taxonomic problems.

Many criteria have been used in this line. Tissa (1977), working on Theaceae of Srilada, used the types of sclereid, basic types of stonata, sizes of adamial and abaxial cells along with the anticlinal cell walls as some of his criteria. Keng (1962) believed that the size and shape of epidermal cells and their anticlinal walls were enough identification characters in the genera and species of Theaceae. On the other hand, Metcalfe and Chalk (1950) employed the number of palisade layers in a leaf as criterion for delimiting genera and species of Theaceae. Cutler (1972) Cutler and Brandham, (1977) and Brandham & Cutler (1978) believed that the leaf surface pattern is a unique taxonomic character and thus they employed it in sorting out the problems in the tribe Aloineae. Others like Aldridge (1977) employed types of vessel elements and their length : breadth ratios in elucidating the evolution of Sonchus subgenus Dendrosonchus.

Khan <u>et</u>. <u>al</u> (1978) still employed vegetative morphological characters coupled with cytological studies in comfirming the taxonomic limit of some species of <u>Solanum</u>. Hence the use of vegetative morphological characters as taxonomic criteria must be supported by evidence (or data) from ecological, anatomical, cytological and genetical studies.

Anthericum L. and Chlorophytum Ker-Gawl. had been considered as separate genera mainly on their vegetative characters. The limitation of this metnod creates many problems. The extent of taxonomic disagreement among workers on them demonstrates the inadequacy of purely morphological characters in resolving their taxonomy. There is therefore need for confirmatory evidence from other areas of the biology of the complex.

Statistical analyses of the vegetative morphology, anatomy, ocology explored and soil multiplent requirements 'overlaps: in the two genera. These confirm the closeness of the taxa in the complex. However, in spite of their close similarities, some objective and crucial characters have been highlighted as presented in Table 16. Wilkins (1968) is of the opinion that a good taxonomic character must be highly correlated with other characters, but Burtt (1964) maintained that <u>Astragelus</u> and <u>Oxtropus</u> were separated by a single character which did not correlate with anything else. Thus a separation of two genera, based on at least fourteen correlated Khan <u>et</u>. <u>al</u> (1978) still employed vegetative morphological characters coupled with cytological studies in comfirming the taxonomic limit of some species of <u>Solanum</u>. Hence the use of vegetative morphological characters as taxonomic criteria must be supported by evidence (or data) from ecological, anatomical, cytological and genetical studies.

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With these new taxonomic criteria, it will be necessary to subject the intermediate species - <u>C</u>. <u>caulescens</u>, <u>Chlorophytum</u> X, <u>C</u>. <u>laxum</u> and <u>C</u>. <u>alismifolium</u> - to taxonomic analysis. A definite character is quantified as a whole, while an intermediate character is halved and distributed between the two genera. With this method, it is easy to determine the position of these taxa as can be seen in Table 17.

<u>C. caulescens</u> is hundred percent <u>Chlorophytum</u> and in agreement with the claim of Marais and <u>Refly</u> (1978).

<u>Chlorophytum</u> X is 71.4% <u>Anthericum</u> and 28.6% <u>Chlorophytum</u>. The leaf surface pattern of this taxon and its meiotic irregularities show that <u>Chlorophytum</u> X is close to one of the ancestral parental stocks in the complex. Hence it is suggested that <u>Chlorophytum</u> X, with 2n = 16 be considered as an old <u>Anthericum</u> species.

C. alismifolium, on the basis of these selected criteria, is 25.0% Anthericum and 75.0% Chiorophytum. This is expected because of its norphologically intermediate position. The leaf surface pattern is primitive. The irregularity in meiotic chromosome behaviour confirms that <u>C. alismifolium</u> is possibly close to the ancestral parent which gave rise to the <u>Chlorophytum</u> species with basic number 7.

