African Crop Science Journal, Vol. 27, No. 4, pp. 585 - 612
ISSN 1021-9730/2019 $4.00

DOI: https://dx.doi.org/10.4314/acsj.v27i4.4

VALIDATION OF EFFECTIVENESS MARKER-ASSISTED GAMETE SELECTION FOR MULTIPLE DISEASE RESISTANCE IN COMMON BEAN

M.J. MONDO[1, 2], P.M. KIMANI[1] and R.D. NARLA[1]

[1]Department of Plant Science and Crop Protection, University of Nairobi, P. O. Box 29053-00625, Nairobi, Kenya
[2]Faculty of Agriculture and Environmental Sciences, Université Evangélique en Afrique (UEA), P. O. Box 3323, Bukavu, Democratic Republic of Congo
Corresponding author: mondo.mubalama@yahoo.fr, mondo.mubalama@uea.ac.cd

(*Received 23 January 2019; accepted 30 October 2019*)

ABSTRACT

Plant breeding is the most cost-effective, practical and environmentally friendly strategy for reducing losses associated with bean diseases, especially in low-input agricultural systems because no additional investment is required from farmers. However, incorporating resistance to one pathogen may not result in a significant change because several diseases co-infection beans at the farm level. Consequently, breeding varieties with multiple disease resistance is a more appropriate, reliable and sustainable approach. In such context, gamete selection is the more appropriate breeding method because it allows simultaneous selection for multiple traits; though as originally proposed and validated, it is largely based on phenotypic evaluation for agronomic traits, which leads to delay in variety development and strong dependence on erratic weather conditions. The objective of this study was to validate 26 $F_{1.8}$ elite bean lines selected for resistance to angular leaf spot (ALS), anthracnose, root rots, common bacterial blight (CBB) and bean common mosaic virus (BCMV), from inter-racial and inter-gene pool populations developed using molecular markers on the gamete selection method in early generations. Pathogens were isolated from diseased plants collected from various locations in central Kenya, multiplied on appropriate media and used to inoculate the test lines in a greenhouse at Kabete Field Station, University of Nairobi. Data on disease incidence and severity were collected at 14, 21, 28th days after inoculation, using the 1-9 CIAT scale; except for the root rot experiments for which data were recorded once at 21st day after seedling emergence. Results showed that five of the 26 elite lines possessed multiple resistance to five pathogens, eight to four pathogens, nine to three pathogens, three to two pathogens and one was resistant to one pathogen. This implied that markers, used in early generations, were effective in the identification and transfer of resistance genes to susceptible commercial varieties. However, there were no significant correlations in the reaction of tested genotypes to pathogens in this study, except between BCMV and ALS (r=0.3942*). This suggests that resistance genes are in different chromosomes and are assorted independently. The presence of genotypes with multiple disease resistance among test elite lines, confirms the effectiveness

of inter-racial crosses and marker-assisted gamete selection to concurrently improve the resistance to common bean major diseases in Eastern Africa.

Key Words: Inter-racial elite lines, Kenya*, Phaseolus vulgaris*

RÉSUMÉ

L'amélioration génétique des plantes est la stratégie la moins coûteuse, la plus pratique et la plus respectueuse de l'environnement dans la réduction des pertes associées aux maladies du haricot, en particulier dans les systèmes agricoles à faible usage d'intrants. Ceci car aucun investissement supplémentaire n'est requis de la part des agriculteurs. Cependant, l'incorporation de la résistance à un agent pathogène ne pourrait entraîner de changement significatif, car plusieurs maladies attaquent simultanément le haricot. Par conséquent, le développement de variétés présentant une résistance multiple aux maladies constitue une approche plus appropriée, fiable et durable. Dans ce contexte, la sélection des gamètes est la méthode d'amélioration génétique la plus appropriée car elle permet la sélection simultanée de plusieurs caractères ; bien que, telle que proposée et validée à l'origine, elle repose en grande partie sur une évaluation phénotypique des caractères agronomiques. Ceci retarde ainsi le développement de la variété et entraîne une forte dépendance à des conditions météorologiques, souvent irrégulières. L'objectif de cette étude était de valider la huitième génération ($F_{1.8}$) de 26 lignées élites de haricot sélectionnées pour leur résistance à la maladie des taches angulaires, à l'anthracnose, à la fonte des semis, à la bactériose commune du haricot et à la mosaïque commune du haricot. Ces lignées viennent des populations interraciales et inter-géniques de haricot, développées en utilisant des marqueurs moléculaires sur la méthode de sélection des gamètes dans leurs premières générations. Des agents phytopathogènes ont été isolés à partir de plantes malades recueillies à divers endroits dans la partie centrale du Kenya, multipliés sur des milieux de culture appropriés et utilisés par la suite pour inoculer les lignées testées sous une serre, dans le champ expérimental de l'Université de Nairobi situé à Kabete. Les données sur l'incidence et la sévérité des maladies ont été enregistrées aux 14, 21 et 28ème jours après l'inoculation, à l'aide de l'échelle de CIAT allant de 1à 9; à l'exception des expériences sur les fontes de semis pour lesquelles les données étaient enregistrées une seule fois, au 21ème jour après la levée des plantules. Les résultats ont montré que cinq des 26 lignées élites présentaient une résistance multiple à cinq agents pathogènes, huit à quatre agents pathogènes, neuf à trois agents pathogènes, trois à deux agents pathogènes et une était résistante à un agent pathogène. Cela démontrait que les marqueurs moléculaires, utilisés dans les premières générations, étaient efficaces dans l'identification et le transfert de gènes de résistance à des variétés commerciales sensibles. Cependant, il n'y avait pas de corrélations significatives dans la réaction des génotypes aux agents pathogènes, sauf entre la mosaïque commune du haricot et la maladie des taches angulaires (r = 0,3942*). Ceci suggère que les gènes de résistance sont dans différents chromosomes et assortis indépendamment. La présence de génotypes, présentant une résistance multiple aux maladies parmi les lignées élites testées, confirme l'efficacité des croisements interraciaux et de la méthode de sélection de gamètes assistée par marqueurs dans l'amélioration simultanée de la résistance aux principales maladies du haricot commun en Afrique de l'Est.

Mots Clés: Lignées élites interraciales, Kenya, *Phaseolus vulgaris*

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is the most important legume crop for human consumption worldwide, contributing protein, complex carbohydrates, dietary fiber, isoflavones and micronutrients (iron, phosphorus, zinc) to diets of large millions of people, especially in Africa and Latin America (Broughton *et al.,* 2003; Beebe *et al.,* 2013).

In addition to its nutritional value, the common bean is also an important source of income for the small-scale and resource-poor farmers of sub-Saharan Africa (CGIAR, 2017). Also, common bean has multiple health benefits; it reduces the risk of chronic diseases such as diabetes, heart disease and cancer (Winham *et al.,* 2018).

Eastern and Central African countries are the major producers and consumers of common bean in Africa, where it contributes up to 25% of total caloric intake and 45% of total dietary protein and, thus, making it the highest level of contribution of protein in the world (Kilimo Trust, 2012; Alladassi *et al.,* 2018). Kenya, Tanzania and Uganda are the leading producers in Africa (Beebe *et al.,* 2013; FAO, 2018). However, Kenya has been a net bean importer for the last two decades because demand exceeds production (Kimani *et al.,* 2005a).

Despite the importance of common bean in Eastern and Central Africa, its productivity is still among the lowest in the world, with an average seed yield of 0.5 t ha^{-1} (FAO, 2018); while potential yields range from 1 to 3 t ha^{-1} for bush genotypes, and could be as high as 5 t ha^{-1} for climbers (Ronner *et al.,* 2018). Many constraints are responsible for poor performance of common bean in the region. Major constraints include drought stress, low soil fertility, plant diseases and pests, poor adaptation of introduced varieties to local conditions, and socio-economic factors such as low and untimely access to external inputs; and poor farming practices (Wortmann *et al.*, 1998; Kimani *et al.,* 2005b).

The major diseases constraining common bean productivity in Eastern and Central Africa include angular leaf spot (ALS) caused by *Pseudocercospora griseola* (Sacc.) (Ddamulira *et al.,* 2014; Leitich *et al.,* 2016) and anthracnose (*Colletotrichum lindemuthianum* (Sacc. and Magn.) (Kiryowa *et al.,* 2016). Other damaging diseases are root rots (*Pythium spp*., *Fusarium spp., Sclerotium rolfsii* and *Rhizoctonia solani*) (Nzungize *et al.,* 2011a; Obala *et al.,* 2012; Buruchara *et al.,* 2015; Mukankusi *et al.,* 2018), bean common mosaic and necrotic viruses (BCMV/ BCMNV) (Mwaipopo *et al.,* 2017), and common bacterial blight (CBB) caused by *Xanthomonas axonopodis* pv. *phaseoli* (Alladassi *et al.,* 2018). These diseases cause severe losses of seed yield and quality of common bean, ranging from 20% to as high as 80 to 100% (Singh and Schwartz, 2010). Wortmann *et al.* (1998) estimated the annual production losses in Eastern Africa caused by ALS at 281,300 t; anthracnose at 247,400 t, root rot at 179,800 t, CBB at 145,900 t and BCMV at 144,600 t.

