

Full Length Research Paper

# Seasonality of the mycoflora of the crown disease complex of the vegetative organs of the grapevine *Vitis vinifera* cvar Anap-e-Shahe

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The mycoflora associated with lesions on the vegetative organs of Anap-e-Shahe cultivar of grapevine in Zaria were monitored for 3 years. The symptoms varied with the seasons but were the same over the years studied. The mycoflora detected on the lesions were consistent with the symptoms. Except for pruning interruptions, *Epicoccum* sp., *Penicillium* sp., *Aspergillus* sp. and *Curvularia* sp. was regularly detected on lesions of all growth seasons. *Alternaria* sp., *Drechslera* sp. and *Pestalotiopsis* aff. *uvicola* occurred only on lesions of the rainy season growths. *Phoma* aff. *multirostrata*, *Colletotrichum gloeosporioides*, *Phomopsis* sp. and *Phoma* sp. were the dominant group on lesions of rainy season growth and were detected from the beginning of the rains in May until the close of the rains in October, which corresponded with the prevailing period of anthracnose, tip die back, commencement of canker development and blight. The occurrence of *Plasmopara viticola* commenced during the downy mildew epiphytotic late in July or early August, a time which corresponded with high relative humidity and a drop in maximum temperature at the peak of the rains. *P. viticola* and *Curvularia* sp. with other regularly occurring fungi detected from the late rainy season persisted into the dry season. The seasonal variability in the mycoflora of the crown disease complex of grapevine provided information which could be exploited in disease diagnosis and management.

**Key words:** Mycoflora, Anap-e-Shahe, viticulture, disease epidemiology.

## INTRODUCTION

The grape berry (*Vitis* spp.) is a renowned cash crop (Nail and Howell, 2005). It has a high dietary fiber and when fermented into wine reduces the risk of, and death from, cardiovascular disease (Stockly and Hoj, 2005; Diaz-Rubis and Saura-Calixto, 2006).

Millions of dollars are lost annually due to fungal infections of grapevines which results in low crop yield, compositional and sensory changes as well as increased levels of haze-forming pathogenesis related proteins in grape juice and wine (Girbau et al., 2004; Stummer et al.,

2005). In view of the above, solutions have been proffered which include multiline cultivars and cultivar mixtures for disease management, increasing the natural antagonistic population present in vineyards to prevent fungal disease and the use of fungicides and clay treatment to control fungal microflora (Mundt, 2002; Robusto et al., 2006; Sholberg et al., 2006).

Viticultural research began in Ahmadu Bello University, Zaria in the 1980s with the introduction of a high yielding cultivar known as Anap-e-shahe (Majanbu, 1995). However, the constraints to cultivation in Northern Nigeria include infectious diseases, pests and water scarcity (Erinle, 1987; Khan, 1987; Kalu, 1997; Roby et al., 2004; Pellegino et al., 2005).

Despite advances in laboratory molecular-based detection techniques, a strong case is presented for the conti-

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**Table 1.** Disease symptoms of the growth stages of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe, pruned mid March to late September/mid October (rainy season growth).