CHARACTERS	AN PHERICUAL	CHLOROPH YTUM	INPERILDIATE LP.CLES
LEAF INDEX	> 20 Chlorophytum :	<ul> <li>≤ 10</li> <li><u>aubpetiol1tum</u></li> <li><u>C. cnuloscens</u></li> </ul>	>10 < 20 * <u>C. laxon</u> <u>C. bequeetii</u> * <u>C. blepharophyll</u>
VEIN SPACING INTERVAL (VSI)	≤0.5mm <u>C. laxun</u> Chlorophytum X	< 1.0mm C. caulescens	> 0.5mm < 1.0mm
SHAPE OF PSEUDOSTEM	Flattened Chlorophytum X C. alismifolium C. laxum	Circular <u>C. caulescens</u>	
LEAF SHAPE	Linear	Lanceolate C. caulescens	Chlorophytum X C. laxum
SHAPE OF PEDUNCLE	Flattened and Angular	Circular C. <u>Caulescons</u>	
ANTHER SIZE	≥ 2.5mm	2.5mm C. cauloscens	
% ANTHER : FILA- MENT RATIO	> 50% Chlorophytum X	<50% <u>C. caulescens</u>	
TEPAI DISPOSI- FION	Spreads out at full anthesis <u>Chlorophytum</u> X <u>C. laxum</u> <u>C. alisaifolium</u>	Bends backward at full Anthesis. <u>C. caulescens</u>	<u>C. toqoense</u>
WIDTH OF NOIGH- BOURING CRLLS	40 microns Chlorophytum X	2 40 microns <u>C. cauloscens</u>	
INDEX OF DEICH- BOURLING CELLS	> 5	C. crulescens	
COF NEIGHBOURING CELL WIDTH: STOMA WIDTH	Chlorophytum X	Z 50% C. chulescens	
% INDEX OF NEIGH- BOURING CELL:INDEX OF STOMA		25. C. cauloscens	
LEAF SURFACE PATTERN	Elongated paral- lelogram in shape	Rhombic shape <u>C. caulescens</u>	Chlorophytum X C. alismifolium C. laxun
LEAF MARGIN	Epidermal cells surround mesophyl cells. <u>Chlorophytum</u> X <u>C. alismifoliu</u>		

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	TABLE 16	
CHARACTERS	SEPAR FING SPHERICUH	AD CHLOROPATION

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# TABLE 17

# STATISTICAL ANALYSIS OF THE INTERMEDIATE SPECIES AND C. caulescens (A. caulescens)

		ulescens ulescens)	<u>c</u> . x	4 # 15 	<u>C. alismi</u>	folium	C. la:	X116
CHARACTERS	AN THER CUM	CHLORO PH YTUM	ANTHERICUM	CHLOROPH YFUM	ANTHERICUM	CHLOROPH YTUM	ANTHERICUM	CHLOROPH YTUM
LEAF INDEX	-	1.0	1.0	AND AND	ging .	1.0	0.5	0.5
VEIN SPACING INTERVAL (VSI)	-	1.0	1.0	-		1.0	1.0	6177 .
SHAPE OF PSEUDOSTEM	-	1.0	1.0	-	1.0	-	1.0	-
LEAF SHAPE	-	1.0	0.5	0.5	-	1.0	0.5	0.5
SHAPE OF PEDUNCLE	-	1.0	-	1.0	-	1.0	-	1.0
SIZE OF ANTHER	- (	1.0	-	1.0	-	1.0	-	1.0
% OF ANTHER : FILAMENT RATIO	-	1.0	1.0	-	-	1.0	-	1.0
PEPAL DISPOSITION	-	1.0	1.0	-	1.0	-	1.0	-
WIDTH OF NEIGHBOURING CELLS	-	1.0	1.0	-	-	1.0	-	1.0
INDEX OF NEIGHBOURING CELLS	-	1.0	-	1.0	-	1.0	-	1.0
% OF NEICHBOURING CELL WIDTH : STOMA WIDTH	-	1.0	1.0	-	-	1.0	-	1.0
% OF INDEX OF NEIGHBOURING CELL : INDEX OF STOMA	-	1.0	1.0	-	-	1.0	-	1.0
LEAF SURFACE PATTERN	-	1.0	0.5	0.5	0.5	0.5	0,5	0.5
LEAF MARGIN	-	1.0	1.0	-	1.0		0.5	0.5
TOTAL	-	14	10.0	4.0	3.5	10.5	5.0	9.0
% OF AFFINITY	0%	100%	71.4	28,6	25	75	35.7	64.3

C.