Several approaches have been used to control those common bean diseases, such as combinations of cultural and chemical controls; but are occasionally found to be ineffective to many diseases (Okii *et al.,* 2017). In addition to negative environmental impacts of chemicals, associated costs are not practical for the widespread low-input systems; and therefore, breeding for resistance is the most cost-effective and environmentally friendly approach for resource-poor farmers of Eastern and Central Africa (Odogwu *et al*., 2017), since there is often no additional cost. This approach can greatly reduce the need for chemicals, hence increasing returns on farmers' investment.

Okii *et al.* (2017) showed that multiple pathogen co-infections on common beans are responsible for complete crop losses in susceptible bean varieties. This suggests that common bean breeding for disease resistance should target multiple pathogens simultaneously, by pyramiding resistance genes in a single genotype for a broader and durable resistance. Because several diseases normally occur in a particular production environment, incorporating resistance to a single disease will not result in significant changes (Singh, 1994; Kimani *et al.,* 2005b).

Development of improved dry bean varieties in Eastern and Central Africa faces four key challenges. First, is the occurrence

of new races and strains of disease pathogens, such as ALS, anthracnose, root rots, and BCMV (Leitich *et al.,* 2016; Mwaipopo *et al.,* 2017). In addition, there is insufficient identification and deployment of new sources of resistance to the emerging pathotypes (Ddamulira *et al.,* 2014; Mukankusi *et al.,* 2018), as well as a narrow genetic base within existing breeding populations, especially for grain yield potential and disease resistance. This threatens progress towards improvement for these traits (Kimani *et al.,* 2005b; Asfaw *et al.,* 2009). Finally, there is also lower efficiency of breeding methodology (Kimani *et al.,* 2005b) leading to high failure rate and longer duration in new varieties development process. These four issues listed above were the main focus of the marker-assisted breeding programme at the University of Nairobi since 2009. The programme initiated studies to determine whether marker-assisted gamete selection could be effective in pyramiding genes for resistance to bean major diseases in Eastern and Central Africa (mainly ALS, anthracnose, CBB, BCMV and root rot); and introduce these genes into susceptible, but popular large- and small-seeded bean varieties (Musyimi, 2014; Njuguna, 2014; Mondo *et al.,* 2018).

Thirty-two inter-racial and inter-gene pool populations were developed from crosses among Middle American (Mesoamerican) and Andean gene pool cultivars to broaden the genetic base of commercial cultivars, and take advantage of attributes of both gene pools. In addition to high yield potential of Middle American cultivars, they are resistant to major diseases of the Andean gene pool counterparts, and possess genes for drought resistance; while the Andean cultivars are the most preferred in Africa for their seed quality, and thus fetch higher prices in local markets (Welsh *et al.,* 1995; Singh *et al.,* 2002; Sichilima *et al.,* 2016).

This, therefore, justified the necessity of inter-racial crosses in developing breeding populations. To shorten the breeding programme and increase its efficiency, the marker-assisted gamete selection method was followed as a possible improvement of the original phenotypic gamete selection developed by Singh (1994). Based on the objective of the breeding programme in the present study, gamete selection is the more appropriate breeding method because it allows simultaneous selection for multiple traits (Beaver and Osorno, 2009) and screening and selection of desirable traits in early generations, and therefore, helps to avoid wastage of scarce resources and time by advancing unpromising genotypes as it is the case for most bean breeding methods.

This study, which is a continuation of the above described breeding programme, aimed to validating the multiple disease resistance of $F_{1.8}$ elite bean lines selected in early generations by combining molecular markers to the gamete selection on populations from inter-racial crosses.

MATERIALS AND METHODS

Study site. This study was carried out at Kabete Field Station of the University of Nairobi, which is located in Kenya at coordinates 01°15' S (latitude); 036°44' E (longitude) and at an altitude of approximately 1820 m above sea level. The station receives an average rainfall of 1059 mm annually, spread over two seasons. It experiences mean maximum and minimum temperatures of 22.5 °C and 12.3 °C, respectively. Soils are well drained, very deep, dark reddish brown, friable clay with acid humic topsoil, humic nitisols (Jaetzold *et al.,* 2006). The pH is about 5.0 to 5.4 and a mean sunshine of 6.6 hours per day.

Plant materials. Plant materials used for this study were 26 elite $F_{1.8}$ lines selected for seed yield and seed quality from multisite testing, conducted during 2017 short rainy season in three agro-ecological conditions of central Kenya (low, medium and high altitudes). Additive main-effects, and multiplicative

interaction (AMMI) analysis for genotype (G) and genotype x environment interactions (GE) was used to identify the 26 elite lines from five market classes. The major characteristics of these lines are presented in Table 1. In addition to these elite lines, 10 parental cultivars used in population development were included as checks.

During population development, Mex54 and G10909 were used as sources of resistance to ALS; G2333 for anthracnose, RWR719 and AND1062 for root rots and BRB191 for BCMV. Commercial check varieties included GLP92 (*Mwitemania*), GLP585 (*Wairimu*), KATB9 and KATB1 which are susceptible parents but with high yield potential, market-demanded traits and good adaptation to agro-ecological conditions of Eastern Africa. Major characteristics of these parental genotypes are described in Table 2.

Population development was done from 2009 using the gamete selection breeding method, as first described by Singh (1994). Development of male gametes involved making single crosses in the first round of crossing. The single crosses were subsequently combined into double crosses. Male gametes with requisite resistance genes were then identified using markers SAB-3 for anthracnose (Garzon *et al.,* 2008); SH-13 for ALS (Mahuku *et al.,* 2011); SW-13 for BCMV (Sharma *et al.,* 2008) and PYAA-19 for *Pythium* root rot (Namayanja *et al.,* 2014). These male gametes were, thereafter, used to construct the F_1 by the final cross of the double-cross gamete to the commercial varieties (Singh, 1994; Mondo *et al.,* 2018). Selection also started in F_1 instead of F_2, in normal cases.

A total of 16 populations were developed. The segregating F_1 and $F_{1.2}$ populations were then evaluated for agronomic attributes, and tested for resistance to target diseases under natural disease infestation in the field at Kabete and Tigoni in 2011 and 2012 in Kenya. Molecular markers were used for screening the male gametes and the segregating F_1. From F1.2 to F1.6 generations, bean progenies were advanced following gamete selection procedure as modified by Mondo *et al.* (2018). This was conducted during the period from 2013 to 2016.

A multisite testing of $F_{1.7}$ bean lines grouped in five major market classes, was conducted in 2017-2018 short rainy season in three agro-ecological conditions representing the low-, medium- and high-altitude environments. Lines used in this study were the $F_{1.8}$ elite lines (with high yield potential) selected from that multisite evaluation.

Experimental procedures

Pathogen isolation, inoculum preparation and plant inoculation. Common bean plant parts (leaves, roots, stems or pods) infected by anthracnose, ALS, root rot, CBB and BCMV were collected from various areas in central Kenya. The collection areas were selected based on previous country-wide surveys (Omunyin *et al.*, 1995; Mwang'ombe *et al.,* 2007; Musyimi, 2014; Njuguna, 2014), which identified regions with the highest prevalence for each of those pathogens. These areas included Kabete (Nairobi County), Tigoni and Limuru (Kiambu County), Mwea (Kirinyaga County) and Naivasha (Nakuru County). Most of the diseased plant samples were collected during the 2017 short rainy season (from October 2017 to February 2018). Specific pathogen isolation and procedures are described below.