Major growth stage	Growth stage description under healthy conditions	Time in the Year	Observed symptoms
Bud development Bud development	<ul style="list-style-type: none"> <li>The dormant bud has scales more or less closed.</li> <li>The dry season bud swells, then turns into a woolly bud in which the brown wool is visible.</li> <li>The bud bursts and the green shoot tips are just visible</li> </ul>	3 <sup>rd</sup> week in March	None
Shoot and inflorescence development	<ul style="list-style-type: none"> <li>The first leaf unfolds and spreads away from the shoot.</li> <li>12 leaves unfold sequentially, inflorescence well developed and single flowers separate.</li> <li>14 leaves separated, cap colour fading from green.</li> </ul>	4 <sup>th</sup> week in March – 1 <sup>st</sup> week in April  2 <sup>nd</sup> week in April  2 <sup>nd</sup> – 4 <sup>th</sup> week in April	None  Leaf spots, blotches and shot holes.  Leaf spots, blotches and shot holes. Spots and blotches on petioles.
Flowering	<ul style="list-style-type: none"> <li>The first flower caps falls away from the receptacle.</li> <li>10, 30, 50, 80% of the flower caps fall away.</li> <li>Cap-fall complete.</li> </ul>	1 <sup>st</sup> – 2 <sup>nd</sup> week in May  3 <sup>rd</sup> week in May	Symptoms evident on immature organs. Leaf spots, blotches and shot holes. Spots and blotches on petioles. Tip dieback.
Berry development	<ul style="list-style-type: none"> <li>Fruit set: the berries begin to swell.</li> <li>Berries are peppercorn size.</li> <li>Berries are pea-sized (7 mm diameter).</li> <li>Berries continue to expand until a lag phase.</li> </ul>	2 <sup>nd</sup> – 3 <sup>rd</sup> week in May 4 <sup>th</sup> week in May 3 <sup>rd</sup> week in June	Symptoms evident on immature organs (safe for berries). Leaf spots, blotches and shot holes. Lesions girdling petioles. Leaf epinasty. Tip dieback. Spots, blotches, scab on canes (early canker) (Figures 1 and 2)
Berry ripening	<ul style="list-style-type: none"> <li>Berries soften and levels of total soluble solids increase.</li> <li>Change in colour and berries expand again.</li> </ul> <ul style="list-style-type: none"> <li>Berries ripe.</li> <li>Berries over-ripe.</li> </ul>	4 <sup>th</sup> week June – 2 <sup>nd</sup> week July          2 <sup>nd</sup> week July	Symptoms evident on immature and mature organs. Leaf blight. Spots, blotches, scab on canes (early canker), spurs and branches (Figure 3) Defoliation of dead leaves. Branch dieback. Early cankers on canes, spurs and branches. Peak of disease intensity. Lesions on berries. Lesions, rot with fungal fructification on berries
Senescence	<ul style="list-style-type: none"> <li>End of wood maturation</li> <li>Pruning</li> </ul>	August  3 <sup>rd</sup> week September – 2 <sup>nd</sup> week in October	Symptoms evident on immature and mature organs. Disease intensity apparently minimized with the production of new leaves in compensatory response to defoliation (Figure 4).

nued use of slower biological tests in programs requiring high levels of confidence in detection of pathogens (Rowhani et al., 2005). With reference to the above the aim of this work is to initialize the study of disease problems in vineyards in Northern Nigeria through disease characterization and identification.

## MATERIALS AND METHODS

The mycoflora associated with a susceptible variety, Anap-e-Shahe, was monitored on samples obtained from vineyards in Zaria (located between latitude  $11^{\circ} 11'$  and longitude  $7^{\circ} 38'$  and 686m above sea level in the Northern Guinea Savanna Zone). Three vineyard sites (in Area A and E of Ahmadu Bello University (ABU), and Federal College of Chemical and Leather Technology (CHELTEC) were selected for being established at same period (dry season of 1995) and having at least five fruiting vines of Anap-e-Shahe. The vineyards were visited for the disease assessment twice a month (at  $14 \pm 2$  days intervals) starting from bud burst after each pruning from March pruning of 1998 to March pruning of 2000 with pruning interruptions. The growing season of the grapevine (from bud breaking to pruning) were divided into six growth stages (G.S.) using the scale of Coombe, (1988a,b). The assessment was on samples obtained by randomly selecting one vine per site. Two (2) canes ( $2 \pm 0.5$  cm diameter thick) purposively selected for the occurrence of disease symptoms from the selected vine were removed and taken to the laboratory for examination. Segments excised from the periphery of lesions on the various organs of each branch were separated according to type of disease lesions, organs, branches, and sites.

