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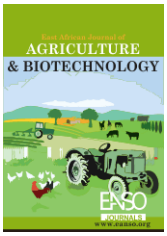

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Original Article

Phenotypic Characterisation of Colletotrichum lindemuthianum Isolates from Major Bean-Growing Regions in Tanzania.

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Colletotrichum lindemuthianum, Races, Resistant genes, Differential cultivars, Anthracnose.

ABSTRACT

Anthracnose, caused by the fungus Colletotrichum lindemuthianum, is a major disease of common bean (Phaseolus vulgaris L.). C. lindemuthianum is highly variable; therefore, understanding the pathogen's diversity is a key step in developing common bean varieties that can resist anthracnose disease. This work provides the first comprehensive update on C. lindemuthianum races in Tanzania since 2017, using a set of differential bean cultivars. A field survey was conducted in 10 bean-growing districts in Tanzania to collect plants. A total of 100 infected samples were collected, and 30 pure isolates were obtained and characterised based on their reactions to 12 common bean differential cultivars. The virulent races identified were race 55 from Morogoro; 820, 944 and 3842 from the Njombe region; 1804 from Kigoma; and 3210 and 3842. The least virulent race was characterised as race 2 from the Arusha region. Races 2, 27, 32, 55, 129, 274, 344, 524, and 776, obtained from this study, have been previously reported, and 24 races were identified for the first time in Tanzania. Races 2 and 55 gave a susceptible reaction only to Andean cultivars, while races 129, 329, 257, 776, and 2056 gave a susceptible reaction only to Middle American cultivars. The remaining races gave a susceptible reaction on both Andean and Middle American cultivars. Races 2056, 3088, 3210, and 3842, three from Kilimanjaro and Arusha and one from Njombe, were susceptible to the resistant genes found in cultivar G2333, which is used for resistant gene pyramiding in common bean. The current study revealed the existence of both Andean and Middle American races in Tanzania. The knowledge on the race structure of C. lindemuthianum provided in this study will help in making breeding decisions regarding the host plant resistance genes required to develop common bean varieties with durable resistance to anthracnose in Tanzania.

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INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is an important food and nutritional security pulse crop that provides a cheap source of vegetable proteins, micronutrients (iron and zinc, and vitamins) and dietary fibre (Lisciani *et al*., 2024). In Tanzania, common bean is not only a crucial protein source but also a significant cash crop for many farmers, in major growing regions including Njombe, Morogoro, Arusha, Kigoma and Kilimanjaro (Nakei *et al.,* 2023). The low productivity of commercially cultivated bean varieties is due to many biotic and abiotic factors. Among the biotic factors are anthracnose, angular leaf spot, Bean common mosaic virus, common bacterial blight, halo blight, and bean anthracnose (Padder *et al.,* 2017). Bean anthracnose caused by the fungus *Colletotrichum lindemuthianum* (Sacc. & Magn.) is one of the most serious diseases in common bean. In fact, under *C. lindemuthianum*, in favourable conditions, anthracnose can reduce bean production up to 100% (Campa *et al.,* 2014). Bean anthracnose can be managed by the important management of seed-borne diseases, which is to eradicate or reduce the pathogen inoculum in the seed production field (Mohammed, 2013). Management strategies used to minimise seed-borne infection in the seed production field include host resistance, cultural, chemical and biological control methods. The use of resistant cultivars to the disease can be effective, but there is a possibility of breakdown of resistance due to adaptation of the pathogen. Therefore, varieties must be tested where they are to be grown to determine their tolerance to the locally prevalent races (Nunes *et al.,* 2021).

Colletotrichum lindemuthianum exists in many physiological forms in all bean-growing regions of the world and infects bean genotypes from both bean gene pools, that is, the Andean and Mesoamerican (Mahuku & Riascos, 2004). The fungus is known to have races that vary across countries, regions, locations and varieties (Nalupya, 2021). The highest diversity and variation are reported from Latin America, which is the centre of origin of the common bean (CIAT, 1997). East African regions are considered the secondary centre of diversity for common bean, and due to the co-evolution of the *C. lindemuthianum* pathogen and its host (Gezahegn *et al.,* 2021). The East African highland regions are expected to have a high variability of *C. lindemuthianum* because of the introduction of new varieties from different gene pools and the movement of bean seed within the country and region.

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The diversity among isolates of different races and even within the same race of *C. lindemuthianum* is attributed to its reproduction, lifecycle and host selection (Padder *et al*., 2017). *Colletotrichum lindemuthianum* has a parasexual cycle that involves heterokaryosis, where different nuclei from fusing hyphae coexist in the same cell, leading to potential genetic recombination without a true sexual phase provide the basis for generating variability (Padder *et al., 2017*).

Assessment of genetic variability within and among races of *C. lindemuthianum* is based on genome sequencing of conserved regions, which includes the Internal Transcribed Spacer (ITS) region (Stewart *et al.,* 1999). The DNA encoding ribosomal RNA (rRNA) has a genomic cluster that repeats hundreds of times in the genome and is composed of several genes, including those for the 18S, 5.8S, and 28S rRNA subunits. These genes are separated by the ITS 1 and ITS 2 regions, which are transcribed, resulting in the rRNA (Balardin *et al.*, 1999). However, differential cultivars provide a simple and effective method for studying pathogen virulence diversity of *C. lindemuthianum*, which has developed over time and in different locations (Pastor-Corrales *et al*., 1995). The sequences of the ITS regions can be compared to assess the relatedness of isolates and characteristics of a species' variability.

The Andean and Mesoamerican groups of *C. lindemuthianum* correspond to the Andean and Mesoamerican gene pools of the common bean. Andean races of the anthracnose pathogen are typically isolated from large-seeded bean hosts in the Andean gene pool (Darben *et al.,* 2017). Conversely, Mesoamerican races are often, though not always, isolated from small- or medium-seeded bean hosts within the Mesoamerican gene pool. Mesoamerican races of *C. lindemuthianum* usually exhibit considerably greater virulence and genetic diversity than Andean races. More importantly, Andean races of the pathogen mostly overcome resistance conferred by Andean common bean, while Mesoamerican races of *C. lindemuthianum* are compatible with Mesoamerican and Andean common bean (de Lima Castro *et al*., 2017). The Tanzanian bean populations belong to both the Andean and Mesoamerican gene pools.

This study aimed at phenotypic characterisation of *C lindemuthianum.* isolates found in bean-growing regions in Tanzania, so as to have a current update on the races that are present in Tanzania, whether they are Mesoamerican races or Andean races. The knowledge of the fungus's variability will improve the management practices by reducing bean anthracnose disease and reducing the use of susceptible cultivars.