Anthracnose. *Collectotrichum lindemuthianum* was isolated from diseased bean leaves following Sicard *et al.* (1997) procedure. The concentration of the inoculum was adjusted to 2×10^6 conidia per ml using a haemocytometer for pathogens as suggested by Bigirimana and Hofte (2001). Twenty one-days-old seedlings were covered with polythene plastic bags to provide a humid environment, 12 hours before inoculation. The plants were then inoculated by spraying spore

TABLE 1. Characteristics of 26 elite lines used in common bean multi-disease resistance validation study in controlled environments, at Kabete Field Station, Kenya

Line	Seed colour	Growth habit	Seed size	[§]Yield (kg ha^{-1})	Recommended areas	Pedigree
KMA13-27-27	Tan red	IV	Medium	2,845	Low- and highland	KATB1 x Mex54 / G2333 // AND1062 / BRB191
KMA13-28-5	Tan red	IV	Medium	1,947	Lowland	KATB1 x Mex54 / G2333 // RWR719 / BRB191
KMA13-28-13	Tan red	IV	Medium	1,869	Midland	KATB1 x Mex54 / G2333 // RWR719 / BRB191
KMA13-31-62	Tan brown	III	Medium	1,989	Lowland	KATB9 x Mex54 / G2333 // AND1062 / BRB191
KMA13-27-12	Black	II	Medium	2,044	Midland	KATB1 x Mex54 / G2333 // AND1062 / BRB191
KMA13-28-21	Black	III	Medium	3,718	Mid- and highland	KATB1 x Mex54 / G2333 // RWR719 / BRB191
KMA13-21-20	Yellow	IV	Medium	2,329	Mid- and highland	GLP92 x G10909 / G2333 // AND1062 / BRB191
KMA13-21-10	Pinto	III	Medium	2,285	Lowland	GLP92 x G10909 / G2333 // AND1062 / BRB191
KMA13-22-21	Pinto	III	Medium	2,748	Low-, mid-, highland	GLP92 x G10909 / G2333 // RWR719 / BRB191
KMA13-22-30	Pinto	III	Medium	2,726	Highland	GLP92 x G10909 / G2333 // RWR719 / BRB191
KMA13-23-13	Pinto	III	Medium	2,031	Midland	GLP92 x Mex54 / G2333 // AND1062 / BRB191
KMA13-23-22	Pinto	III	Medium	2,360	Highland	GLP92 x Mex54 / G2333 // AND1062 / BRB191
KMA13-24-7	Pinto	III	Medium	2,136	Highland	GLP92 x Mex54 / G2333 // AND1062 / BRB191
KMA13-26-32	Red kidney	III	Large	2,370	Lowland	KATB1 x G10909 / G2333 // RWR719 / BRB191
KMA13-27-31	Red kidney	III	Large	2,136	Lowland	KATB1 x Mex54 / G2333 // AND1062 / BRB191
KMA13-28-2	Red kidney	II	Large	2,318	Highland	KATB1 x Mex54 / G2333 // RWR719 / BRB191
KMA13-30-22	Red kidney	III	Medium	3,226	Mid- and highland	KATB9 x G10909 / G2333 // RWR719 / BRB191
KMA13-21-11	Red kidney	II	Large	2,448	Midland	GLP92 x G10909 / G2333 // AND1062 / BRB191
KMA13-17-25	Red mottled	I	Large	2,038	Midland	GLP585 x G10909 / G2333 // AND1062 / BRB191
KMA13-29-21	Red mottled	II	Large	3,860	Low-, mid-, highland	KATB9 x G10909 / G2333 // AND1062 / BRB191
KMA13-29-24	Red mottled	IV	Medium	2,640	Low- and highland	KATB9 x G10909 / G2333 // AND1062 / BRB191
KMA13-17-17	Red mottled	II	Large	2,525	Midland	GLP585 x G10909 / G2333 // AND1062 / BRB191
KMA13-23-14	Small red	IV	Medium	3,022	Low- and highland	GLP92 x Mex54 / G2333 // AND1062 / BRB191
KMA13-25-9	Small red	IV	Medium	3,385	Low- and highland	KATB1 x G10909 / G2333 // AND1062/ BRB191
KMA13-30-14	Small red	III	Medium	2,787	Highland	KATB9 x G10909 / G2333 // RWR719 / BRB191
KMA13-32-28	Small red	III	Medium	2,453	Lowland	KATB9 x Mex54 / G2333 // RWR719 / BRB191

[§]Yield data is from a multi-environment evaluation at three locations during the 2017 short rainy season (Mondo *et al.,* 2019a; 2019b).

TABLE 2. Major characteristics of parental lines used for population development

Genotypes	[1]Gene pool	Seed colour	[2]Growth habit	[3]Reaction to diseases				Linked markers	Reference
				ALS	ANT	RR	BCMV		
Donor parents									
G2333	M	Red	Ⅳ	R	R	S	S	SAB-3	Garzón *et al.* (2008)
Mex54	M	Cream beige	Ⅳ	R	S	S	S	OPE4^{708}	De Queiroz *et al.* (2004)
G10909	M	Red	Ⅳ	R	S	S	S	SH13^{520}	Mahuku *et al.* (2011)
RWR719	M	Red	I	S	S	R	S	PYAA19^{800}	Buruchara *et al.* (2015)
AND1062	A	Red Kidney	I	S	S	R	S	PYAA19^{800}	Namayanja *et al.* (2014)
BRB191	A	Red Mottled	I	S	S	S	R	SW13^{690}	Sharma *et al.* (2008)
Susceptible parents									
GLP585	M	Red	I	S	S	S	S	N/A	
GLP92	M	Pinto	Ⅱ	S	S	S	S	N/A	
KATB1	M	Green	I	S	S	S	S	N/A	
KATB9	M	Red	I	S	S	S	S	N/A	

[1]A = Andean, M = Mesoamerican; [2]I = determinate, II = indeterminate bush, erect stem and branches, III = indeterminate bush with weak and prostrate stem and branches, IV = indeterminate climbing habit with weak, long and twisted stem and branches; [3]R = resistant, S = susceptible, ALS = angular leaf spot, ANT = anthracnose, BCMV = bean common mosaic virus, RR = *Pythium* root rot

suspension on the leaves evenly with a handheld atomiser. After inoculation, the plants were covered with moistened polythene bags and transferred into the greenhouse.

Angular leaf spot (ALS). The *Pseudocercospora griseola* causing the ALS was isolated from infected leaves by transferring ALS lesions on the underside of leaves on V8 agar, using an inoculating needle; then incubated and multiplied following procedures by Correa and Saettler (1987) and Wagara *et al.* (1999). Spores for inoculation were obtained by gently scraping the surface of sporulating colonies incubated for 14 days in sterile distilled water. Inoculations were done on both sides of the first and second trifoliolate leaves 21 days after planting.

Root rots. Bean plants were uprooted based on the presence of root rot-like symptoms prevailing on leaves, roots and stems. Isolation procedure described by White (1988) and modified by Nzungize *et al.* (2011b) was used. *Fusarium*, *Rhizoctonia* and *Pythium* root rots were then multiplied by plating mycelia on autoclaved millet grains (300 g) mixed with 200 ml of water in 1000 ml bottles. After two weeks of incubation under darkness and at 25 °C, a pre-sterilised soil was mixed with the infested millet at a ratio of 1:10 v/v in polyphene pots three days before planting (Buruchara *et al.,* 2015). Three weeks after emergence of the seedlings, the surviving plants were uprooted and washed with water to remove soil.

Bean common mosaic virus (BCMV). Young infected leaves of bean with distinct mosaic symptoms were collected and the standard inoculum obtained using the procedure by Verma and Gupta (2010). The plant inoculation was done following suggestions by Strausbaugh *et al.* (1999), when primary leaves were fully expanded. This corresponded with 14 days after seedling emergence.

Common bacterial blight (CBB). CBB pathogen was isolated from leaves and stems. Isolation, inoculum preparation and spraying were following procedures previously described by Harveson and Schwartz (2007). The inoculum was sprayed on plants 14 days after seedling emergence. Inoculated plants were covered with plastic bags, and placed into incubators. After four days, plants were then transferred in the greenhouse until symptom development.

Experimental design and data collection. The screening experiments for ALS, anthracnose, BCMV and CBB resistance were conducted in a greenhouse at Kabete Field Station of the University of Nairobi. Screening for resistance to *Fusarium solani* pv. *phaseoli*, *Rhizoctonia solani*, and *Pythium ultimum* root rots was conducted in an insect proof screenhouse at Kabete Field Station.

The experimental design for each trial was a randomised complete block design (RCBD), with four replications. Treatments consisted of 36 genotypes including 26 elite bean lines and 10 parents used as check varieties. A separate, but similar experiment was set for each disease in which treatments were clearly labeled and randomly arranged within the greenhouse. Each plot consisted of four pots, each containing four plants making a total of 16 plants for each genotype in a replication.

Pots were uniformly filled with pre-sterilised soils, mixed with cattle manure in compost form, and sand at a ratio of 3:1:1. As described previously in the study site section, Kabete' soils used for this experiment are well drained, very deep, dark reddish brown, friable clay with acid humic topsoil, humic nitisols and a pH ranging from 5.0 to 5.4.

Diammonium phosphate (DAP) (N 18%: P_2O_5 46%) at a rate of 80 kg ha^{-1} (12.8 g per pot) was applied at planting, in each pot. The pots were irrigated to field capacity to ensure moisture-free conditions for the study plants, except the root rot experiments which relied exclusively on rain for water. Rainfall

distribution during the study period was favourable for disease development in the insect-proof screenhouse; as mean monthly rainfall was approximately 275.7 mm from March to June 2018; while the mean temperature was 18.2 °C.

Data on disease incidence and severity were recorded at seven day intervals (14, 21 and 28 days) after inoculation for ALS, BCMV, anthracnose and CBB. Data on root rots were taken once, 21 days after seedling emergence. The disease severity was rated using a 1-9 CIAT scale: 1-3 being resistant, 3.1-6 intermediate and 6.1-9 susceptible (Schoonhoven and Pastor-Corrales, 1987; Okii *et al.,* 2017). The disease incidence was the percentage of diseased plants from the total number of plants initially inoculated.