The presence of fungi and/or bacteria on the vine segments was verified at the initial stage of assessment. Thereafter, sampling continued by direct examination of the segments using a hand lens and by agar plate method for isolation and identification of the micro-organisms. The dishes were incubated on laboratory benches under prevailing conditions (temperatures of  $25 \pm 2^{\circ}\text{C}$  and 12 h day light) for 48 h. The developing colonies were isolated and sub-cultured onto potato dextrose agar amended with streptomycin (PDA + S). The detected species were identified on the basis of microscopic examination of the fine morphological diagnostic features, the cultural characteristics and analysis of observed symptoms as described in literature (Ellis, 1966; Benoit and Mathus, 1970; Barnett and Hunter, 1972; Booth, 1977; Pico and Rodolfi, 2004; Hajjeh et al., 2005; Rowhani et al., 2005). Cultures of the fungi were matched with already identified colonies in the repository of collection of mycological cultures at the Crop Protection Department of ABU and Nigerian Institute for Oil Palm Production. The isolated fungi were stored as stock cultures on PDA + S slants in McCartney bottles at  $15^{\circ}\text{C}$  and transferred every two months for comparative purposes. Day-old-cultures and infected tissues were dispatched to CABI Bioscience UK Centre for confirmatory identification. Weather records were obtained from the Meteorological Unit of Soil Science Department, Institute of Agricultural Research (IAR), located in ABU campus in Samaru, one and a half (1.5) kilometers away from the furthest sample source.

At each sampling five segments were randomly selected from each type of disease lesion on two branches from the three separate vineyards. Thus for each type of disease lesion there were 30 segments. Counts of the number of times a species was detected on segments of each type of disease lesion were taken at each sampling. The frequency of occurrence of a species was determined by averaging the counts of the species from each of the four organs at each sampling. The relative frequencies of occurrence of the species for each month were calculated by averaging

the frequencies of the two (2) samplings of each month.



**Figure 1.** Leaf and cane lesions on rainy season growth of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe.

## RESULTS

### Characteristics of the disease complex

The growth stages and the disease symptoms of the crown disease complex were similar in the vineyards at all samplings and were similar for the years. The rainy season growth stages in relation to symptoms of the crown disease complex are summarized in Table 1 while those of the dry season are summarized in Table 2. The pictorial view of the various disease states are portrayed in Figures 1 - 6 which are referenced in Tables 1 and 2. The downy mildew epiphytic in conjunction with the pathogen was an observed distinctive symptom during the samplings. All other symptoms were necrotic and atypical of any specific disease, necrotic and hence lumped as necroses.

Disease symptoms on the rainy season growths commenced as necrosis of immature organs that were usually initiated as dark leaf spots which enlarged to blotches. The leaf infection progressed downwards as spots and blotches on petioles but as spots, blotches and scabs on canes. Blotches and scab on leaf, petioles and canes persisted until the influx of downy mildew at the peak of the rains towards the end of July to early August when maximum and minimum temperatures dropped. From the advent of downy mildew the progression of necrosis on leaves and petioles of all ages, on immature canes and spurs accelerated and subsequently resulted in death of the host organs. Death of extensive organs of the crown was evident as leaf blight with foliar epinasty (Figure 4).

Lesions on internodes of canes, spurs and branches enlarged to girdle off all distal portions resulting in cane, spur and branch dieback of areas above the lesions. The dead leaves fell off and in compensatory response new leaves were produced which apparently minimized the

**Table 2.** Disease symptoms of the growth stages of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe, pruned late September/mid October to mid March (dry season growth).