MATERIALS AND METHODS

Study Area for Sample Collection

Anthracnose-infected common bean samples were collected from major bean-growing regions in Tanzania. The regions included Morogoro, Arusha, Kilimanjaro, Njombe and Kigoma; the specific sites and their GPS are indicated in Table 1. The diseased samples were collected during 2023/2024 during the Masika; the common bean plants were at the reproductive growth stage. The selection of regions was based on potential common bean production. The sample collection followed a method by Hughes *et al*. (1996) that involved cluster sampling. Cluster sampling was employed, with major bean-growing regions of Tanzania (Morogoro, Njombe, Kilimanjaro, Arusha and Kigoma) designated as primary clusters. Within each region, four bean-producing areas were selected as secondary clusters based on production intensity. From each area, bean farms were randomly selected with assistance from agricultural extension officers, and diseased plant samples exhibiting anthracnose symptoms caused by *Colletotrichum lindemuthianum* were collected. GPS coordinates of each sampling site were recorded to ensure spatial representation of the pathogen population. Therefore, using a $1m^2$ quadrat, whereby all diseased plants within the quadrat were observed, and diseased pods were

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collected. In each quadrat, six plants were sampled. The pod samples collected were immediately wrapped in paper towels to absorb their moisture, placed in paper bags and labelled with a pencil. The common bean varieties grown in each farm was also recorded. The collected bean samples were transported to the Pathology laboratory at the University of Dar es Salaam and stored at 4℃ for further testing.

Figure 1: Pod Samples Collected

Table 1: Areas for Bean Sample Collection

REGION	Area	GPS Location
Morogoro	Mlali	6° 57' 15" S and 37° 32' 15" E
	Mgeta	7° 02' 45" S and 37° 34' 15" E
	Tangeni	6° 55' 45" S and 37° 36' 30" E
	Konga	6° 54' 50" S and 35° 55' 40" E
Njombe	Shaurimoyo	10° 02' 46" S and 34° 43' 24" E
	Mavanga	9° 54' 06" S and 35° 06' 39" E
	Amani	9° 56' 40" S and 34° 53' 40" E
	Lugarawa	9° 48' 20" S and 34° 42' 48" E
Kilimanjaro	Moshi urban	3° 18' 30" S and 37° 22' 30" E
	Sanya Juu	3° 10' 30" S and 37° 08' 30" E
	Machame	3° 12' 35" S and 37° 13' 20" E
	Kiboriloni	3° 19' 45" S and 37° 22' 30" E
Arusha	Oldonyo Sambu	2° 09' 30" S and 35° 54' 30" E
	Ngaramtoni	3° 20' 45" S and 36° 35' 30" E
	Tengeru	3° 22' 45" S and 36° 47' 20" E
	Ilboru	3° 20' 45" S and 36° 40' 45" E
Kigoma	Kitagata	4° 22' 30" S and 30° 22' 30" E
	Nyakitonto	4° 30' 20" S and 30° 12' 35" E
	Mugombe	4° 26' 45" S and 30° 18' 15" E
	Busunzu	3° 57' 45" S and 30° 35' 45" E

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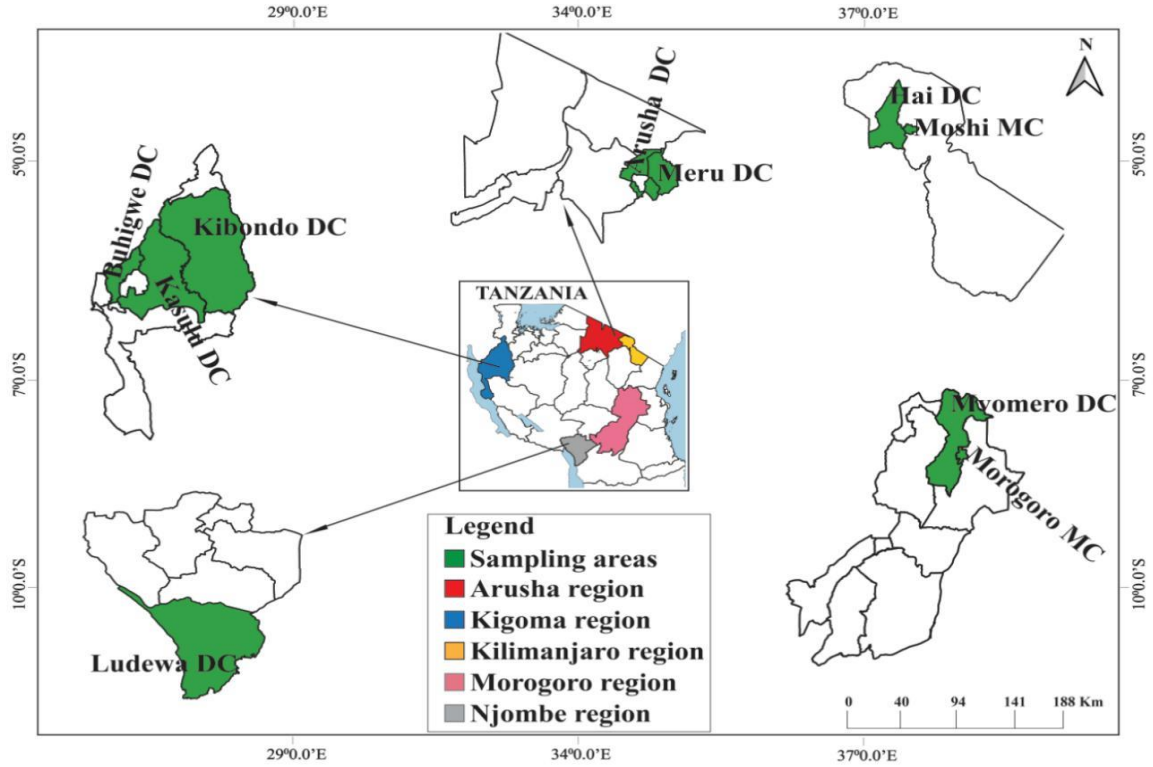
Figure 2: Map of Regions where Anthracnose Bean Samples were Collected



Isolation of the Pathogen

The collected common bean pods were processed at the Pathology laboratory, at the College of Natural and Applied Sciences at the University of Dar es Salaam, located at 6° 46' 52" S and 39° 12' 22" E. The pod samples were surface disinfected in 1% Sodium hypochlorite for 3 minutes, then rinsed three times in sterile distilled water. Small cuts of 5cm were taken from the symptomatic areas and placed on Potato Dextrose Agar in Petri dishes. The petri plates were incubated in the darkroom at 22-25 ℃ for seven days. The colony growth of the fungus was recognised based on its morphological characteristics, such as colony colour and conidia shape. The fungal colony and monoconidial were obtained by picking pinpoint colonies using a sterile needle and then sub-cultured on fresh PDA media to get a pure culture using the single spore technique and hyphae tip method (Harlapur *et al*., 2007). Continuous sub-culturing was made until pure cultures with no contamination were obtained. During culturing, streptomycin sulfate (5 mg L-1 in PDA) was used to suppress bacterial colony growth. A total of 30 pure isolates were obtained, and they were kept in the refrigerator at $4^{\circ}C$ for later use.