Data analysis. GenStat 15th edition software (VSN Int., 2013) was used for analysis of variance. Fisher's protected least significant difference (LSD) was used for mean separation at 1 and 5 percent probability levels. Area under disease progression curve (AUDPC) was performed for each genotype using the midpoint rule method (Campbell and Madden, 1990) as follows:

$$\text{AUDPC} = \sum_{i=1}^{n-1}\left(\frac{y_i + y_{i+1}}{2}\right)(t_{i+1} - t_i)$$

Where:

t represents the time in days of each observation, y is disease severity at observation and, n is the number of observations.

The AUDPCs were then subjected to ANOVA to compare amounts of disease among different bean lines for each disease pathogen. The highest values corresponded to more susceptible; while the lowest values corresponded to more resistant varieties.

RESULTS

Disease severity and AUDPC. Disease severity score showed no significant ($P>0.05$) differences in the elite lines and check varieties to the three root rot pathogens (Table 3). However, genotypes reacted differently to ALS ($P<0.05$), BCMV ($P<0.01$), CBB ($P<0.01$) and anthracnose pathogen ($P<0.001$). The differences among genotypes were even highly significant ($P<0.001$) when referring to computed AUDPC values, regardless of the pathogens (Table 4).

Reaction to root rot diseases. Figure 1 presents symptoms of the three root rot pathogens as observed on susceptible genotypes during the screenhouse testing at Kabete, University of Nairobi. The *Fusarium* root rot was the most damaging on the tested materials; its incidence ranged from 43.3% on KMA13-27-31 to 96.1% on the check variety BRB191 (Table 5). Disease severity was also high, ranging from 2.8 on KMA13-27-31 to 6.9 on KMA13-17-25. KMA13-27-31, a red kidney genotype, was the only elite line which showed resistance to *Fusarium* root rot. *Rhizoctonia* root rot affected more than 50% of plants for all the genotypes, but the severity was very low (1.5 to 5.0).

The *Pythium* root rot incidence was also very high, ranging from 53.9 to 84.6% (Table 5). Severity of *Pythium* root rot varied from 2.1 on KMA13-32-28 to 5.8 on the check variety KATB1. None of the elite lines or check varieties combined concurrently, resistance to the three root rot-causing agents. However, 6 elite lines (KMA13-21-11; KMA13-23-14; KMA13-25-9; KMA13-28-5; KMA13-30-14 and KMA13-32-28) had combined resistance to *Rhizoctonia* and *Pythium* root rots; while KMA13-27-31 had concurrent resistance to *Fusarium* and *Rhizoctonia* root rots. Fortunately, more than 80% (21 of the 26) of

TABLE 3. Mean squares of incidence and severity scores for the foliar pathogens on elite bean lines at the final score (28 days after inoculation) in a greenhouse at Kabete, University of Nairobi, Kenya

Sources of variation	DF	ALS		BCMV		CBB		ANTH	
		Incidence	Severity	Incidence	Severity	Incidence	Severity	Incidence	Severity
Replication	3	5605.6	62.3	2322.2	6.1	4170.9	100.3	5.6	13.3
Genotype	35	712.4^{ns}	1.7*	1504.9^{ns}	3.1**	1235.2***	3.4**	952.1***	2.8^{ns}
Residual	35	493.5	0.78	864.6	0.92	173.0	1.0	2.1	2.4
Total	73								
Mean		28.7	2.7	75.1	3.5	50.9	3.6	28.9	2.6
$LSD_{0.05}$		45.1	1.8	59.7	1.9	26.7	2.0	2.9	59.1
CV (%)		77.5	32.9	39.2	27.8	25.9	27.5	5.0	52.1

DF = degree of freedom, $LSD_{0.05}$ = least significant difference at 5% P-value threshold, CV = coefficient of variation, ns = not significant, *, **, *** = significant at P = 0.05, 0.01 and 0.001, respectively. ALS = angular leaf spot, BCMV = bean common mosaic virus, CBB = common bacterial blight, ANTH=anthracnose

TABLE 4. Mean squares of AUDPC for the foliar pathogens on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Sources of variation	DF	ALS		BCMV		CBB		ANTH	
		Incidence	Severity	Incidence	Severity	Incidence	Severity	Incidence	Severity
Replication	3	17564.	741.1	122896.	115.0	158109.	5270.2	345	1378.1
Genotype	35	81206.***	78.8***	133628.***	364.9***	207554.***	418.4***	195616***	337.6^{ns}
Residual	35	8782.	16.6	21915.	18.8	20882.	74.5	153	274.6
Total	73								
Mean		486.5	32.2	1002	41.7	614.7	42.8	342.7	31.8
$LSD_{0.05}$		190.2	8.3	300.5	8.8	293.4	17.5	25.1	33.6
CV (%)		19.3	12.7	14.8	10.4	23.5	20.2	3.6	52.1

DF = degree of freedom, $LSD_{0.05}$ = least significant difference at 5% P-value threshold, CV = coefficient of variation, ns = not significant, *** = significant at P = 0.001. ALS = angular leaf spot, BCMV = bean common mosaic virus, CBB = common bacterial blight, ANTH = anthracnose

***Pythium* root rot**

***Fusarium* root rot**

***Rhizoctonia* root rot**

Figure 1. Root rot symptoms on susceptible inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya.

TABLE 5. Incidence and severity of *Fusarium*, *Rhizoctonia* and *Pythium* root rots on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotype	*Fusarium*			*Rhizoctonia*			*Pythium*		
	Incidence (%)	Severity	RC	Incidence (%)	Severity	RC	Incidence (%)	Severity	RC
KMA13-17-17	69.3	4.2	I	53.9	2.0	R	69.3	3.6	I
KMA13-17-25	92.3	6.9	S	50.0	1.5	R	84.6	4.4	I
KMA13-21-10	73.1	3.9	I	53.9	3.0	R	65.4	3.6	I
KMA13-21-11	65.4	6.1	S	50.0	1.5	R	61.6	2.9	R
KMA13-21-20	73.1	4.9	I	53.9	4.2	I	65.4	3.4	I
KMA13-22-21	73.1	4.7	I	53.9	4.0	I	53.9	2.3	R
KMA13-22-30	76.9	5.4	I	53.9	3.2	I	73.1	4.4	I
KMA13-23-13	69.3	4.1	I	53.9	2.3	R	69.3	3.6	I
KMA13-23-14	73.1	5.2	I	53.9	3.0	R	61.6	2.9	R
KMA13-23-22	84.6	6.2	I	57.7	2.1	R	61.6	3.3	I
KMA13-24-7	61.6	3.8	I	50.0	1.5	R	57.7	3.1	I
KMA13-25-9	65.4	4.6	I	53.9	2.5	R	53.9	2.5	R
KMA13-26-32	69.3	5.0	I	53.9	1.8	R	69.3	3.9	I
KMA13-27-12	84.6	5.9	I	57.7	3.1	I	69.3	4.5	I
KMA13-27-27	80.8	5.5	I	50.0	1.5	R	59.1	3.9	I
KMA13-27-31	49.3	2.8	R	50.0	1.5	R	57.7	3.9	I
KMA13-28-13	80.8	6.5	S	53.9	2.0	R	65.4	3.3	I
KMA13-28-2	61.6	4.1	I	50.0	1.5	R	57.7	3.9	I
KMA13-28-21	65.4	3.4	I	53.9	5.0	I	53.9	2.8	R
KMA13-28-5	80.8	6.0	I	53.9	3.0	R	59.1	2.6	R
KMA13-29-21	69.3	5.0	I	53.9	2.0	R	73.1	4.4	I
KMA13-29-24	76.9	4.9	I	50.0	1.5	R	61.6	3.4	I
KMA13-30-14	88.5	5.9	I	53.9	3.0	R	53.9	2.5	R
KMA13-30-22	80.8	5.6	I	53.9	1.8	R	84.6	5.6	I
KMA13-31-62	73.1	4.9	I	61.6	3.7	I	65.4	4.1	I
KMA13-32-28	61.6	4.9	I	53.9	2.0	R	57.7	2.1	R
AND1062	69.3	4.4	I	50.0	1.5	R	73.1	3.6	I
BRB191	96.2	6.5	S	53.9	1.8	R	80.8	4.9	I
G10909	80.8	6.1	S	57.7	2.1	R	69.3	3.4	I
G2333	57.7	3.9	I	50.0	1.5	R	53.9	2.3	R
GLP585	61.6	2.9	R	50.0	1.5	R	69.3	3.6	I
GLP92	92.3	5.9	I	50.0	1.5	R	69.3	3.9	I
KATB1	69.3	3.9	I	53.9	2.3	R	80.8	5.8	I
KATB9	80.8	5.9	I	50.0	1.5	R	57.7	3.0	R
Mex54	88.5	6.1	S	50.0	1.5	R	61.6	2.4	R
RWR719	76.9	4.2	I	50.0	1.5	R	57.7	2.6	R
$LSD_{0.05}$	29.1	2.7		8.1	2.0		25.0	1.9	
CV (%)	19.3	26.4		7.5	45.1		18.9	27.9	

RC = reaction category; R = resistant; I = intermediate; S = susceptible; LSD = least significant difference at P-value threshold of 0.05; CV = coefficient of variation

the elite lines combined moderate resistance (scores of 4 to 6) for reaction to the three root rots.