The intermediate position of C. laxum (2n = 14) moves more to Chlorophytum side. It is 35.7% Anthericum and 64.3% Chlorophytum . It may be that C. larum was an old bydrid of Anthericum and Chlorophytum, which is now tending towards Chlorophytum (1, e introgressive hybridization). This phenomenon had been reported in maize X Tripsacum hybrid by Harlan and De Wet (1977). It may be that C. laxum with 2n = 16. (i.e. the Indian material) is more of Anthericum or Chlorophytum. One would have liked to examine this material, but all efforts to get seeds from India failed. However, Naik (1977) reported that C. laxum with 2n = 16 had a pair of lagging chromosomes as has been observed in C. alismifolium. It is therefore speculated that C. laxum, 2n = 16, might have been one of the parlies hybrids resulting from the two closely related ancestral parents. From this taxon, some Anthericum and Chlorophytum species might have evolved.

## ii. STATUS OF THE TWO FORMS OF C. STENOPETALUM Bak.

The similarity in vegetative attributes of <u>C</u>. <u>stenopetalum</u> I and <u>C</u>. <u>stenopetalum</u> II needs further analysis. The morphology of the two taxa is very similar except that <u>C</u>. <u>stenopetalum</u> II, collected from the Northern part of the country, has more leaves than <u>C</u>. <u>stenopetalum</u> I, collected from the southern part of the country. A careful uprooting of the two species reveals a marked difference in their root system. While <u>C</u>. <u>stenopetalum</u> I usually has one mediam or distally located root tuber per root, <u>C</u>. <u>stenopetalum</u> II has ten or more root tubers which are alternately arranged along each root length. This distinction is found to be stable.

The morphological difference has been correlated with evidence from cytological studies. It will therefore be reasonable to separate the two taxa because:

i. genotypic differences have been established between the two taxa,

and ii. the phenotypic characters are manifestations of their genetic constitutions.

Borril (1961) employed cytological data to elevate two forms of <u>Dactylis glomerata</u> to species level. Khan, <u>et. al</u> (1978) confirmed the separation of <u>Solanum nigrum</u> by cytological studies and statistical comparison of morphological characters. Thus the separation of the two taxa on the bases of (i) the different patterns of root system; (ii) difference of their karyo-morphology (see Table 12) and (iii) their genetical difference as shown by their chiasma formation (as presented in Table 13 and discussed in Chapter 5) is justified.

It is hereby suggested that <u>C</u>. <u>stenopetalum</u> I should still be known as <u>C</u>. <u>stenopetalum</u> Bak. <u>C</u>. <u>stenopetalum</u> II, with ten or more

2

leaves and with many root tubers, should be separated from it and given a different identity. Meanwhile, both taxa can be identified easily, using the following key:

Leaves few, less than 10,

Root tubers usually one, median to distal on root, of wet and shaded habitat in rain forest.

..... C. stenopetalum I

Leaves many, usually 10 or more, Root tubers usually many and alternately arranged along the length of each root, of open savanna grassland habitat.

iii. TAXCNOMIC KEY.

of the fourteen separating criteria listed in Table 16, seven characters are easily observable, and decisive and thus considered to be of great taxonomic importance to any herbarium

..... C. sp. nov.

worker or taxonomist. Using these criteria, the two genera can be separated as follows:

Leaf linear, usually with leaf index > 20,

Veins closely spaced usually < 0.5mm, apart; Leaf surface cells usually elongated

parallelogram with vertical anticlinal walls, Epidermal cells surround the mesophyll

cells at leaf margin, Pseudostem, more or less, flattened, Tepals open to base and rotate, Peduncle more or less flattened.

Anthericun

Leaf lanceolate usually with leaf index < 10, Veins widely spaced usually > 1.0mm apart, Leaf surface cells more or less rhombic in shape with horizontal anticlinal walls; Epidermal cells taper at leaf margin

forming one or two cell layers, Pseudostem more or less circular; Tepals not open to base but reflexed; Peduncle more or less circular.

come Chlorophytum

Using this key and analysis of the intermediates in Table 17 with other facts earlier enunciated, it is suggested that:

- i. The transfer of <u>C</u>. <u>caulescens</u> (Bak.) Marais & Reilly from <u>Anthericum</u> to <u>Chlorophytum</u> genus by Marais and Reilly (1978) is supported by this work. Marais and Reilly (1978) based their separation mainly on the seed and capsule characteristics. The 100% justification of <u>C</u>. <u>caulescens</u> as <u>Chlorophytum</u> strengthens the new criteria that I have put forth. In view of the fact that most specimens do not usually have fruits at the time of their collections, it is therefore better to use the criteria that are hereby proposed for the separation of the two genera.
- ii. C. alismifolium and C. laxum should still remain in Chlorophytum genus.
- iii.