Major growth stage	Growth stage description under healthy conditions	Time in the year	Observed symptoms
Bud development	<ul style="list-style-type: none"> <li>The dormant bud has scales more or less closed.</li> <li>The wet season bud swells, then turns into a woolly bud in which the brown wool is visible</li> <li>The bud bursts and the green shoot tips are just visible.</li> </ul>	October	none
Shoot and inflorescence development	<ul style="list-style-type: none"> <li>The first leaf unfolds and spreads away from the shoot</li> <li>12 leaves unfold sequentially, inflorescence well developed and single flowers separate.</li> <li>14 leaves separated, cap colour fading from green.</li> </ul>	2 <sup>nd</sup> week in October  3 <sup>rd</sup> week in October  4 <sup>th</sup> week in October	None  Leaf spots, blotches and shot holes. Downy mildew on leaves  Leaf spots, blotches and shot holes. Downy mildew on leaves Spots and blotches on petioles.
Berry ripening	<ul style="list-style-type: none"> <li>Berries soften and levels of total soluble solids increase.</li> <li>Change in colour and berries expand again.</li> </ul>	2 <sup>nd</sup> – 3 <sup>rd</sup> week in December  1 <sup>st</sup> week in January 1 <sup>st</sup> week in March	Downy mildew blotches and shot holes on leaves. Leaf blight; and cane, spur and branch dieback resulting from girdling lesions on internodes of canes, spurs and branches (Figure 6). Lesions on berries. Lesions, rot and fungal mycelia and fructification on berries.
Senescence	<ul style="list-style-type: none"> <li>End of wood maturation</li> <li>Pruning</li> </ul>	2 <sup>nd</sup> week March	Leaf blight; and cane, spur and branch dieback.



**Figure 2.** Tip dieback on rainy season growth of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe.



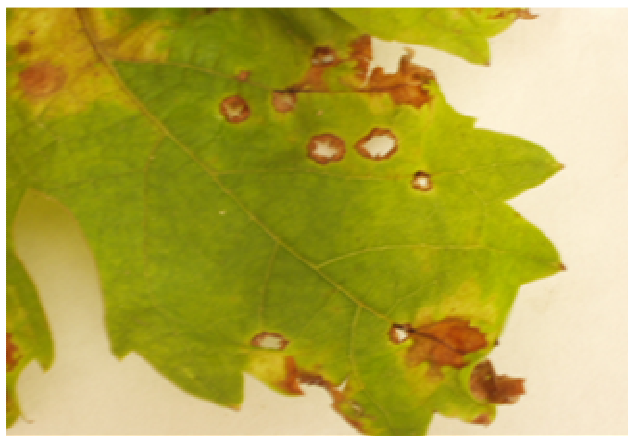
**Figure 3.** Blight of the crown of rainy season growth (July) of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe.

disease intensity (Figure 5). Lesions on internodes of canes, spurs and branches sporadically developed into cankers which also girdle to kill off all distal portions above the canker zone resulting in cane, spur and branch dieback. Dieback from cankers was most problematic where cankers escaped pruning and subsequently resulted in holdover cankers which caused extensive death, evident as fire blight.

The disease of the vegetative organs of the dry season growths was downy mildew with symptoms evident on mature and immature leaves and canes. The downy mildew symptoms vanished during the middle of the dry season growths (starting from February) and the lesions appeared like necrosis which accelerated drying off and senescence of affected organs.



**Figure 4.** Close of rainy season growth (compensatory response to defoliation in August) of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe.



**Figure 5.** Leaf section of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe, from dry season growth necrosis at the cessation of downy mildew epiphytotic consequent on increased temperatures in February.



**Figure 6.** Downy mildew on dry season growth of the grapevine, *Vitis vinifera* cvar Anap-e-Shahe in January.

### Micro-organisms of the disease complex

The two specific stains for bacteria failed to reveal bacteria on the infected segments of the disease complex over the study period. The signs of the fungi detected directly and in conjunction with cultural and morphological characteristics of isolates indicated mixture of fungi at each observation. Fungi were encountered on the outer portions of lesions on the variously symptomatic organs (spots, shot holes and necroses on leaves; tip die back; spots and blotches on petioles; early cankers comprising spots, blotches, scab on canes and spurs; and cankers on canes, spurs and stem). Dark hyphae were preponderant on the outer layers of lesions on cankers. Pycnidia were common on the outer bark lesions. The detected signs of fungi were used for their placement into the corresponding family and genus as follows: on the leaves and shoot tips were *Phoma* spp. (Spheropsidales: Spheropsidaceae); anthracnose, *Colletotrichum gloeosporioides* (Melanconiales: Melanconiaceae), *Pestalotiopsis*, *Alternaria*, *Curvularia* spp. *Penicillium* sp., *Aspergillus* sp., *Epicoccum* sp. (Moniliales: Dematiaceous Hyphomycetis); on cankers on branches and stems, *Phoma* spp., *Phomopsis* sp. (Spheropsidales: Spheropsidaceae), *C. gloeosporioides* anamorph: *Gloeosporium* sp., *Fusarium* sp., and *Epicoccum* sp. (Moniliales: Moniliaceae). Under the study conditions the perfect forms of the fungi were neither observed *in situ* nor in cultures of segments from the leaves, petioles, shoot tips, canes nor spurs.