Bean common mosaic virus (BCMV). Field illustration of the BCMV disease progression is presented in Figure 2 for the 14, 21 and 28 days after inoculation. A total of 13 elite lines were resistant to BCMV; while the other 13 were moderately resistant (Table 6). However, none of the elite lines was completely immune or highly susceptible to BCMV. Four of the 10 checks were resistant, five were intermediate and one (KATB1) was highly susceptible. The BCMV incidence was very high and increased over time from 34.8 percent 14 days after inoculation, to 88.2 percent after 21 days and to 93.4 percent on the 28th day after inoculation.

The disease severity score increased from 2.5 on the 14th day after inoculation, to 3.0 and 3.5 on the 21st and 28th days after inoculation, respectively. There were highly significant differences ($P<0.001$) among genotypes for their reaction to BCMV for severity AUDPCs. The highest levels of infection were recorded on the check variety KATB1 (82.2). Line KMA13-30-14 (24.5) was the most resistant genotype among all the elite lines and checks. Other elite lines with low

Figure 2. Bean common mosaic virus disease progression on susceptible cultivar (KATB1) used as check in a greenhouse at Kabete, University of Nairobi, Kenya. Days = days after plant inoculation.

TABLE 6. Incidence and severity of bean common mosaic virus on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotype	14 days after inoculation		21 days after inoculation		28 days after inoculation		Severity AUDPC	RC
	Incidence (%)	Severity	Incidence (%)	Severity	Incidence (%)	Severity		
KMA13-17-17	16.7	1.5	100.0	3.0	100.0	4.5	42.0	I
KMA13-17-25	50.0	2.0	100.0	2.0	100.0	4.0	35.0	I
KMA13-21-10	16.7	2.0	66.6	4.0	80.0	5.5	54.2	I
KMA13-21-11	35.0	4.0	94.5	4.0	100.0	4.5	57.8	I
KMA13-21-20	25.0	2.0	100.0	3.0	100.0	4.0	42.0	I
KMA13-22-21	8.4	1.5	33.3	2.0	58.4	2.0	26.2	R
KMA13-22-30	35.0	3.0	33.3	4.0	80.0	3.5	50.8	I
KMA13-23-13	41.7	3.5	91.7	3.0	98.4	2.5	42.0	R
KMA13-23-14	28.6	2.0	100.0	3.0	100.0	3.5	40.2	I
KMA13-23-22	50.0	1.5	100.0	2.0	100.0	2.0	26.2	R
KMA13-24-7	21.7	2.5	100.0	3.0	100.0	4.0	43.8	I
KMA13-25-9	43.8	2.0	100.0	2.0	100.0	2.5	29.8	R
KMA13-26-32	54.6	3.0	100.0	3.0	100.0	4.0	45.5	I
KMA13-27-12	8.4	1.5	100.0	2.0	100.0	2.0	26.2	R
KMA13-27-27	16.7	1.5	40.0	2.0	55.0	2.5	28.0	R
KMA13-27-31	50.0	2.5	100.0	2.0	100.0	4.5	38.5	I
KMA13-28-13	31.3	3.0	37.5	3.0	68.8	2.5	40.2	R
KMA13-28-2	28.6	4.5	100.0	5.5	100.0	5.5	73.5	I
KMA13-28-21	10.0	1.5	100.0	2.0	100.0	2.0	26.2	R
KMA13-28-5	54.8	3.5	28.6	2.0	56.0	3.0	36.8	R
KMA13-29-21	50.0	1.5	100.0	3.0	100.0	2.5	35.0	R
KMA13-29-24	16.7	1.5	100.0	2.0	100.0	3.0	29.8	R
KMA13-30-14	12.5	1.5	87.5	2.0	90.0	1.5	24.5	R
KMA13-30-22	12.5	1.5	75.0	3.0	82.5	3.5	38.5	I
KMA13-31-62	20.6	2.5	87.5	4.0	94.3	3.0	47.2	R
KMA13-32-28	57.5	3.5	100.0	5.0	100.0	4.5	63.0	I
AND1062	50.0	4.0	100.0	4.0	100.0	5.5	61.2	I
BRB191	47.9	4.0	100.0	4.0	100.0	3.5	40.2	I
G10909	87.5	4.0	100.0	3.0	100.0	2.5	43.8	R
G2333	14.3	1.5	100.0	2.0	100.0	3.0	29.8	R
GLP585	8.4	1.5	100.0	2.0	100.0	2.0	26.2	R
GLP92	18.4	2.0	100.0	3.0	100.0	2.5	36.8	R
KATB1	90.0	4.5	100.0	6.0	100.0	7.0	82.2	S
KATB9	25.0	2.0	100.0	3.0	100.0	4.5	43.8	I
Mex54	50.0	2.5	100.0	4.0	100.0	3.5	49.0	I
RWR719	66.7	3.0	100.0	3.0	100.0	4.0	45.5	I
$LSD_{0.05}$	22.1	1.8	13.8	1.2	29.7	1.9	8.8	
CV (%)	37.9	34.9	17.7	13.9	19.2	27.8	10.4	

RC = reaction category; R = resistant; I = intermediate; S = susceptible; LSD = least significant difference at P-value threshold of 0.05; CV = coefficient of variation

levels of infection were KMA13-22-21, KMA13-23-22, KMA13-27-12, and KMA13-28-21 with an AUDPC value of 26.2.

Angular leaf spot (ALS). The ALS disease progression on a susceptible genotype at the 14, 21 and 28 days after inoculation is illustrated in Figure 3. Table 7 shows that 18 of the 26 elite lines were resistant to infection by ALS (*Pseudocercospora griseola*); eight were intermediate, and none was highly susceptible. The pathogen effects were almost static (stable) over time as the severity scores were 2.0, 2.5 and 2.8 at 14, 21 and 28 days after inoculation, respectively. However, disease incidence increased from 35.1% on the 14th day after inoculation to 45.7% on the 21st day, and to 51.5% on the 28th day.

Computed AUDPCs showed that there were highly significant (P<0.001) differences among the genotypes for reaction to the ALS infections. The elite line KMA13-17-25, with an AUDPC value of 14.0, was the most resistant genotype to ALS compared to all other lines and parental checks. Other elite lines with low levels of infection were KMA13-27-12 (24.5), KMA13-17-17, KMA13-23-14, KMA13-26-32, and KMA13-28-21, all with an AUDPC value of 26.2.

Common bacterial blight (CBB). Table 8 shows that six of 26 elite lines were resistant to CBB, among which KMA13-17-17, KMA13-28-2, KMA13-28-21 and KMA13-30-14 were completely immune, since not a single plant showed CBB symptoms. Eighteen of the 26 elite lines had moderate resistance (3.1 to 6.0); while two were highly susceptible (6.1 to 9). None of the check varieties was resistant to CBB; yet eight were moderately resistant; while two were highly susceptible (Mex54 and G2333).

The CBB severity and incidence on tested lines increased over time (Fig. 4). There were highly significant (P<0.001) differences among genotypes for reactions to CBB, compared to their severity AUDPCs. Based on computed AUDPC values, check variety Mex54 was the most susceptible. The lowest infection level was recorded on elite lines KMA13-17-17, KMA13-28-2 and KMA13-30-14.

Anthracnose pathogen. Figure 5 illustrates the disease progression on the susceptible check KATB1 using photos. The elite lines were resistant to anthracnose (Table 9). The disease severity ranged from 1.0 on elite lines KMA13-21-20, KMA13-28-21, and KMA13-29-21 to 6.0 on the check variety KATB1. Disease incidences were also low; averages were 20.9, 24.1, and 28.9% at 14, 21 and 28 days after inoculation. As for AUDPC values, KMA13-21-20, KMA13-28-21, and KMA13-29-21 were the most resistant; having recorded the lowest infection levels. The

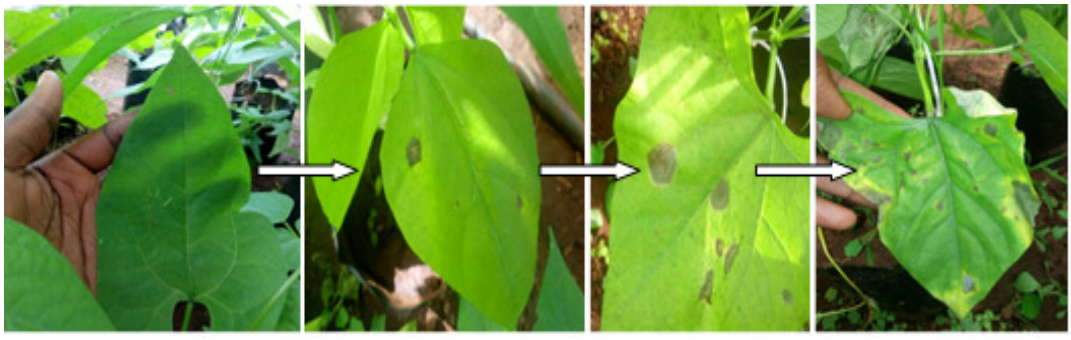

Figure 3. ALS disease progression on susceptible cultivar (RWR719) used as check in a greenhouse at Kabete, University of Nairobi, Kenya.