The two forms of <u>C</u>. <u>stenopetalum</u> complex need to be separated into two taxa at species level. a. <u>C. stenopetalum</u> I can still be regarded as C. stenopetalum Bak.

b. C. stenopetalum II is a different species from C. stenopetalum Bak. It can be regarded as a new species for the moment until. its identity is confirmed as being new or otherwise at Kew. be transferred from Chlorophytum genus to

#### Anthericum genus.

Meanwhile, taxonomic nomenclature of the new taxa would be stayed until their fifferent identities are confirmed from Kew.

### B. EVOLUTION.

## i. CHROMOSOME EVOLUTION:

It has been suggested that the possible parental stock of the two genera must have had  $\mathbf{x} = 4$  basic chromosome number. It has also been speculated that natural hybridization occurred between two closely related taxa. Hybridization was followed by chromosome doubling, which resulted in 2n = 16, to ensure balanced genetic system. It has also been argued that n = 7 possibly evolved from n = 8 which is primitive. Basic number 8 is primitive because C. alismifolium and C.  $\mathbf{X}$  with n = 8 are perennial while other diploid Chlorophytum species with n = 7 are annual. Perennial habit is primitive while annual habit is derived (Stebbins, 1950; Baker, 1965). The perennial habit of these taxa (tetraploids with  $x \rightarrow 4$ ) is to enable them to evolve genetic systems that will confer sexual reproduction on them (Olorode, 1973). The partial sterility and meiotic irregularities observed in these taxa support the above idea.

Evolution does not usually take one single course. During the early hybridization and chromosome doubling meiotic irregularities would occur. During this process, some chromosome pair(s) might have been lost through fusion or elimination. On the other hand, supernumerary division might have occurred to give rise to spores having n = 4, n = 6 and n = 3. Supernumerary division has been reported in <u>C</u>. elatum (2n = 28) by Storey (1968) who observed hemi-haploid with n = 7 and sesquihaploid with n = 21. Koul (1970) also reported different chromosome numbers in the spore cells of <u>C</u>. elatum. The taxa in this complex easily propagate vegetatively. Hence any mis division in both somatic and meiotic cells can give rise to a different individual. If such individuals were able to establish themselves, plants with different basic number would emerge.

Crosses between these plants possibly account for their speciation, while the suggestion that they all evolved from the same parental stock ensures their similarities. Thus species with 2n = 14 might have arisen from two possible lines. The first possibility has been discussed in Chapter 5, that is, by fusion of two pairs of chromosomes from 2n = 16. The second possibility is that a cross occurred between one of the parental stock with x = 4 and a tetraploid hybrid with n = 8. This gave rise to 2n = 12. A second cross then occurred between n = 6 and n = 8 and give rise to 2n = 14. The formation of three associated quadrivalents and a bivalent in <u>C</u>. inornatum supports this idea. The three bivalents from n = 6 parent associated with three of the four bivalents from n = 8 parent since they evolved from a common ancestral stock.