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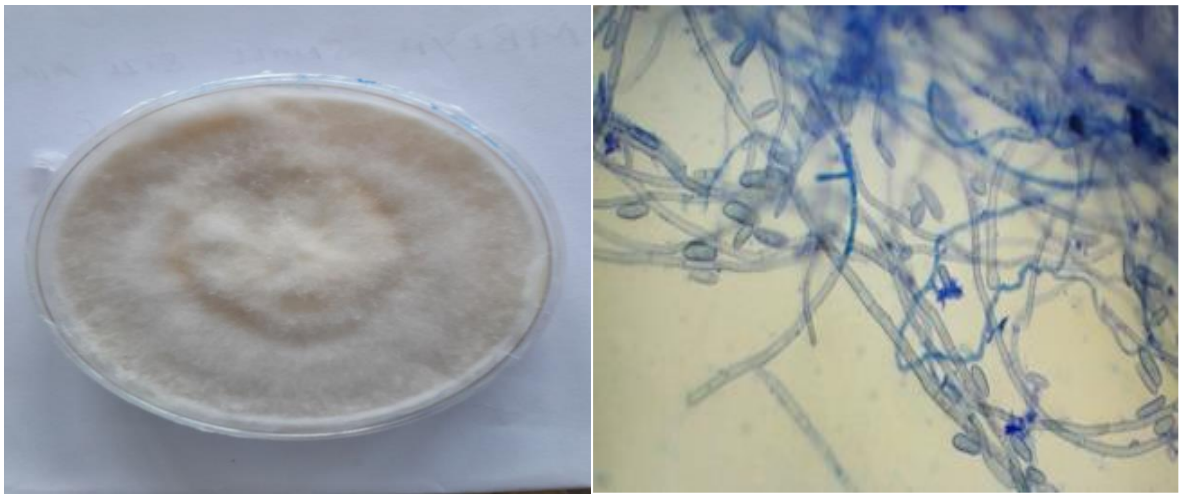
Plate 1: a. C. *lindemuthianum* colony on PDA b. C. *lindemuthianum* conidia under Microscope X40 Magnification were kept in the Refrigerator for Later Use



Differential Cultivars

A set of 12 bean differential cultivars that were used for race classification comprised two gene pools: four Andean and eight Mesoamerican origins, as shown in Figure 4 (Kiryowa *et al.*, 2017). The designated binary code and characteristics of the standard cultivars as described by Pastor-Corrales (1991) were obtained from CIAT and are presented in Table 2. To obtain sufficient seed for race identification, race-differential seeds obtained from CIAT were multiplied in the screenhouse at the Department of Crop Science and Horticulture at SUA in 2023. The seeds were surface sterilised with 0.1% chlorox solution for three minutes, washed or rinsed thoroughly with sterile distilled water and dried at room temperature (Gezahegn *et al.*, 2021). The purpose of surface disinfecting the seeds with 0.1% chlorox solution was to remove microorganisms (such as fungi and bacteria) present on the seed surface.

Table 1: Standard Differential Cultivars of Common Bean Used to Characterise *Colletotrichum lindemuthianum*, Their Binary Codes, Resistance Genes and Gene Pool

S/N	Differential cultivars	Seed type	Notation	Binary code	Host genes	Gene pool
1	Michelite	S	0	1	Co-1^1	MA
2	Michigan Dark Red Kidney	L	1	2	Co-1	A
3	Perry Marrow	L	2	4	Co-1^3	A
4	Cornell 49-242	S	3	8	Co-2	MA
5	Widusa	L	4	16	Co-1^5	A
6	Kaboon	L	5	32	Co-1^2	A
7	Mexico 222	S	6	64	Co-3	MA
8	PI 207262	S	7	128	Co-4^3 Co-9	MA
9	TO	S	8	256	Co-4	MA
10	TU	S	9	512	Co-5	MA
11	AB 136	S	10	1024	Co-6	MA
12	G2333	S	11	2048	Co-42 Co-52 Co-7	MA

**S: Small seeded; L: Large seeded MA = Middle American gene pool; A = Andean gene pool of Phaseolus vulgaris. Binary code: 2n, where n is equivalent to the place of the cultivar within the series (0-11).*

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Plate 2: Appearance of Seeds of Anthracnose Differential Bean Cultivars a. Andean Gene Pool b. Mesoamerican Gene Pool

Raising the Differential Cultivars.

Four seeds of each set of the 12-anthracnose differential cultivars were sown in separate pots in a screen house at the College of Natural and Applied Sciences, at the University of Dar es Salaam. The potted soil media had a mixture of sterilised forest soil and peat moss at a 1:1 ratio. After sowing the seeds, irrigation was done daily to facilitate seed germination. Thinning was done before inoculation to maintain only two seedlings per pot after the plants had developed a set of true leaves.

Pathogenicity Test on the Differential Cultivars

The experiment was laid out in a complete randomised design in three replications. Each pot was considered a replication. Inoculation on differential cultivars was done 7 days after sowing, and for each fungal isolate, two plants were inoculated. The differential cultivars were at the V1 growth phase with two cotyledons and primary leaves at nodes 1 and 2. Single spore/monosporic cultures of C. *lindemuthianum* were grown on PDA and incubated in the dark for 10 to 15 days. For inoculation, conidial spores were scraped off the growth medium using 10 ml of sterile distilled water to make a suspension. The spore concentration used was quantified and adjusted to a spore concentration of 1.2×10^6 spores/ml (Pastor-Corrales *et al.*, 1995) using the hemocytometer (Neubauer-Preciss Chamber) under the microscope, and 0.1% Tween 20 was added as a surfactant. Seven-day-old seedlings were used for inoculation. Using a hand sprayer, all 12 differential cultivars were inoculated separately, each with the respective isolate suspension, until all 30 isolates were sprayed on both the abaxial and adaxial surfaces of the leaves. The spore suspension was sprayed until runoff. Control plants were inoculated with sterile distilled water and were included for each set of the differential cultivars and each isolate of the pathogen. A relative humidity of approximately 95% was maintained at a temperature of 22±2 ℃ to enhance infection. Each plant was covered with a transparent polythene sheet for four days after inoculation, followed by regular management such as watering and fertiliser application.

Race Determination

Symptoms evaluation on inoculated plants was done 14 days after inoculation, based on a 1-9

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severity scale, depicting the percentage of the leaf covered with anthracnose lesions as described by (Balardin *et al.*, 1997); Where; 1 = no symptoms (resistant), 2 – 3 = very small lesions mostly on primary leaves (resistant) and 4-9 = numerous enlarged lesions or sunken cankers on the lower side of the leaves or hypocotyls (susceptible). Cultivars with scores 1-3 were regarded as resistant, while those with scores 4-9 were regarded as susceptible. A differential cultivar considered susceptible had 60% (3/5) of its inoculated plants exhibiting the same disease, with scores within the susceptibility range (Ansari *et al*., 2004). Race identification was done as the sum of binary numbers of all differential cultivars showing susceptible reaction to the particular isolate, and each isolate was given a race name (number) based on the binary nomenclature system by adding the numerical binary values of susceptible cultivars together (CIAT, 1988).

Data Analysis

Race isolation frequency was calculated based on the number of times a race was identified among all isolates. The number of areas in which a race was represented provided insight into the distribution of races. Isolation frequency, resistant index, and virulence index was calculated using the formula below.