TABLE 7. Incidence and severity of angular leaf spot on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotype	14 days after inoculation		21 days after inoculation		28 days afterinoculation		Severity AUDPC	RC
	Incidence (%)	Severity	Incidence (%)	Severity	Incidence (%)	Severity		
KMA13-17-17	33.4	2.0	36.7	2.0	36.7	1.5	26.2	R
KMA13-17-25	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-21-10	56.3	2.0	62.9	2.5	69.3	4.0	36.7	I
KMA13-21-11	38.1	2.0	38.6	4.0	50.0	3.5	40.2	I
KMA13-21-20	42.9	2.0	50.0	3.0	56.8	3.0	35.0	R
KMA13-22-21	59.1	3.0	64.4	3.5	68.9	4.5	49.0	I
KMA13-22-30	22.8	2.0	28.1	2.0	59.1	4.5	36.8	I
KMA13-23-13	87.5	2.0	95.0	3.5	97.5	4.5	42.0	I
KMA13-23-14	10.0	2.0	20.0	1.5	20.0	2.0	26.2	R
KMA13-23-22	64.3	2.0	72.9	4.0	72.9	4.0	42.0	I
KMA13-24-7	83.4	2.0	83.4	3.0	90.8	3.0	35.0	R
KMA13-25-9	53.6	2.0	62.2	3.5	68.6	3.0	36.8	R
KMA13-26-32	25.0	2.0	33.3	2.0	36.7	1.5	26.2	R
KMA13-27-12	0.0	2.0	33.3	1.0	38.4	2.0	24.5	R
KMA13-27-27	32.5	2.0	37.5	2.0	40.0	2.5	29.8	R
KMA13-27-31	37.5	2.0	37.5	2.5	38.8	2.5	31.5	R
KMA13-28-13	33.3	2.0	44.4	2.0	52.2	3.0	31.5	R
KMA13-28-2	42.9	2.0	42.8	2.0	55.7	2.5	29.8	R
KMA13-28-21	8.4	2.0	42.9	1.5	44.3	2.0	26.2	R
KMA13-28-5	30.3	2.0	33.3	2.0	37.5	2.5	29.8	R
KMA13-29-21	12.5	2.0	25.0	1.5	32.5	2.5	28.0	R
KMA13-29-24	18.8	2.0	25.0	2.0	28.6	3.5	33.2	I
KMA13-30-14	28.4	2.0	33.3	3.5	50.0	3.5	38.5	I
KMA13-30-22	20.0	2.0	40.0	2.0	46.7	2.0	28.0	R
KMA13-31-62	44.3	2.0	46.4	3.0	51.6	2.0	31.5	R
KMA13-32-28	12.5	2.0	25.0	1.5	33.6	2.5	28.0	R
AND1062	37.5	2.0	40.0	2.0	45.0	3.5	29.8	I
BRB191	62.5	2.0	70.0	2.5	77.5	3.0	29.8	R
G10909	63.1	2.0	67.1	2.0	68.6	2.0	38.5	R
G2333	25.0	2.0	50.0	2.0	56.7	3.5	29.8	I
GLP585	43.8	2.0	47.5	3.5	48.8	3.5	35.0	I
GLP92	0.0	3.0	50.0	1.0	50.0	4.0	28.0	I
KATB1	41.7	2.0	66.6	3.0	70.0	3.5	33.2	I
KATB9	26.8	2.0	35.0	2.5	42.5	3.5	28.0	I
Mex54	50.0	2.0	60.0	2.0	65.0	2.2	35.0	R
RWR719	15.6	2.0	44.4	3.0	51.7	4.6	35.0	I
$LSD_{0.05}$	29.0	0.7	28.1	1.2	45.1	1.8	8.3	
CV (%)	40.7	14.4	23.0	24.6	37.5	32.9	12.7	

RC = reaction category; R = resistant; I = intermediate; S = susceptible; LSD = least significant difference at P-value threshold of 0.05; CV = coefficient of variation

TABLE 8. Incidence and severity of common bacterial blight on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotype	14 days after inoculation		21 days after inoculation		28 days afterinoculation		Severity AUDPC	RC
	Incidence (%)	Severity	Incidence (%)	Severity	Incidence (%)	Severity		
KMA13-17-17	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-17-25	12.5	1.5	50.0	3.0	50.0	4.0	36.8	I
KMA13-21-10	45.2	3.0	69.1	4.0	74.6	6.0	56.0	I
KMA13-21-11	18.8	2.0	50.0	3.0	56.3	4.0	38.5	I
KMA13-21-20	56.3	4.0	56.3	4.0	62.5	7.0	59.5	S
KMA13-22-21	31.3	2.5	60.7	3.5	60.7	5.0	47.2	I
KMA13-22-30	75.0	4.5	75.0	3.5	75.0	5.0	54.2	I
KMA13-23-13	66.7	4.5	100.0	4.5	100.0	8.0	68.2	S
KMA13-23-14	58.3	4.5	83.3	4.5	83.3	6.0	61.2	I
KMA13-23-22	39.3	3.0	53.5	3.5	65.3	6.0	50.8	I
KMA13-24-7	58.4	3.5	66.7	3.5	66.7	5.0	50.8	I
KMA13-25-9	33.3	2.5	33.3	3.0	44.5	5.0	42.0	I
KMA13-26-32	31.3	2.0	38.8	2.0	45.0	2.0	28.0	R
KMA13-27-12	37.5	2.5	55.0	3.5	62.5	5.0	45.5	I
KMA13-27-27	37.5	3.5	50.0	3.5	56.3	6.0	52.5	I
KMA13-27-31	37.5	2.0	37.5	1.5	37.5	2.0	22.8	R
KMA13-28-13	43.8	3.0	43.8	2.5	43.8	5.0	42.0	I
KMA13-28-2	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-28-21	0.0	1.0	8.4	1.5	0.0	1.0	17.5	R
KMA13-28-5	44.4	3.5	66.7	4.5	74.5	6.0	61.2	I
KMA13-29-21	16.7	1.5	33.3	2.0	83.3	5.0	31.5	I
KMA13-29-24	75.0	3.5	87.5	3.0	87.5	5.0	49.0	I
KMA13-30-14	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-30-22	31.3	2.5	56.3	3.0	68.8	5.0	45.5	I
KMA13-31-62	42.9	3.5	64.3	2.5	71.4	5.0	43.8	I
KMA13-32-28	25.0	3.5	35.0	3.0	36.7	6.0	47.2	I
AND1062	20.0	2.0	30.0	2.5	50.0	4.0	35.0	I
BRB191	30.0	2.5	30.0	3.5	40.0	6.0	47.2	I
G10909	10.0	1.5	46.7	2.5	56.7	6.0	36.8	I
G2333	13.4	2.0	25.9	3.5	52.7	8.0	50.8	S
GLP585	5.6	1.5	18.1	2.5	22.2	5.0	35.0	I
GLP92	46.7	3.5	75.0	3.0	76.7	5.0	47.2	I
KATB1	31.3	3.5	56.3	3.0	56.3	4.0	43.5	I
KATB9	40.0	3.0	60.0	3.0	70.0	5.0	47.2	I
Mex54	52.7	5.0	72.3	5.0	79.5	8.0	71.8	S
RWR719	41.0	2.0	42.5	2.5	47.8	4.0	35.0	I
$LSD_{0.05}$	37.4	1.3	31.1	1.7	26.7	2.0	17.5	
CV (%)	54.9	23.1	33.6	28.8	25.8	27.5	20.2	

RC = reaction category; R = resistant; I = intermediate; S = susceptible; LSD = least significant difference at P-value threshold of 0.05; CV = coefficient of variation

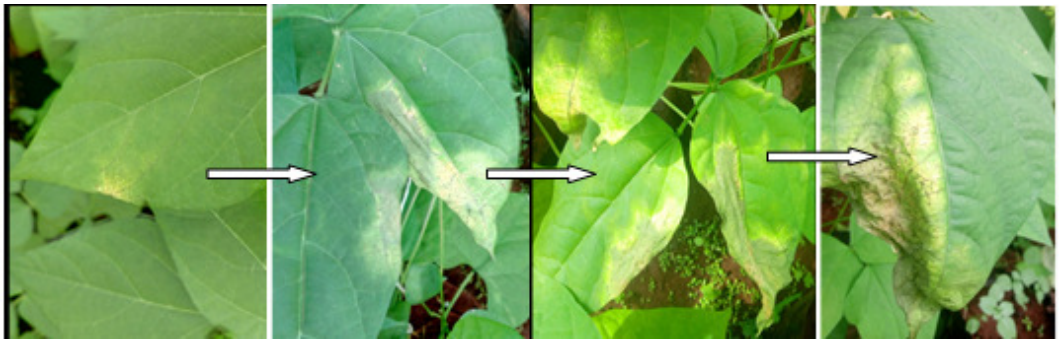

Figure 4. CBB disease progression on susceptible cultivar (Mex54) used as check in a greenhouse at Kabete, University of Nairobi, Kenya.