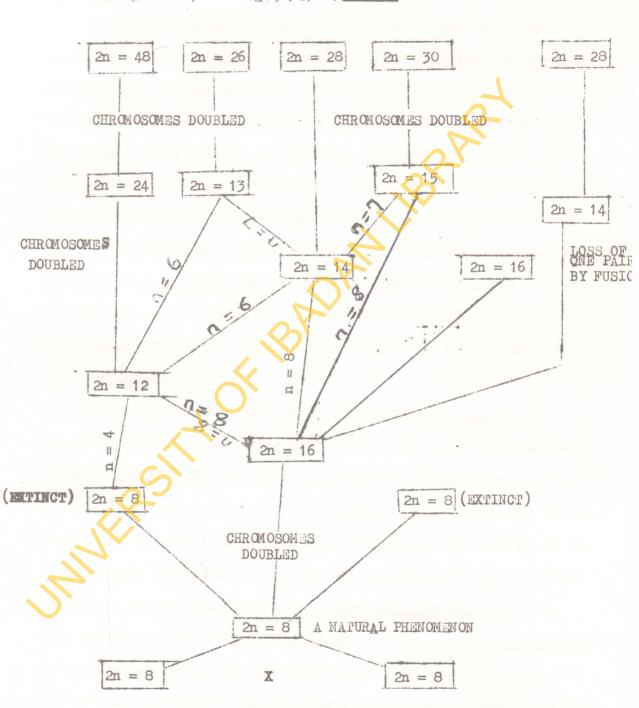
Some taxa in this complex have been reported to have different chromosome basic numbers from n = 8, n = 6 and n = 7. Sarkar <u>et. al</u> (1973) reported <u>C</u>. <u>khasianum</u> and <u>A</u>. <u>ramosum</u> as having 2n = 30. In like manner, Strandhede (1963) reported 2n = 60 for <u>A</u>. <u>liliago</u> and 2n = 30 for <u>A</u>. <u>ramosum</u>. These plants might have evolved from crosses of n = 8 and n = 7 resulting in 2n = 15. Due to possible irregularities in pairing, the chromosome doubled to give tetraploid with 2n = 30.

Naik (1975) in his proposal for the evolution of species of <u>Chlorophytum in India recorded C. nepalense</u> and <u>C. khasianum</u> as having 2n = 26. He was of the opinion that species with 2n = 28, lost a pair of chromosomes and gave rise to 2n = 26. The other possibility is that a cross occurred between n = 6 and n = 7 and gave rise to 2n = 13 which doubled and resulted in 2n = 26.

Stransburger (1888) reported <u>C</u>. comosum and <u>C</u>. sternbergianum as having 2n = 24. A species of <u>Anthericum</u> (? <u>A. pterocanlon</u> I) worked upon by the author in 1977 had 2n = 24 (Plate VIIId). It was however lost under cultivation. These taxa might have possibly evolved from the old plants with 2n = 12 by chromosome doubling. Species having 2n = 24 should better be regarded as tetraploids with x = 6 rather than as triploids with x = 8.

This study has revealed that the chromosome evolution in <u>Anthericum</u> and <u>Chlorophytum</u> complex is multi-directional after the initial hybridization of their ancestral parents and the doubling of the hybrids chromosome complements. The secondary basic chromosome number is therefore x = 8 from which x = 6 and x = 7 are possibly derived. The combinations of these basic numbers coupled with fusion and/or elimination of different chromosome pair(s), translocations of chromosomes and gene inversions, rearrangements of genes and possibly reciprocal crosses, gave rise to the present different chromosome/diploid levels and differences in the present species. The proposition that they all evolved from the same common ancestral stock ensures their similarities.

The proposed trend of chromosome evolution in the <u>Chlorophytum</u> and <u>Anthericum</u> complex is presented in Figure IV.



PROPOSED TREND OF CHROMOSOME EVOLUTION IN CHLOROPHYTUM - ANTHERICUM COMPLEX

FIG. IV

R

#### ii. SPECIATION

Evidence of aneuploid alteration of basic number has been carefully studied in <u>Crevis</u> and its relatives - <u>Lactuca</u>, <u>Prenanthes</u> and <u>Hieracium</u> (Stebbins, 1940). It has also been concluded that the most primitive species have the highest basic numbers (Babcock and Cameron, 1934; Babcock <u>et</u>. <u>al</u>, 1937; Babcock, 1942, Babcock & Jenkins, 1943). It is believed that aneuploid alteration had occurred in the ancestral hybrid(s) of this complex and that n = 8is the primitive basic number. A study of the leaf surface anatomical patterns of <u>C</u>. <u>alismifolium</u> and <u>Chlorophytum</u> X, both with n = 8 shows that they are more primitive than their relatives with n = 7(see Plates III a & b). Also their perennial habit is evidence of their primitiveness.