Resistant index, for each of the 12 differential cultivars

$$RI = \frac{RC \times 100}{T}$$

RC = Total number of races that each resistant differential cultivar

T = Total number of races *C. lindemuthianum*

Virulence index

$$VI = \frac{S \times 100}{T}$$

S = Total number of differential cultivars susceptible to the race

T = Total number of differential cultivars (12)

Isolation Frequency (%) = $\frac{\text{Number of isolates belonging to a specific race}}{\text{Total number of isolates identified}} \times 100$

RESULTS

Race Determination

In this study, 30 tested pure isolates were categorised into 25 physiological races based on the reaction recorded on the differential cultivars, as shown in Table 3. The physiological races obtained from the study were races 2, 27, 30, 32, 55, 66, 129, 140, 166, 257, 267, 274, 329, 344, 524, 534, 776, 820, 944, 1092, 1804, 2056, 3088, 3210 and 3842. However, races 66, 129, and 267 were characterised twice from isolates obtained from different regions and had an isolation frequency of 6.7%. Meanwhile, race 524 was characterised thrice from isolates in different regions and had an isolation frequency of 10%.

Race Distribution

The races 55, 820, 944, 1804, 3210, and 3842 showed the highest level of susceptibility reactions on the differential cultivars, as shown in Table 4, whereby three were from the Njombe region, and the remaining races were each characterised from isolates obtained from Morogoro, Kigoma, and Kilimanjaro regions. The most virulent races affected five differential cultivars. Furthermore, race 66 was characterised from isolates in Morogoro and Njombe, as shown in Table 3. Race 524 was the most widely distributed across the sampled regions since it was characterised from three regions, namely: Arusha, Kilimanjaro and Morogoro. These are all high-altitude bean-growing regions ranging from 1400–2000m above sea level and therefore offer favourable conditions for the development and spread of bean anthracnose disease. Four races were

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able to infect the cultivar G233, these were race 3842 from Njombe region and races 2056, 3210 from Kilimanjaro and 3088 from Arusha. Race 129 and 776 affected only the Middle American cultivars, as shown in Table 3. Cornell 49-242 was the most infected bean cultivar, as shown in Table 4, and was infected by races from all regions. Race 66, 129, 257 and 2056 affected only two differential cultivars. Race 2 from Arusha region affected only the Andean bean cultivars. Races collected from Arusha and Kilimanjaro regions affected the most differential cultivars. The races that caused a susceptible reaction to only Mesoamerican cultivars were races 129, 257, 776, and 2056. Moreover, some races infected only one gene pool and not the other, they overcame some of the anthracnose-resistant genes and not others (the anthracnose resistance genes in parentheses). Meanwhile, races that affected only the Andean cultivars were races 2, 32 and 329, as shown in Table 4.

Table 2: The number of Isolates and Races of *Colletotrichum lindemuthianum* Identified in Each Region by Phenotypic Characterization

Regions	Cultivars grown	Races
Morogoro	Njano	30, 55, 66, 129, 140, 524,
Njombe	Lyamungo 90, Njano	32, 66, 776, 820, 944, 3842
Arusha	Lyamungo 90, Njano, Lyamungo 85	2, 267, 524,534, 1092, 3088,
Kilimanjaro	Lyamungo 90, Njano, Lyamungo 85	27, 344, 524, 534, 2056, 3210
Kigoma	Njano, Morogoro, Mworo Kabhengwa	129, 166, 257, 274, 329, 1804

Table 3: Responses of 12 Differential Cultivars to C.lindemuthianum

	DIFFERENTIAL CULTIVARS												
ISOLATES	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**RACES**
SHZ 5	R	R	R	R	R	S	R	R	R	R	R	R	32
SHZ 6	R	S	R	R	R	R	S	R	R	R	R	R	66
SHZ 7	R	R	S	R	S	S	R	R	S	S	R	R	820
SHZ 8	R	R	R	S	R	R	R	R	S	S	R	R	776
SHZ 9	R	R	R	R	S	S	R	S	S	S	R	R	944
SHZ 10	R	S	R	R	R	R	R	R	S	S	S	S	3842
EZ11	R	S	R	R	R	R	S	R	R	R	R	R	66
EZ12	R	S	S	S	S	R	R	R	R	R	R	R	30
EZ13	S	S	S	R	S	S	R	R	R	R	R	R	55
EZ14	S	R	R	R	R	R	R	S	R	R	R	R	129
EZ15	R	R	S	S	R	R	R	R	R	S	R	R	524
EZ16	R	R	S	S	R	R	R	S	R	R	R	R	140
NZ17	R	R	S	R	R	R	S	R	R	R	S	R	1092
NZ18	R	S	R	R	R	R	R	R	R	R	R	R	2
NZ19	S	S	R	S	S	R	R	R	R	R	R	R	27
NZ20	S	S	R	S	R	R	R	R	S	R	R	R	267
NZ21	R	R	R	R	S	R	R	R	R	R	S	S	3088
NZ22	S	S	R	S	R	R	R	R	S	R	R	R	267
WZ23	S	R	R	R	R	R	R	S	R	R	R	R	129
WZ24	S	R	R	S	R	R	S	R	S	R	R	R	329
WZ25	R	S	S	R	R	S	R	S	R	R	R	R	166
WZ26	S	R	R	R	R	R	R	R	S	R	R	R	257

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	DIFFERENTIAL CULTIVARS												
ISOLATES	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**RACES**
WZ27	R	S	R	R	S	R	R	R	S	R	R	R	274
WZ28	R	R	S	S	R	R	R	R	S	S	S	R	1804
NZ29	R	R	S	S	R	R	R	R	R	S	R	R	524
NZ30	R	R	R	S	S	R	S	R	S	R	R	R	344
NZ31	R	S	S	R	S	R	R	R	R	S	R	R	534
NZ32	R	R	R	S	R	R	R	R	R	R	R	S	2056
NZ33	R	S	R	S	R	R	R	S	R	R	S	S	3210
NZ34	R	R	S	S	R	R	R	R	R	S	R	R	524

*Races characterised using the binary system (Pastor Corales, 199); Resistant reaction **(R); Susceptible(S)**

**Differential cultivars: 1= Michelite, 2= Michigan Dark Red Kidney, 3= Perry Marrow, 4= Cornell 49-242, 5= Widusa, 6= Kaboon, 7= Mexico 222, 8= PI 207262, 9= TO, 10= TU, 11= AB 136, 12=G2333.*

Virulence Spectrum of C. *lindemuthianum* Isolates to Resistant Genes

The differential cultivars with the resistant genes were ineffective against most of the isolates tested. The differential cultivar response to *C. lindemuthianum* isolate interaction, a resistance or susceptibility range with virulence gene frequency in the range of 0% to 46.7%. The differential cultivar, Cornell 49-242, was found to be the most susceptible, followed by Michigan Dark Red Kidney virulence frequency of 43.3%, and lastly Perry marrow and TO both had a virulence frequency of 36.7%. These cultivars were susceptible to 14 and 13 different isolates of *C. lindemuthianum*, respectively. Likewise, the differential cultivars Widusa and TU were found to be moderately resistant to *C. lindemuthianum* isolates, all with a virulence gene frequency of 30%, meanwhile Michelite had 26.7 %. On the other hand, the differential cultivar PI 207262 that carries resistance gene Co-4^3 and Co-9 was only infected by 6 *C. lindemuthianum* isolates and had a virulence frequency of 20%. Kaboon, Mexico 222, and AB136 that carry the resistant gene Co-1^2, Co-3 and Co-6 were all infected by 5 different *C. lindemuthianum* isolates all had a virulence frequency of 16.7%. According to this evaluation, these differential cultivars were highly resistant to different *C. lindemuthianum* isolates. Lastly, 4 isolates were found to be pathogenic to the G2333 differential cultivar, which consists of resistant genes "Co-42, Co-5, Co-7" (Fig. 2.2). This difference in resistance among the differentials, or in virulence and virulence in C. lindemuthianum, is genetically controlled and determined by the gene-for-gene model. Common bean (*Phaseolus vulgaris* L.) has different pre-existing structural or chemical defence mechanisms that are present in plants before the pathogen attack and induce structural or chemical defences activated by the presence of the pathogen in the host tissue to *C. lindemuthianum* (Agrios, 2005). This made the cultivar responses to the pathogen different from each other.