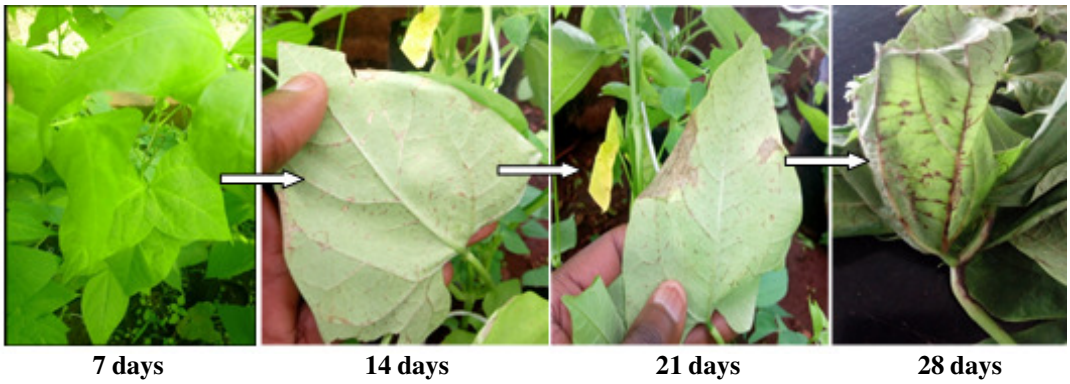

Figure 5. Anthracnose disease progression on susceptible cultivar (KATB1) used as check in a greenhouse at Kabete, University of Nairobi, Kenya.

highest levels of infection were recorded on check varieties KATB1 and KATB9.

Multiple disease resistance in elite lines. All the elite lines possessed resistance to at least one of the pathogens (Table 10). In summary, five of the 26 elite lines possessed a multiple resistance to five pathogens (KMA13-25-9, KMA13-27-31, KMA13-28-21, KMA13-28-5, and KMA13-30-14); eight genotypes were resistant to four pathogens (KMA13-17-17, KMA13-23-14, KMA13-26-32, KMA13-27-27, KMA13-28-13, KMA13-28-2, KMA13-29-21, and KMA13-32-28); nine genotypes were resistant to three pathogens, three of the 26 elite lines possessed resistance to two pathogens and one had resistance to one disease. No significant correlations in reaction of tested genotypes to the seven diseases used in this study (Table 11), except the significant correlation between the BCMV and the ALS (r=0.39*).

DISCUSSION

Inter-racial crosses and marker-assisted gamete selection method proved to be effective in pyramiding genes for disease resistance to major common bean diseases in Eastern and Central Africa. Up to 96% of the tested elite lines (25 of the 26) had combined resistance to at least two pathogens; while five lines had

TABLE 9. Incidence and severity of anthracnose on inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotype	14 Days after inoculation		21 Days after inoculation		28 Days afterinoculation		Severity AUDPC	RC
	Incidence (%)	Severity	Incidence (%)	Severity	Incidence (%)	Severity		
KMA13-17-17	20.0	2.0	22.0	2.0	27.0	2.0	28.0	R
KMA13-17-25	9.0	2.0	11.0	2.0	19.0	2.0	28.0	R
KMA13-21-10	16.7	2.0	19.0	2.0	22.0	3.0	31.5	R
KMA13-21-11	16.7	2.0	17.5	2.0	22.0	2.0	28.0	R
KMA13-21-20	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-22-21	57.1	2.0	64.0	3.0	69.0	3.0	38.5	R
KMA13-22-30	40.0	2.0	43.0	2.0	44.0	2.0	28.0	R
KMA13-23-13	33.3	2.0	39.0	2.0	39.0	3.0	31.5	R
KMA13-23-14	36.4	2.0	38.0	2.0	40.0	3.0	31.5	R
KMA13-23-22	71.4	3.0	77.0	4.0	77.0	4.0	52.5	I
KMA13-24-7	75.0	3.0	87.0	3.0	100.0	3.0	42.0	R
KMA13-25-9	0.0	1.0	0.0	1.0	11.0	2.0	17.5	R
KMA13-26-32	40.0	2.0	48.0	2.0	52.0	2.0	28.0	R
KMA13-27-12	18.2	2.0	20.0	2.0	26.0	2.0	28.0	R
KMA13-27-27	22.2	2.0	25.0	2.0	27.0	2.0	28.0	R
KMA13-27-31	0.0	1.0	0.0	1.0	11.0	2.0	17.5	R
KMA13-28-13	37.5	2.0	38.0	3.0	40.0	3.0	38.5	R
KMA13-28-2	14.3	2.0	15.0	2.0	19.0	2.0	28.0	R
KMA13-28-21	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-28-5	10.0	2.0	15.0	2.0	22.0	2.0	28.0	R
KMA13-29-21	0.0	1.0	0.0	1.0	0.0	1.0	14.0	R
KMA13-29-24	16.7	2.0	18.0	2.0	24.0	2.0	28.0	R
KMA13-30-14	20.0	2.0	22.0	2.0	32.0	3.0	31.5	R
KMA13-30-22	50.0	2.0	55.0	2.0	62.0	2.0	28.0	R
KMA13-31-62	11.1	2.0	15.0	2.0	17.0	2.0	28.0	R
KMA13-32-28	0.0	1.0	9.0	2.0	14.0	2.0	24.5	R
AND1062	0.0	1.0	11.0	3.0	24.0	5.0	42.0	I
BRB191	6.0	2.0	17.0	4.5	22.0	5.0	56.0	I
G10909	25.0	2.0	30.0	2.0	37.0	2.0	28.0	R
G2333	25.0	2.0	25.0	2.5	29.0	2.5	33.2	R
GLP585	27.3	2.0	32.0	3.0	38.0	3.5	40.2	I
GLP92	0.0	1.0	11.0	2.0	19.0	2.0	24.5	R
KATB1	16.7	3.0	18.0	5.5	25.5	6.0	70.0	I
KATB9	5.5	2.5	14.5	5.5	25.5	5.5	66.5	I
Mex54	0.0	1.0	0.0	1.0	12.0	2.0	17.5	R
RWR719	11.1	2.0	14.0	2.0	16.0	3.0	31.5	R
$LSD_{0.05}$	2.6	1.0	1.7	2.8	2.9	3.1	33.6	
CV (%)	6.2	25.6	3.4	60.7	5.0	59.1	52.1	

RC = reaction category; R = resistant; I = intermediate; S = susceptible; LSD = least significant difference at P-value threshold of 0.05; CV = coefficient of variation

TABLE 10. Multiple disease resistance of elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Genotypes	[1]Pathogens							[2]Resistances	Number
	ALS	BCMV	CBB	ANT	*Fusarium*	*Rhizoctonia*	*Pythium*		
KMA13-17-17	R	I	R	R	I	R	I	A, C, AN, R	4
KMA13-17-25	R	I	I	R	S	R	I	A, AN, R	3
KMA13-21-10	I	I	I	R	I	R	I	AN, R	2
KMA13-21-11	I	I	I	R	S	R	R	AN, R, P	3
KMA13-21-20	R	I	S	R	I	I	I	A, AN	2
KMA13-22-21	I	R	I	R	I	I	R	B, AN, P	3
KMA13-22-30	I	I	I	R	I	I	I	ANT	1
KMA13-23-13	I	R	S	R	I	R	I	B, AN, R	3
KMA13-23-14	R	I	I	R	I	R	R	A, R,AN, P	4
KMA13-23-22	I	R	I	I	I	R	I	B, R	2
KMA13-24-7	R	I	I	R	I	R	I	A, AN, R	3
KMA13-25-9	R	R	I	R	I	R	R	A, B,AN, R, P	5
KMA13-26-32	R	I	R	R	I	R	I	A, C, AN, R	4
KMA13-27-12	R	R	I	R	I	I	I	A, AN, B	3
KMA13-27-27	R	R	I	R	I	R	I	A, B, AN, R	4
KMA13-27-31	R	I	R	R	R	R	I	A, C, AN, F, R	5
KMA13-28-13	R	R	I	R	S	R	I	A, B, AN, R	4
KMA13-28-2	R	I	R	R	I	R	I	A, C, AN, R	4
KMA13-28-21	R	R	R	R	I	I	R	A, B, C, AN, P	5
KMA13-28-5	R	R	I	R	I	R	R	A, B, AN, R, P	5
KMA13-29-21	R	R	I	R	I	R	I	A, B, AN, R	4
KMA13-29-24	I	R	I	R	I	R	I	B, AN, R	3
KMA13-30-14	I	R	R	R	I	R	R	B, C, AN, R, P	5
KMA13-30-22	R	I	I	R	I	R	I	A, AN, R	3
KMA13-31-62	R	R	I	R	I	I	I	A, B, AN	3
KMA13-32-28	R	I	I	R	I	R	R	A, AN, R, P	4