### The fungi on the lesions

The Fungi detected on the organs of the crowns in all the sites and their frequencies of occurrence from the various lesions (leaf lesions, tip die back and branch cankers) were consistently similar for each season and are given in Table 3.

The relative frequencies of detecting the fungi and weather records on temperature, relative humidity and temperature plotted over time are illustrated in Figure 7. Except for *Plasmopara viticola*, the fungi detected on lesions of the rainy season growths were generally detected at frequencies which increased with increasing rainfall and relative humidity and the peaks of the relative frequencies of their occurrence varied and were attained at various times. The peaks of relative frequencies of occurrence of other fungi detected on lesions of the rainy season growths on the characteristic darkish spots and blotches on leaves and canes, scab on canes, and tip dieback were attained between April and May except for *Phoma* sp., *Epicoccum* sp. and *Curvularia* sp. 2. The frequency of detecting *Phoma* sp., *C. gloeosporioides* and *Phomopsis* sp. on the necrotic lesions of the rainy season growths corresponded with the distribution of the rains. Their occurrence commenced from the beginning of the season at relative frequencies which increased

**Table 3.** Occurrence of species on lesions of the disease complex of the various vegetative organs on the annual seasonal growths of the crown of grapevine (cvar Anap-e-shahe).

Species	Leaf necrosis		Tip dieback		Stem cankers			
					Cane lesions		Basal cankers	
	Season		Season		Season		Season	
	Rainy	Dry	Rainy	Dry	Rainy	Dry	Rainy	Dry
<i>Pestalotiopsis</i> aff. <i>uvicola</i>	+ <sup>a</sup>	0	+	0	+	0	0	0
<i>Phoma</i> aff. <i>multirostrata</i>	+	0	+	0	+	0	+	+
<i>Alternaria</i> sp.	+	0	+	0	+	0	0	0
<i>Fusarium</i> sp.	+	+	+	+	+	+	+	+
<i>Dreschlera</i> sp.	+	0	+	0	+	0	0	0
<i>Curvularia</i> sp. 1	+	+	+	+	+	+	+	+
<i>Colletotrichum gloeosporioides</i>	+	0	+	0	+	+	+	+
<i>Phomopsis</i> sp.	+	0	+	0	+	0	+	+
<i>Phoma</i> sp. 11	+	0	+	0	+	0	0	+
<i>Epicoccum</i> sp.	0	0	0	0	+	+	+	+
<i>Penicillium</i> sp.	+	+	+	+	+	+	+	+
<i>Aspergillus</i> sp.	+	+	+	+	+	+	+	+
<i>Curvularia</i> sp. 2	+	+	+	+	+	+	+	+
<i>Plasmopara viticola</i>	+	+	+	+	+	+	0	0

+: detected.

0: not detected.