If the leaf surface anatomical pattern in <u>C</u>. <u>alismifolium</u> (Plate IIIa) is taken as being typical of one of the ancestral parent A while that of <u>C</u>. I (Plate IIIt) is typical of the other parent B, One would see that the leaf surface patterns in the other taxa of the complex are intermediates. Thus they are possibly the hybrids of the two patterns. Hybridization of the two patterns will give a pattern which will be much like that of <u>C</u>. <u>blepharophyllum</u> (Appendix <sup>5a</sup>.). This resembles that of <u>C</u>. <u>alismifolium</u> more than that of <u>Chlorophytum</u> X. It is thus speculated that the cross was between plant A and plant B and that the patterns in <u>Chlorophytum</u> species are modifications of this type. The leaf surface pattern in ?A. <u>pterocaulon</u> I tends more to that of  $\underline{C}$ .  $\underline{I}$  . It is thus speculated that the cross in this case was between parent B X parent A and that the pattern in Anthericum species evolved from this cross.

Choudhuri (1944: 1948: 1972) reported differences in the phenotypic characters of reciprocal crosses in potato and Solanum melongena. Adeyemi (1975) also observed differences in the lobing of leaf and shapes of fruit in reciprocal crosses of Solanum melongena varieties. Von Wettstein (1928) demonstrated the cytoplasmic hereditary potentiality in interspecific and intergeneric crosses in Bryophytes. Swanson (1965) is modest in his own contribution and suggested that the cytoplasm should be regarded as the cellular clay that is moulded into shape by the nucleus. He concluded that the quality of the clay (cytoplasm) must be considered as gene-determined although modified by the environment. Hence, the end product - the cell - achieves its final delineation not only by the moulding process but also by the sum total of the ingredients that enter into its makeup. This agrees with the opinion of Willmar (1969) who stated that the phenotypic manifestation of any taxon is a combined interplay of its genome (i.e. its set of chromosomes and other genetic materials), the hegemon (i.e. the part of the genome which regulates the cell activity at any one time), the econome (i.e. the particular pattern of the cell's cytoplasmic and nuclear membrane and its organelles), and the

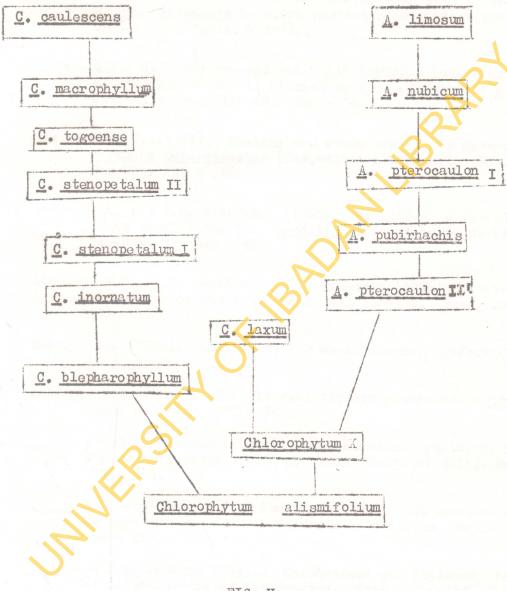
agoranome (i.e. the organised environment of the cell with its neighbours and the surrounding tissue).

In view of the above facts, and knowing the important effects of the cytoplasmic inheritance including the cytogenes on hybrids' phenotypic characters, I believe that <u>Anthericum</u> and <u>Chlorophytum</u>, which had the same ancestral parents evolved from reciprocal crosses. This was followed by parallel evolution.

Considering the possible evolution of the chromosomes (Fig. IV), the vegetative morphology and the leaf surface patterns, the possible evolution of Nigerian species of <u>Anthericum</u> and <u>Chlorophytum</u> species is proposed in Figure V.

The ancestral parents are unknown and might have been eliminated by natural selection. The leaf surface patterns of <u>C</u>. <u>alismifolium</u> and <u>Chlorophytum X</u> are the possible preserved ancestral patterns, hence they are placed low in the evolutionary scale. It has been argued that <u>Anthericum</u> species possibly evolved from <u>Chlorophytum</u> X parents while <u>Chlorophytum</u> species are derived from old