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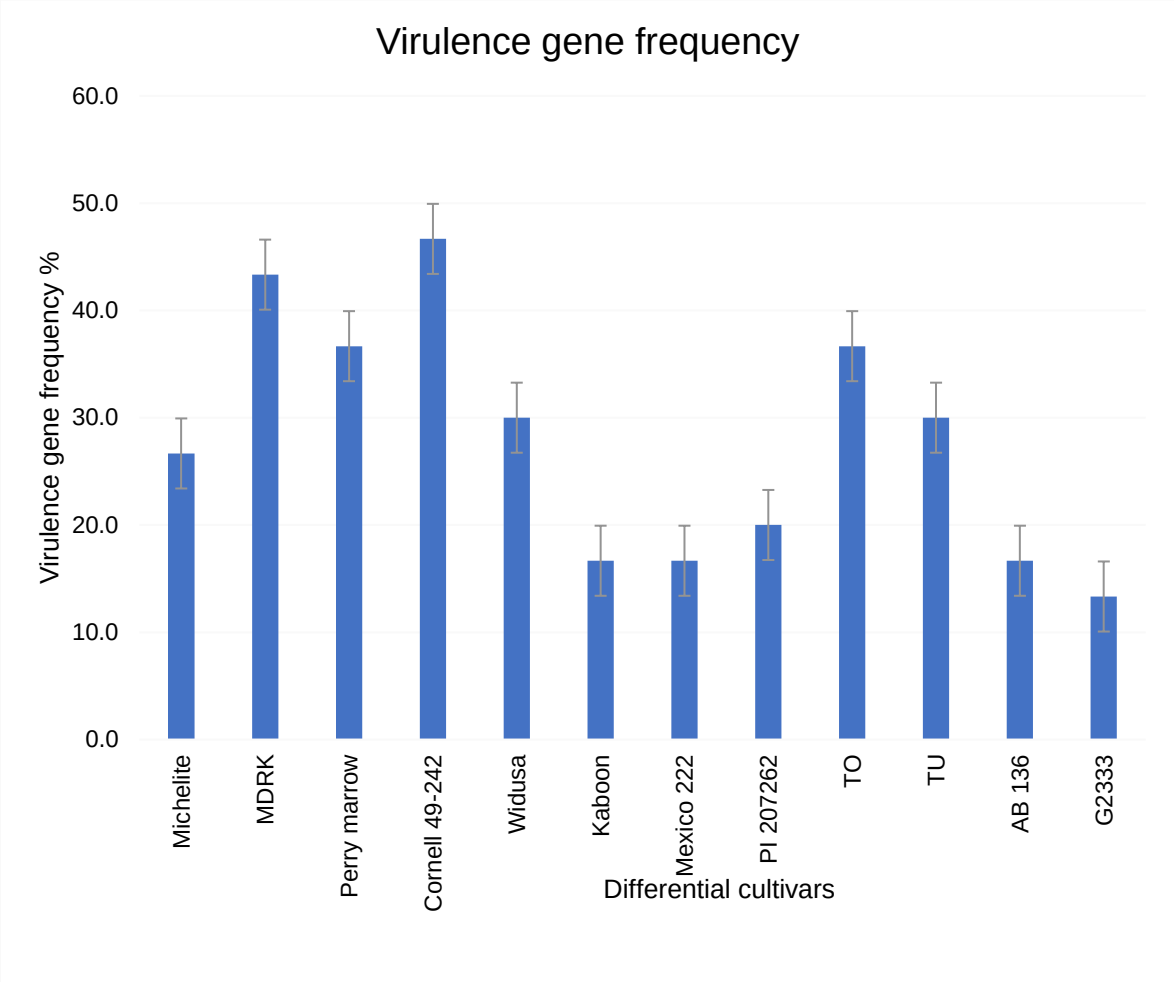
Figure 3: Virulence Gene Frequency Depicting Susceptible Reaction of Common Bean Differential Cultivars to the Tested Isolates



NB. *Cultivars MDRK, Perry Marrow, Widusa and Kaboon are of Andean origin, whereas Michelite, Cornell 49242, Mexico 222, PI 207262, To, TU, AB 136 and G 2333 are of Mesoamerican origin.*

Isolation Frequency of the Physiological Race of *C. lindemuthianum*

The isolation frequency of 25 physiologically distinct races of *C. lindemuthianum* obtained in this study ranged from 3.3% to 10.7%. The most predominant and widely distributed races identified were 66 and 524, with an isolation frequency of 10%, while races 129 and 267 had an isolation frequency of 6.7%. The lowest frequency of 3.3 was recorded on the remaining races, as shown on table 1.96% was recorded on races 1403 and 1033 (Tables 5 and 6).

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Table 5: Isolation Frequency of Races of C. *lindemuthianum*

RACES	Differential cultivar infected	No.of isolates	Isolation frequency (%)
32	Kaboon	1	3.3
66	MDRK, Mexico 222,	2	10.0
820	Perry marrow, Widusa, Kaboon, TO, TU	1	3.3
776	Cornell 49-242, TO, TU	1	3.3
944	Widusa, Kaboon, PI 207262, TO, TU	1	3.3
3842	MDRK, PI 207262, TO, AB 136, G2333	1	3.3
30	MDRK, Perry marrow, Cornell 49-242, Widusa	1	3.3
55	Michelite, MDRK, Perry marrow, Widusa, Kaboon	1	3.3
129	Michelite, PI 207262	2	6.7
524	Perry marrow, Cornell 49-242, TU	3	10.0
140	Perry marrow, Cornell 49-242, PI 207262	1	3.3
1092	Perry marrow, Mexico 222, AB 136	1	3.3
2	MDRK	1	3.3
27	Michelite, MDRK, Cornell 49-242, Widusa	1	3.3
267	Michelite, MDRK, Cornell 49-242, TO	2	6.7
3088	Widusa, AB 136, G2333	1	3.3
329	Michelite, Cornell 49-242, Mexico 222, TO	1	3.3
166	MDRK, Perry marrow, Kaboon, PI 207262	1	3.3
257	Michelite, TO	1	3.3
274	MDRK, Widusa, TO	1	3.3
1804	Perry marrow, Cornell 49-242, TO, TU, AB 136	1	3.3
344	Cornell 49-242, Widusa, Mexico 222, TO	1	3.3
534	MDRK, Perry marrow, Widusa, TU	1	3.3
2056	Cornell 49-242, G2333	1	3.3
3210	MDRK, Cornell 49-242, PI 207262, AB 136, G2333	1	3.3

Susceptibility and Resistance of Differential Cultivars to C. *lindemuthianum* Isolates

In this study, the resistance index (RI) of the 12 differential cultivars to 30 isolates of *C. lindemuthianum* identified in the present study was also calculated. Differential cultivar G2333 has the highest resistant index (86.7%), followed by Kaboon, AB136 and Mexico 222, whereby all had 83.3% as shown in Figure 4. The least resistant index was observed on the differential cultivar Cornell 49-242 (53.3%) and Michigan Dark Red Kidney (60.7%). This shows that the genes present in the two cultivars cannot succumb to most races from the study area. The differential cultivar G2333 showed the highest number of resistant reactions, thus few susceptible reactions followed by cultivars AB 136, Mexico 222, TU, Kaboon and PI 207262 respectively, as shown in Figure 4. The differential cultivars with the highest number of susceptible reactions were Cornell 49-242, Michigan Dark Red Kidney, Perry marrow and TO respectively, as shown in Figure 4.