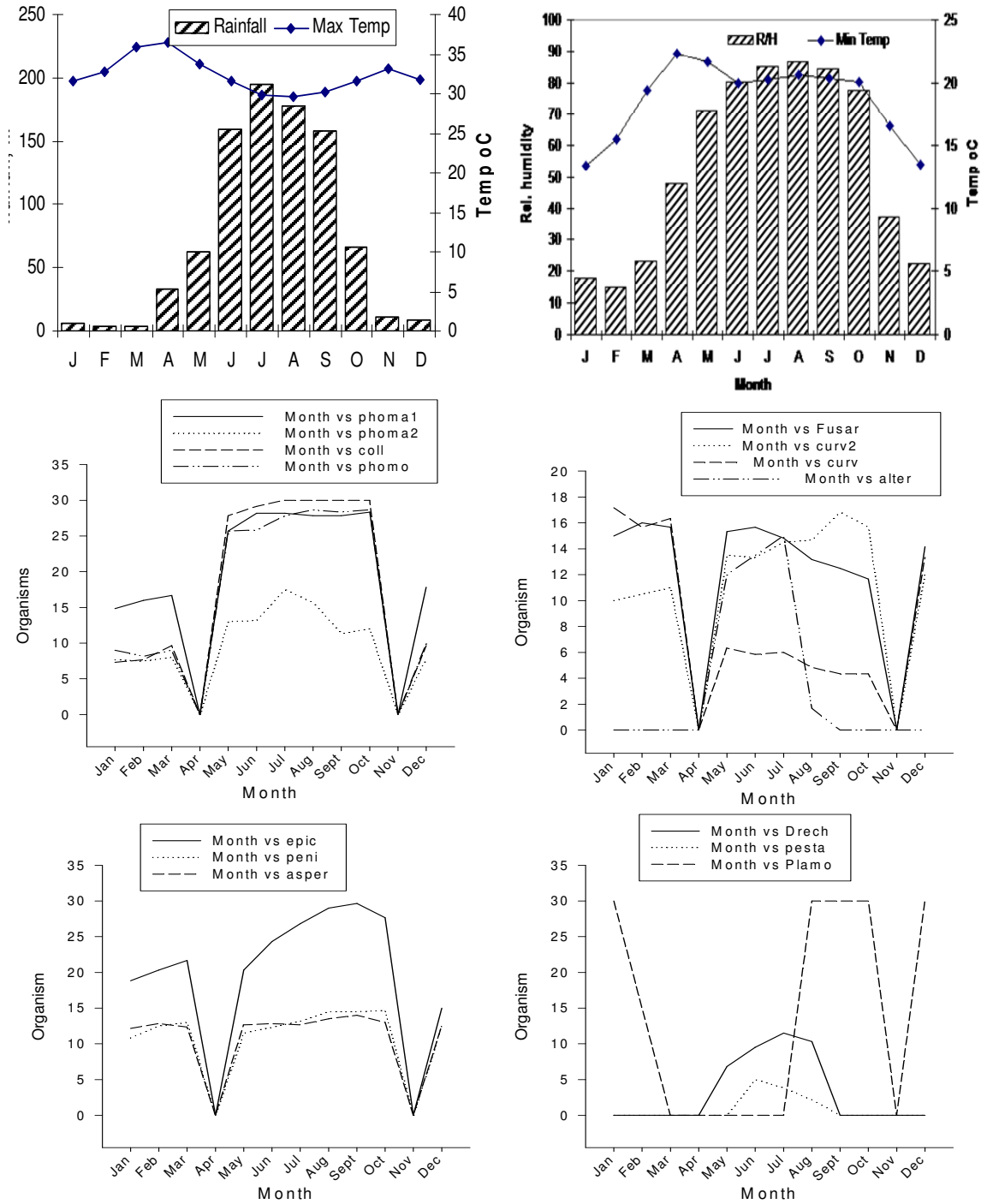
with increasing rainfall and relative humidity. The peaks of the relative frequencies of occurrence were highest for *Phoma* aff. *multirostrata*, *C. gloeosporioides*, *Phomopsis* sp., *Epicoccum* sp. and *P. viticola*. *Penicillium* sp. and *Aspergillus* sp. occurred at frequencies with relatively low peaks. The relative frequencies of detecting *Fusarium* sp. and *Curvularia* sp. also followed a similar pattern, however, after the peaks of the relative frequencies in May their occurrence started decreasing slightly before the pruning at the close of the rains in October. The detection peaks for *Dreschlera* sp. and *Epicoccum* sp., similarly detected throughout the rainy season growths were attained in July, which usually was mid rainy seasons when rains and relative humidity were at their peak. However, the relative frequencies of their occurrences were slightly lower than the peaks of *Phoma* aff. *multirostrata*, *C. gloeosporioides*, *Phomopsis* sp., *Epicoccum* sp. and *P. viticola*. *Alternaria* sp. detected on necrotic lesions from the beginning of the rains in April also attained high peak relative frequencies of occurrence during the peak of the rains, however, the occurrence abruptly ceased after the peak occurrence in July. The occurrence of *Pestalotiopsis* aff. *uvicola* and *P. viticola* on lesions of the rainy season growths delayed to May and July respectively with *Pestalotiopsis* aff. *uvicola* having very low short-lived peak of relative frequencies of occurrence in June, while *P. viticola* had very high long-lasting peak of relative frequencies of occurrence beginning from July to February. *P. viticola* started being detected during the cold rainy season growths from the end of July, which was the general peak for all the fungi dete-