C. alismifolium stock.



PROPOSED EVOLUTION OF SOME NIGERIAN SPECIES OF ANTHERICUM AND CHLOROPHYTUM,

FIG. V

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#### APPENDIX 1

SPECIES	AREAS OF COLLECTION				
<u>A. dalzielii</u> Huth. ex Hepper	Bauchi Plateau, Vom, Jos, Bukuru, Tangale-Waya in Banchi Privince				
<u>A. limosum</u> Bak:	Naraguta F.R., Jos, Borgu Game Reserve Ibuya, Upper Ogun Games Research, Panshin,				
A. <u>nigericum</u> Hepper	Zaria, Birnin Gwari; Vom, Yola. Zaria; Anara F.R.; Zaria Province, Jos Plateau.				
<u>A. nubicum</u> Bak.	Kontagora, Minna, Vom, Naraguta Wana; Yola; Vogel Peak; Zaria, Birnin Gwari,				
<u>A. pterocaulon</u> Welw. ex Bak.	Old Oyo F.R., Ilorin, Borgu G.R., New Bussa, Bauchi Plateau. Anara F.R. Zaria Province; Zaria to Samaru; Wana, Keana, Benue District, Yola				
A. pubirhachis Bak.	Sardauna, Gombi, Vogel Peak Jos, Buruku to Gimi. Nupe; Zaria to Jos; Upper Ogun, Iseyin; Igbeti, Zaria, Kaduna to Jos, Zaria to Kaduna.				

#### DISTRIBUTION CHART OF ANTHERICUM AND CHIOROPHYTUM IN NIGERIA

SPECIES	AREAS OF COLLECTION		
A. <u>subpetiolatum</u> Bak.	Abuja; Jos Plateau; Bukure to Hepham, Birni		
	Kwaya District; Bornu; Mambilla Plateau,		
	Bornu to Kwaya Kaduna to Jos		
A. speciosum	Zaria.		
A. <u>uyuiense</u> Rendle	Liruwen-Kano Hill; Samaru, Giwa Neill's		
	Valley, Jos; Naraguta F.R.; Bornu, Tangale-		
	Waja, Bauchi Province, Offa Panshanu Pass,		
	Bauchi, Birnin Gwari.		
A. Zenkeri Engl.	Vogel Peak, Sardauna Province.		
C. <u>caulescens</u> (Bak)	Nupe Florin to Jebba, Ibadan, Kaduna,		
(A. caulescens (Bak)	Kagara, Ado, Iseyih.		
C. alismifolium Bak.	Ekeji-Ipetu F.R., Ilesha District. Omo F.R.		
	Etioni, to Atijere, Ijebu-Ode, Akure F.R.		
S	Ondo; Obubra Ore; Idanre.		
. <u>aureum</u> Engl.	Zomi (N. of Igeyin); Upper Ogun Cattle R.		
	Oli River; Borgu G.R.		
C. blepharophyllum	Nupe, Barter Mokwa; Anara F.R., Zaria;		
schweinf. ex Bak	Afaka F.R. Jos; Zelau, Banchi District;		
	Jebba, Gimi; Fadau Ayu; Soba between		
	Zaria-Jos; Gembu Mambilla, Kishi, Jos;		

SPECIES	AREAS OF COLLECTION					
	New Bussa, Bida; Kan Gimi; Igbeti; Wawa;					
	Oturkpo; Kontagora; Ogoja, Ilorin, Zaranda					
	Mt (Bauchi); Oyo.					
C. gallabatebse	Jebba; Sanga E.R., Tede, Birnin Gwari;					
Schweinf. ex Bak.	Shika in Samaru, Biological Garden,					
	University of Ife; Bornu.					
C. geophilum	Boronu, Niger Province; Naraguta; Samaru					
Peter ex v. Poelln.	to Shika, Zaria; Gwari.					
C. inornatum Ker-Gawl	Idanre F.R., Ondo District,; Ibadan to Oyo;					
	Sapoba, Benin Dist.; Oyo; Unife Campus;					
Supposed of the second second	Dumbi Wood, Zaria to Kaduna.					
C. laxum R. Br.	Nupe Barter; Zungeru; Dogondaji Kurmin,					
	Jemaa Dist.; Oban; Plateau Dist.; Serti;					
WERS'	Gongola; Upper Ogun; Zaria, Niger Quara					
	Falls (Bonu F.R.) Oyo; Ogboro Hill near					
	Kishi; Soba.					
C. macrophyllum (A. Rich)	Olokemeji F.R.; Ibadan F.R.; Omo F.R.					
Aschers.	Atikiriji; Afi River F.R. Oyo to Iseyin Road					
	Onigambari, Ijebu-Ode Road; Wushishi;					
	Tungna - Biba Road, Wamba, Akwanga Naraguta;					