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Figure 4: Resistance Index of 12 Common Bean Differential Cultivars to 30 Races Characterised in the Present Study

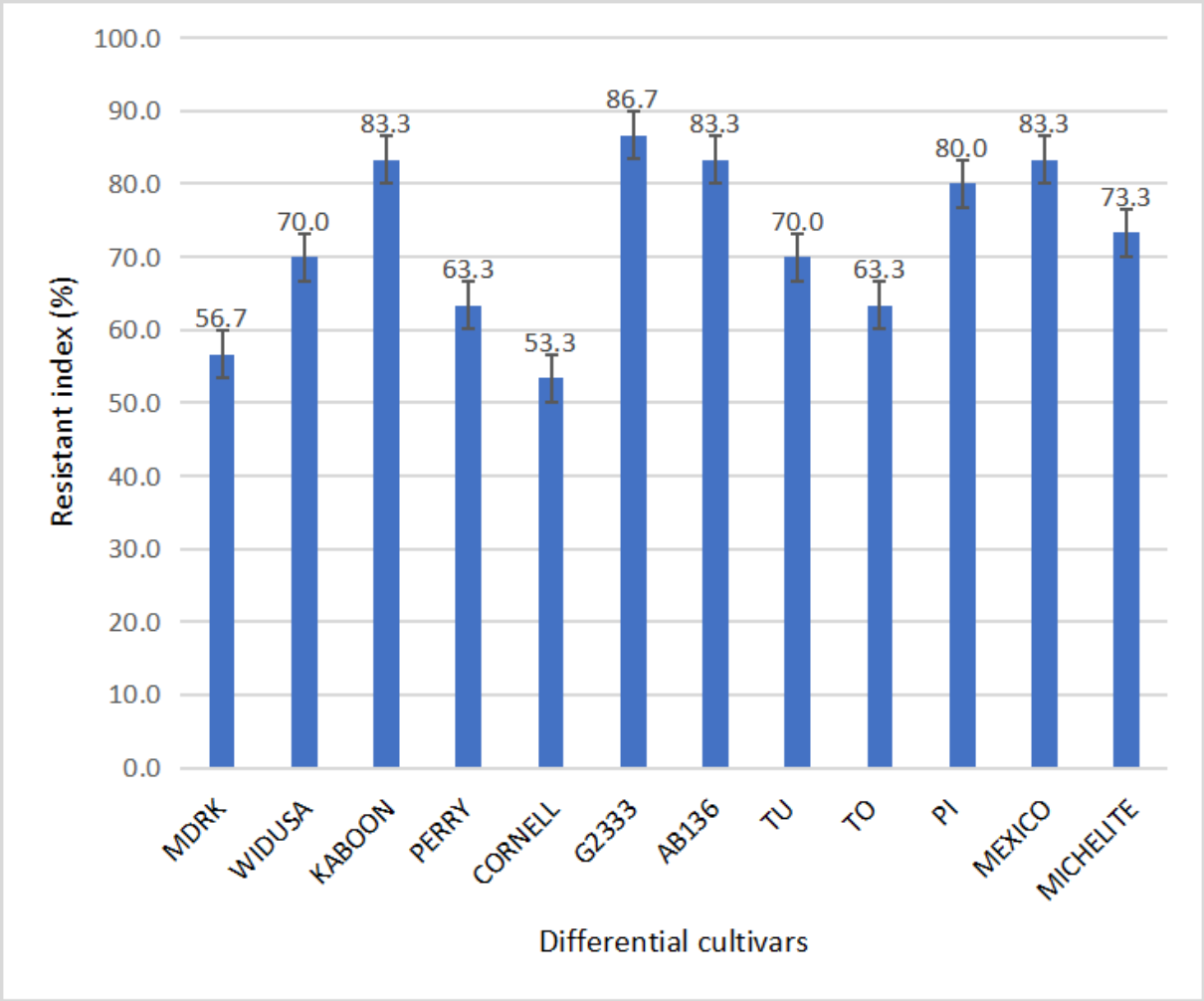

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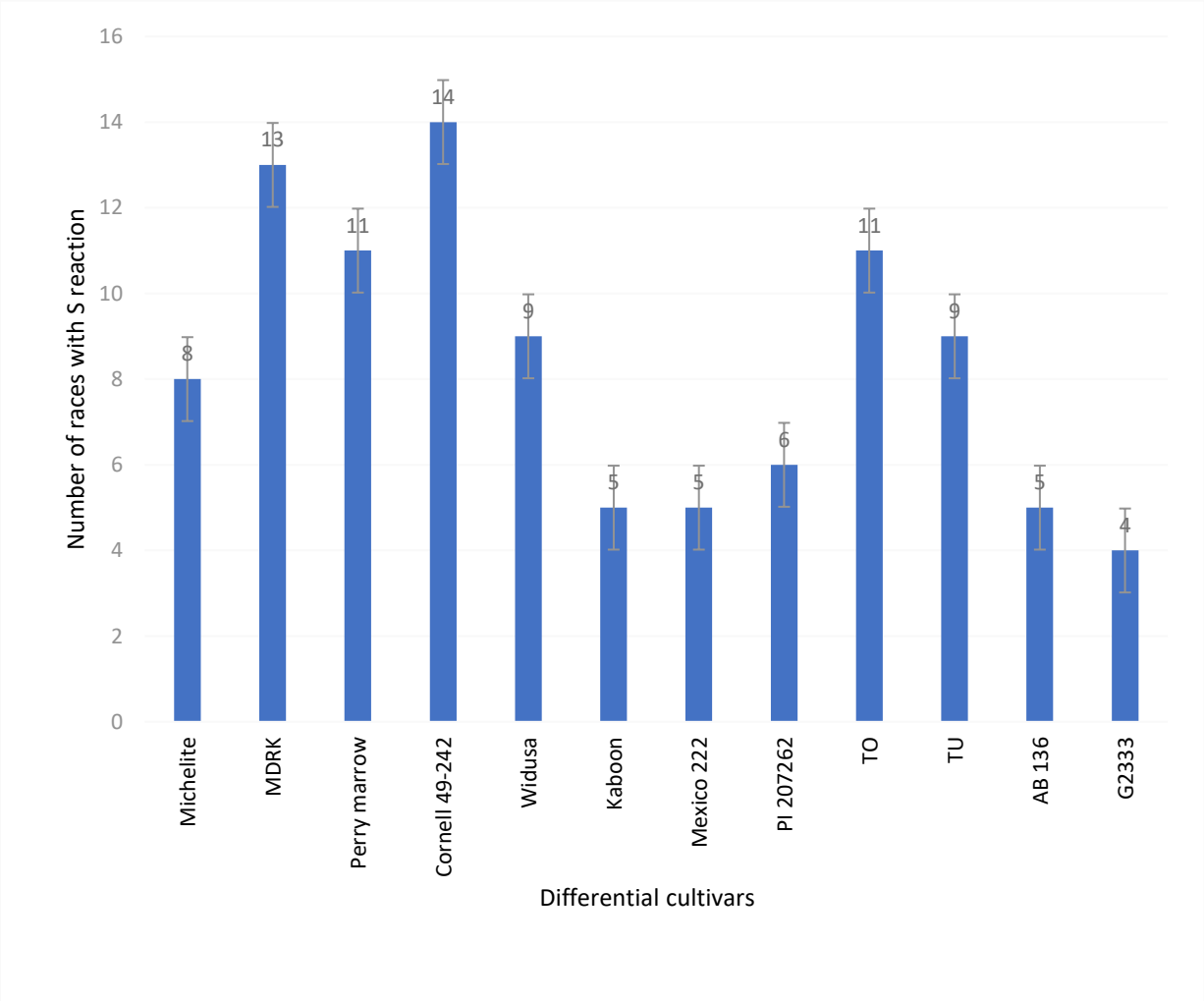
Figure 5: Number of C. *lindemuthianum* Isolates with Susceptible Reactions