cted during the rains. The peak occurrence of *P. viticola* persisted into the dry seasons, even with the pruning interruptions of September-October. However, there was a resurgence of *P. viticola* as the cool months persisted.

*P. viticola* was detected on downy mildew lesions on leaves and canes from the early dry season growths, during the periods of low rainfall, low humidity and low maximum temperature which persisted till February. Thereafter, the downy mildew with the attendant epiphytotic of *Plasmopara* sp. was no longer evident when temperatures began to rise late in the dry seasons and downy mildew symptoms started appearing as mere necrosis. *Epicoccum* sp., *Fusarium* sp. and *Curvularia* sp. were also detected on lesions during the dry season; however, the peaks of their relative frequencies of occurrences were lower than realized for *P. viticola*.

There were fungi with relative frequencies of occurrence that differed for the two seasons of the sampling years. The fungi which varied with the seasons included *Phoma* spp., *C. gloeosporioides*, *Phomopsis* sp., *Curvularia* sp. and *Epicoccum* sp. detected on lesions of the rainy season growth at higher peaks of frequencies of occurrence than on lesions of the dry season growth. *Alternaria*, *Dreschlera* and *Pestalotiopsis* sp. were detected on lesions of the rainy season growths only and never on the dry season growths. Though *P. viticola* was detected on rainy season lesions the most conspicuous occurrences were the dry season peaks.

The peak relative frequencies of detecting *Curvularia* sp. were also higher on dry season lesions than on rainy season lesions.



**Figure 7.** Frequency of detection of the organisms plotted against the month of their detection. Fusar = *Fusarium* sp., phoma 1 = *Phoma* aff. *multirostrata*, phoma 2 = *Phoma* sp., curv = *Curvularia* sp., curv2 = *Curvularia* sp. 2, coll = *Colletotrichum gloeosporioides*, phomo = *Phomopsis* sp., alter = *Alternaria* sp., epic = *Epicoccum* sp., Drech = *Drechslera* sp., peni = *Penicillium* sp., pesta = *Pestalotiopsis* aff. *uvicola*, asper = *Aspergillus* sp., and Plamo = *Plasmopara viticola*.

On the other hand there were regularly detected species with little or no difference in the relative frequencies of occurrence for the two seasons of the sampling years. The regularly detected species included *Penicillium* sp.,

*Aspergillus* sp. and *Fusarium* sp. that were regularly detected, regardless of the growth season and the type of lesion whether spots, blotches, cankers, or downy mildews.