SPECIES	AREAS OF COLLECTION
	Abeokuta; B. Gwari,; Zaria Badeggi-Lapai;
	Gimi; Idanre Hill, Ondo ; Kabba, Ibadan,
	Ife.
C. orchidastrum	Olokemeji, Cross River ; Dekina, Benue
Lindt.	District, Calaba, Obubra; Abuja; Ibadan,
	University of Ife Biological Garden;
	Upper Ogun F.R.
C. polystachys Bak.	Yankari:
C. pusillum Schweinf.	Panshanu; Kilba Hills, near Yola; and
ex Bak.	Pankshin.
C. sparsiflorum Bak.	Ikwette, Obudu District; Oban F.R. Calabar;
S Abrances of	Awi to Ikou Road; Momgo: to Kumba; Ogoja.
C. stenopetalum Bak.	Nupe Narter, Zungeru, Samaru, Anara F.R.,
	Zaria; Naraguta; Zaria-Funtua Road, Birnin
S	Gwari; Badeggi-Lapai (Niger State); Lapai;
	Igboho to Kisi Rd.; Mokwa; Kaduna; and Bukur
<u>C. togoense</u> Engl.	Wawa; Gindiri, Jos Plateau; Abinsi; Ikoyi Wo
	Lagos; Ibada North F.R.; Ewohimi to Idumujun, Ishan Dist.; Olokemeji, Upper Ogun R. (Oyo-
	Iseyin); Awka, Onitsha Prov., Oban; Benin;
	Pankshin, Nsukka; Ijaiye, Ibadan; Ikare; Ogo,
	Unife; Fasakai, Plateau; R. Lere near
	Gindiri; Ondo, Mokwa-Kainji Road.
C. tuberosum (Roxb.) Bak.	Katagum; Filiya, Bauchi Prov.; Biu, Bornu
	Province, Yola: Panyam.

### APPENDIX 2

# SOIL SAMPLES' LOCATIONS

af an mean at the state of the state at the
LOCATION
About 10 km. to the North of Jebba Bridge, lying
few metres on the right side of the road from Jebba.
Onigambari, on Ibadan-Ijebu-Ode Road, opposite
the Forestry Research Institute of Nigeria
Station, gate.
Along a footpath on the outskirts of Jebba
from Ilorin.
2 km. to Seba on Zaria-Jos New Road from Zaria.
Outskirts of Mokwa along the A.B.U. Research
station road; around the Guest House area.
Azadirachta indica plantation. 24 km to Kaduna
along Ibadan - Kaduna Road.
Outskirt of Iseyin, about 2 km. to the town
from Oyo.
Cutskirts of Ore, about 3 km. to the right on
the New Benin-Lagos Road.
Botanical Garden, U.I. Ibadan on a slope of a
rock just few metres from the main garden road. Around the railway crossing in Jos. A few metres
on the left on Jos-Bukuru Road.
Outskirts of Jos on weathered rock by the
right from Zaria.
f km. to Jengre on Zaria-Jos Road, from Zaria end, a few metres on the left.

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# APPENDIX 3

### MECHANICAL SOIL ANALYSIS

	SPECIES	% FIED WATER	% SOIL	% OF	% OF	% OF
		HOLDING CAPACITY	WATER	SAND	SILT	CLAY
C.	blepharophyllum	3.2	1.37	88.85	2.63	8.52
<u>c</u> .	macrophyllum	11.7	3.54	55.43	31.72	12.85
C.	inornaium	16.8	5.10	39.95	34.34	25.71
<u>e</u> .	laxum	8.5	1.14	84.83	6.67	8.50
C.	stenopetalum I	12.7	1.96	86.87	4.65	8.48
<u>C</u> .	stenopetalum II	4.3	1.27	86.83	4.66	8.5
c.	togoense	11.5	2.44	41.57	47.77	10.66
C.	alismifolium	14.8	2.82	55.76	23.25	20.99
<u>c</u> .	caulescens	13.0	2.14	70.37	12.87	16.76
<u>A</u> .	limosum	9.7	1.83	70.46	20.98	8.56
<u>A</u> .	pterocaulon	11.8	2.13	74.46	16.96	8.58
Δ.	putrirhachis	8.6	1.70	62.37	25.07	12.61