DISCUSSION

In this study, 30 Anthracnose isolates from 5 major bean-growing regions in Tanzania were characterised into 25 distinct physiological races, and the race structure of the pathogen was elucidated. The observed variation of *C. lindemuthianum* in Tanzania is consistent with the previous study that reported significant diversity of *C. lindemuthianum* in Tanzania (Mpeguzi *et al*., 2020). There has been a significant shift in the race structure in Tanzania since the two studies by Ansari *et al*. (2004) and Mpeguzi *et al*. (2020). The introduction of new races of *C. lindemuthianum* is contributed by seed exchange, the finding of previously undetected races, or the evolution of races, which could all be responsible for the observed variations in race structure throughout time. Furthermore, there have been changes in common bean varieties grown in the five regions where diseased samples were collected. This change in the varieties could potentially contribute to a change in the race structure in Tanzania.

The characterised races in this study were distributed differently across the five bean growing regions that included Arusha, Kilimanjaro, Kigoma, Morogoro and Njombe. However, some characterised isolates from different regions had the same race number. This shows that some common bean seeds are sold and used across regions, which makes the *C. lindemuthianum* races distributed in

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different regions. For instance, race 524 was characterised from isolates collected from Arusha, Kilimanjaro and Morogoro and race 129 was characterised from isolates collected from Kigoma and Morogoro. Furthermore, some races reported in the current study have been characterised in Brazil, for example, races 2, 27 and 32 (Martiniano-Souza *et al*., 2021). Race 55 has been reported in Kenya and Uganda (Nunes *et al*.,2021).

The prevalence of the same races in the different regions, for instance, 524, implies that the previously resistant bean varieties may no longer be effective, thus creating a need for region-specific breeding programs. Moreover, some regions had more virulent this could have been caused by host selection pressure due to farmers repeatedly planting the same resistant bean varieties, leading to an increase in the selection pressure promoting survival of more aggressive races.

The distribution of *Colletotrichum lindemuthianum* races across different regions in Tanzania has several important implications in disease management, as it enables the farmers and breeders in the selection of resistant cultivars. For instance, knowing races that are prevalent in a particular region allows farmers and breeders to select bean cultivars with resistance to those specific races. This is crucial for effective disease management and minimising yield losses. The differences in the pathogen races between these regions reflect the differences in the common bean germplasm used and the agricultural practices employed in different regions.

Virulence of *Colletotrichum lindemuthianum* races is essential for understanding the diversity and adaptability of the pathogen population in bean-growing regions. This study shows that resistance genes in common bean cultivars are still effective and have been overcome. This information is critical for guiding breeding programs toward developing durable, broad-spectrum resistant varieties. Knowledge of virulence patterns also supports early detection of emerging aggressive races that could cause severe yield losses. Additionally, it informs the strategic deployment of resistant cultivars to minimise disease pressure in the field. Monitoring virulence contributes to improved disease management and sustainable bean production systems. Overall, it enhances food security by reducing the risk of widespread anthracnose outbreaks.

Furthermore, isolation frequency helps in identifying the distribution and prevalence of different *C. lindemuthianum* strains within a population. Identifying the most frequently isolated races reveals which strains are currently prevalent in a given area. For instance, in this study, races 66 and 524 were the most prevalent, and genes to that race are being overcome. Therefore, those genes should not be deployed in that area race 2, 129, 166, 274, 344, 524, 776,944 characterized in this study, also appeared in the previous study conducted in Tanzania (Mpeguzi *et al*., 2020). This indicates that some races of *Colletotrichum lindemuthianum* have not changed since the last study conducted in Tanzania. The number of different races identified indicates the diversity of the pathogen population. High diversity can make disease management more challenging, as it increases the likelihood of new, more virulent races emerging. Identification of races that affected both Andean and Middle American races shows that it is important to develop common bean cultivars that involve gene pyramiding.

The differences in the pathogen populations between these regions reflect the differences in common bean germplasm used and the agricultural practices employed in different agroecological zones (Mwesigwa, 2009). Comparing isolation frequency data over time can reveal how pathogen populations are evolving and help track the emergence of new races and the spread of existing ones. For example, a race that was previously rare may become dominant over time, showing the pathogen's ability to evolve. Therefore, the distribution and dynamics of *C. lindemuthianum*

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races is essential for studying the epidemiology of anthracnose, and this can help predict disease outbreaks and develop strategies to prevent them.

Resistant index implies that these cultivars possess genes that confer broad-spectrum resistance to *diverse C. lindemuthianum* races in Tanzania and can be used in future bean breeding programs. A high resistance index indicates that a cultivar is highly resistant to a particular race of the pathogen. This information is crucial for farmers and breeders to select appropriate cultivars for specific regions where those races are prevalent. By using cultivars with high resistance indices, farmers can reduce disease incidence and severity, leading to improved yields and reduced reliance on fungicides. In this study, the cultivars AB 136, Kaboon and G2333 had a high resistant index, meaning they have avirulent genes for most virulent genes of *C. lindemuthianum*. This aligns with another study, Keterew *et al*. (2018), whereby the differential cultivar AB 136 showed promising results. Therefore, its gene can be successfully incorporated into the commercial cultivars planted.

Most of the common beans grown in Africa are large-seeded market class of the Andean gene pool. Based on the pathogen-host co-evolution of the Andean and Middle American gene pools theory, it is anticipated that the races that are most likely to cause anthracnose infections in the majority of African bean production systems are those that can overcome the *Co-1* (Andean) resistance locus. The cultivars Cornell 49-242, MDRK, Perry marrow, and TO, respectively had the lowest number of resistant reactions. This implies that the resistance genes they carry are less broad-spectrum. However, these genes could still be useful in breeding programs targeting resistance gene pyramiding for broad-spectrum resistance or specific resistance for certain races.