## DISCUSSION

Our study provides the first documentation of the mycoflora of the crown disease complex of the vegetative organs of grapevine in Nigeria, though many fungi have been reported as causal agents of grapevine diseases in Nigeria. The results detected species that were more or less seasonal, specific to particular symptoms and which had occurrence frequency patterns that were consistent with succession of microorganisms (Dickinson, 1971). Starting with lesions on early rainy season growths, succession commenced with occurrence of every other detected fungi, except *P. viticola* and *Pestalotiopsis* sp. The periods of increases in the relative frequencies of occurrence of the various species is connected with the pathogenic roles of the detected fungi. Although reports on the pathogenic role of *C. gloeosporioides* on grapevines have been inconsistent (Mirica, 1988; Quimio and Quimio, 1975; Swaroop et al., 1994), the pathogenicity of *C. gloeosporioides* and *Alternaria* sp. on grapevines in Nigeria has been reported (Chindo, 1991; Majanbu, 1995). The fact that *C. gloeosporioides*, *Phoma* aff. *multirostrata*, *Phoma* sp., *Phomopsis* sp., *Pestalotiopsis* aff. *uvicola*, and *Alternaria* sp. are pathogenic to grape vines and other crops is consistent with earlier reports (Lopes and Boiteux, 1994; Forster and Adaskaveg, 1999; Mostert et al., 2001). Though *Drechslera* sp. has not been implicated in the causation of crown disease of vegetative parts of grapevines, the genus induces leaf spot and stem blight of crops (Mehta, 2001). Whether the decreasing relative frequency of detecting *Alternaria* sp., *Drechslera* sp. and *Pestalotiopsis* aff. *uvicola* should be attributed to their intrinsic weakness (inability to compete with other associated fungi) is open to further elucidation but Lesney and Felker, (1993) reported a species of *Pestalotiopsis* as a weak wound pathogen. However, the notion that high frequency of occurrence of a species is a measure of dominance is simplistic for Lopes and Boiteux (1994) reported on the saprophyte, *C. gloeosporioides* that thrived on leaf spot and stem blight lesions of sweet potato which often out grew the pathogenic *Alternaria* sp. when infected tissues were surface sterilized and plated on PDA. The late occurrence of *Pestalotiopsis* sp. when the relative frequency of occurrence of other implicated pathogenic species were on the increase, coupled with the fact that it is a wound pathogen of young leaves, shoot tips and cane tissues suggest that it could thrive as a secondary pathogen, invading immature tissues wounded by other primary pathogens. In addition, it was capable of neither infecting older tissues through pruning wounds nor infecting tissues directly early in the season.

The sudden enhanced disease condition observed late in July while in the mid rainy seasons and the peak of the rains can be attributed to *P. viticola* acting in concert with other concomitant fungi along with the effect of secondary dispersal and infection of propagules of resident fungi including saprophytes with the exudates from deco-

posing tissues enhancing further infections and colonization by microorganisms (Pico and Rodolfi, 2004; Hajjeh et al., 2005). The high rainfall, high relative humidity, cool tropical maximum and minimum temperatures could have favoured the multiplication and the interplay of the abounding species, hence the disease severity.

That the regularly detected species of mycoflora which include *Fusarium* sp., *Penicillium* sp., *Aspergillus* sp. and *Curvularia* spp. exist as opportunistic infections, deriving nourishment from the dead tissues and further disintegrating host tissues through their feeding corroborates earlier reports (Dickinson, 1971; Diem, 1971; Melgar et al., 1994).

The mycoflora detected on the crown disease complex of the vegetative organs of grapevine in Zaria in the Northern Guinea Savanna over time has shown four categories of fungi acting in concert. Among the species *P. viticola* occurred as a primary pathogen inducing downy mildew during the cold months prevailing from the rainy season late in July, and persisted beyond the rainy season pruning, into the dry season while *Alternaria* sp., *Phoma* aff. *multirostrata*, *C. gloeosporioides*, *Phomopsis* sp., and *Phoma* sp. were primary invaders of the rainy season. The wound pathogen, *Pestalotiopsis* aff. *uvicola*, thrived only during the early rainy season while *Fusarium* sp., *Penicillium* sp., *Aspergillus* spp., *Curvularia* spp. and *Epicoccum* sp. were opportunistic infections, deriving nourishment from the dead tissues and further disintegrating dead tissues through their feeding. Future study of antagonism or synergism among the species of the fungal complex and the influencing weather parameters would provide useful epidemiological facts for the management of the disease complex.

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