CONCLUSION

This study has reported pathogenic and genetic variability of *C. lindemuthianum* in Tanzania to understand the virulence and variability of *C. lindemuthianum*. The physiological variation in *C. lindemuthianum* was revealed on the basis of 28 pathotypes, which were defined by standard bean differential cultivars. The differential cultivar G2333 showed strong resistance to most of the isolates and can be used as a potential source of resistance in the breeding program. The three common bean differential cultivars, Kaboon, AB136 and Mexico are promising sources of resistance to be used as alternative parental lines for developing anthracnose resistance in the study area. This is because the particular differential cultivars have avirulent genes that overcame most of the *C. lindemuthianum* genes. Understanding the distribution of Avirulent genes in *C. lindemuthianum* populations helps guide the deployment of resistant genes in common bean. Therefore, by matching R genes to the prevalent avirulent genes in a particular region, breeders can maximise the effectiveness of resistance and prevent the pathogen from overcoming the resistance.

Overall, this study generated useful information for designing anthracnose management strategies and for the selection and development of resistant common bean varieties that can help farmers increase their income by producing disease-free and high-yielding varieties resistant to *C. lindemuthianum***.**

REFERENCES

Ansari, K. I., Palacios, N., Araya, C., Langin, T., Egan, D., & Doohan, F. M. (2004). Pathogenic and genetic variability among *Colletotrichum lindemuthianum* isolates of different geographic origins. *Plant pathology*, *53*(5), 635-642.

Balardin, R. S., Smith, J. J., & Kelly, J. D. (1999). Ribosomal DNA polymorphism in

 This work is licensed under a Creative Commons Attribution 4.0 International License.

Article DOI : https://doi.org/10.37284/eajab.9.2.4903

C*olletotrichum lindemuthianum.* Mycological Research, 103(7), 841–848.

CIAT (International Center for Tropical Agriculture) (1997) Bean program annual report 1995. Working document No. 163.Centro Internacional de Agriculture Tropical Colombia.

Darben, L. M., Gonela, A., Elias, H. T., da Silva, C. R., & Pastre, H. H. (2017). Common bean germplasm resistant to races 73 and 2047 of *Colletotrichum lindemuthianum*. *African Journal of Biotechnology*, *16*(19), 1142-1149.

de Lima Castro, S. A., Gonçalves-Vidigal, M. C., Gilio, T. A. S., Lacanallo, G. F., Valentini, G., da Silva Ramos Martins, V., ... & Pastor-Corrales, M. A. (2017). Genetics and mapping of a new anthracnose resistance locus in Andean common bean Paloma. *BMC Genomics*, *18*, 1-12.

Gezahegn, A., Chala, A., & Ayana, G. (2021). Physiological Races of Colletotrichum lindemuthianum from South and Central Ethiopia. *J Plant Pathol Res*, *3*(1), 17-23.

Harlapur, S.I., Kulkarni, M.S., Yashoda, H. and Shrikant, K. (2007). Variability in Exserohilum turcicum (Pass) Leonard and Suggs, causal agent of turcicum leaf blight of maize. Karnataka Journal of Agriculture Science, 20(3), 665-666.

Hughes, G., Madden, L. V., & Munkvold, G. P. (1996). Cluster sampling for disease incidence data. *Phytopathology*, *86*(2), 132-137.

Keterew, Y., Dawit, W., Selvaraj, T., & Lencho, A. (2018). Physiological characterization and evaluation of common bean genotypes against anthracnose (Colletotrichum lindemuthianum (Sacc. and Magnus) Lams-Scriber) races in West Shewa, Ethiopia. *Int J Life Sci*, *6*(3), 744-754.

Kiryowa, M. J., Ebinu, A., Kyaligonza, V., Nkalubo, S. T., Paparu, P., Mukankusi, C., & Tukamuhabwa, P. (2017). Pathogenic variation of Colletotrichum lindemuthianum causing anthracnose of beans (Phaseolus vulgaris) in Uganda. International Journal of Phytopathology, 5(3), 89-98.

Lisciani, S., Marconi, S., Le Donne, C., Camilli, E., Aguzzi, A., Gabrielli, P., ... & Ferrari, M. (2024). Legumes and common common bean in sustainable diets: nutritional quality, environmental benefits, spread and use in food preparations. Frontiers in Nutrition, 11, 1385232.

Mahuku, G. S., & Riascos, J. J. (2004). Virulence and molecular diversity within *Colletotrichum lindemuthianum* isolates from Andean and Mesoamerican bean varieties and regions. European Journal of Plant Pathology, 110, 253-263.

Martiniano-Souza, M. D. C., Gonçalves-Vidigal, M. C., da Costa, A. F., Vidigal Filho, P. S., Martins, V. D. S. R., Vaz Bisneta, M., ... & Valentini, G. (2021). Virulence and genetic diversity of Colletotrichum lindemuthianum and resistance of local common bean germplasm to anthracnose in Pernambuco State, Brazil. *European Journal of Plant Pathology*, *159*, 727-740.

Mohammed, A. (2013). An overview of distribution, biology and the management of common bean anthracnose. *J Plant Pathol Microbiol*, *4*(8), 1-6.

Mpeguzi, M., Susan, N. M., Mabagala, R., & Chilagane, L. A. (2020). Races of Colletotrichum lindemuthianum (Sacc. Magnus) Briosi Cavara in major bean growing regions in Tanzania. *African Journal of Plant Science*, *14*(8), 308-314

Mwesigwa J. B., 2009. Diversity of Colletotrichum lindemuthianum and reaction of common bean germplasm to anthracnose disease. MSc.thesis. Makerere University, Uganda. pp96.

Nakei, M. D., Venkataramana, P. B., & Ndakidemi, P. A. (2023). Preliminary symbiotic performance of indigenous soybean (Glycine max)-nodulating

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Article DOI : https://doi.org/10.37284/eajab.9.2.4903

rhizobia from agricultural soils of Tanzania. *Frontiers in Sustainable Food Systems*, *6*, 1085843.

Nalupya, K. (2021). *Evaluation of the ciat phaseoulus core collection for resistance to anthracnose* (Doctoral dissertation, The University of Zambia).

Nunes MPB, MC Gonçalves-Vidiga, VSR Martins, LFS Xavier, G Valentini, M Vaz Bisneta, PSF Vidiga (2021). Relationship of Colletotrichum lindemuthianum races and resistance loci in the Phaseolus vulgaris L. genome. Crop Sci 61:3877–3893

Padder, B. A., Sharma, P. N., Awale, H. E., & Kelly, J. D. (2017). Colletotrichum lindemuthianum, the causal agent of bean anthracnose. *Journal of Plant Pathology*, 317-330.

Pastor-Corrales, M. A., Otoya, M. M., Molina, A., & Singh, S. P. (1995). Resistance to *Colletotrichum lindemuthianum* isolates from middle America and Andean South America in different common bean races. *Plant Disease, 79*, 63–67

Stewart, E., Liu, Z., Crous, P. W., & Szabo, L. J. (1999). Phylogenetic relationships among some cercosporoid anamorphs of *Mycosphaerella* based on rDNA sequence analysis. *Mycological Research, 103*(11), 1491–1499.

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