1: R = resistant; I = intermediate; S = susceptible. 2: A = ALS; B = BCMV; C = CBB; AN = anthracnose; F = *Fusarium*; R = *Rhizoctonia* and P = *Pythium*

TABLE 11. Pearson's correlation coefficients among pathogens for disease resistance of inter-racial elite common bean lines grown in a greenhouse at Kabete, University of Nairobi, Kenya

Pathogens	ALS	ANT	BCMV	CBB	FRR	PRR
ANT	0.16^{ns}					
BCMV	0.39*	-0.11^{ns}				
CBB	0.12^{ns}	0.03^{ns}	0.18^{ns}			
FRR	-0.18^{ns}	-0.02^{ns}	-0.07^{ns}	0.18^{ns}		
PRR	-0.19^{ns}	0.04^{ns}	0.07^{ns}	-0.10^{ns}	0.08^{ns}	
RRR	-0.01^{ns}	-0.07^{ns}	-0.22^{ns}	0.03^{ns}	0.07^{ns}	-0.20^{ns}

ns = not significant; * = significant at 0.05 P-value threshold; ALS = angular leaf spot; ANT = anthracnose; BCMV = bean common mosaic virus; CBB = common bacterial blight; FRR = *Fusarium* root rot; PRR = *Pythium* root rot; RRR = *Rhizoctonia* root rot

multiple resistance to five pathogens (Table 10). This implies that markers are effective in identifying and transferring of resistance genes to susceptible commercial varieties in early generations. Pyramiding genes for disease resistance in a genotype is a more durable and sustainable strategy to control diseases, as multiple coinfections of pathogens are common in production fields and have been reported to substantially affect productivity of the common bean (Singh, 2001; Valentini *et al.,* 2017; Okii *et al.,* 2018).

While developing inter-gene pool multiple-parent genotypes, Okii *et al.* (2017) showed the effectiveness of marker-assisted selection to pyramid genes of resistance as well as improve the agronomic qualities. In their study, disease resistance was associated with small-seeded Mesoamerican genotypes, except for the BCMV where the Andean and Mesoamerican genotypes behaved similarly. This could explain the growing interest in inter-racial crosses among genotypes belonging to these two gene pools. Thus, the low levels of disease infection recorded on test elite lines in the greenhouse could be attributed to effects of inter-gene and inter-racial crosses performed between Andean and Mesoamerican cultivars as they allowed to broaden the genetic base and increase levels of resistance to both biotic and abiotic stresses (Welsh *et al.,* 1995; Singh *et al.,* 2002; Singh and Schwartz, 2010; Singh, 2013).

Gamete selection method was effective as it allowed pyramiding resistance genes to target pathogens, and thus reached the primary objective of this breeding programme, which was to ascertain the effectiveness of the gamete selection in pyramiding resistance genes to major bean diseases in Eastern Africa in susceptible popular cultivars.

Many other successful applications of the gamete selection to improve the common bean disease resistance have been reported (Singh *et al.,* 1998; Asensio-S.-Manzanera *et al.*, 2005; 2006; Singh *et al.*, 2008; Terán and Singh, 2009). The innovation of using markers on the gamete selection during this breeding programme allowed to increase precision and efficiency, and therefore, made it easy to pyramid desirable genes as previously stated by Miklas *et al.* (2006).

There was an independent inheritance of genes controlling the major common bean diseases, as no significant correlations were reported among them (no co-segregation of resistance genes), except the significant correlation between the BCMV and the ALS (Table 11).

More surprising were reactions of elite lines to root rot-causing agents (Table 5). *Fusarium* root rot was the most damaging among

common bean root rots, both for disease incidence and severity. Only one elite line from the 26 tested and one check variety of the 10 used were resistant to *Fusarium* root rot. A study carried out by Mukankusi (2008) confirmed the virulence the *Fusarium* root rot as, among the 147 accessions evaluated in that study, none of them showed resistance to this pathogen. Spence (2003) found that *F. solani* was more damaging than the two common species of *Pythium* (*P. torulosum* and *P. spinosum*) in Uganda.

Although the plant materials used in the present study were improved for *Pythium* root rot resistance, its incidence and severity were still very high. Only eight out of 26 elite lines possessed the *Pythium* root rot resistance. None of the genotypes had shown concurrent resistance to *Pythium* and *Fusarium* root rot, even though the parental line RWR719, which was used in study populations, has been reported to possess genes of resistance to both pathogens (Otsyula *et al.,* 2003; Mukankusi, 2015). Similar results were reported by Mukankusi *et al.* (2018) as only 21.5% of tested inter-specific lines combined concurrent resistance to *Fusarium* and *Pythium* root rot. These results supported those of Ongom *et al.* (2012), who concluded that although quantitative trait loci (QTLs) linked to *Fusarium solani* resistance have been mapped on the same chromosome as that for resistance to *Pythium ultimum*, their resistances were inherited independently and the correlations between them were very low (Table 11). In addition, resistance to *Fusarium solani* is believed to be much more complex as it is controlled by two or more genes (Mukankusi *et al.,* 2011; Obala *et al.,* 2012); while the *Pythium ultimum* resistance is only conditioned by a single dominant gene, marked by a dominant SCAR marker-PYAA19^{800} (Otsyula *et al.,* 2003; Mahuku *et al.,* 2005; Otsyula, 2010). This could explain why breeding for *Pythium* root rot resistance did not improve significantly the *Fusarium* root rot resistance, even if a donor parent (RWR719) resistant to both pathogens was involved in the crosses.

CONCLUSION

This study has confirmed the effectiveness of inter-racial crosses and marker-assisted gamete selection to concurrently improve resistance of common bean to major diseases in Eastern and Central Africa. From the 26 elite lines tested in this experiment, five lines possess a concurrent resistance to five pathogens; eight are resistant to four pathogens; nine are resistant to three pathogens, three show resistance to two pathogens and one has a resistance to one pathogen.

Efficient use of markers in the gamete selection method at early generations is effective for pyramiding resistance genes into susceptible genotypes. However, there are no significant correlations in the reaction of tested genotypes to pathogens used in this study, except the significant correlation between the reaction of genotypes to BCMV and the ALS. This suggests that most of the genes controlling resistance to these major bean diseases are inherited independently.

Further field experiments in areas with a high prevalence of these diseases should be conducted to confirm the multiple disease resistance of these elite lines before releasing to farmers. In addition, more sources of resistance to these pathogens should be sought and introgressed for durable resistance, especially to CBB and *Fusarium* root rot.

ACKNOWLEDGMENT

Financial support from the Université Evangélique en Afrique is gratefully acknowledged.

REFERENCES

Alladassi, B.M.E., Nkalubo, S.T., Mukankusi, C., Kayaga, H.N., Gibson, P., Edema, R.,

Urrea, C.A., Kelly, J.D. and Rubaihayo, P.R. 2018. Identification of common bean genotypes with dual leaf and pod resistance to common bacterial blight disease in Uganda. *African Crop Science Journal* 26(1):63-77.

Asensio, S.M.M.C., Asensio, C. and Singh, S.P. 2006. Gamete selection for resistance to common and halo bacterial blights in dry bean inter-gene pool populations. *Crop Science* 46(1):131-135.

Asensio-S.-Manzanera, M.C., Asensio, C. and Singh, S.P. 2005. Introgressing resistance to bacterial and viral diseases from the Middle American to Andean common bean. *Euphytica* 143(1-2):223-228.

Asfaw, A., Blair, M.W. and Almekinders, C. 2009. Genetic diversity and population structure of common bean (*Phaseolus vulgaris* L.) landraces from the East African highlands. *Theoretical and Applied Genetics* 120(1):1–12.

Beaver, J.S. and Osorno, J.M. 2009. Achievements and limitations of contemporary common bean breeding using conventional and molecular approaches. *Euphytica* 168:145–175.

Beebe, S., Rao, I.M., Blair, M.W. and Acosta-Gallegos, J.A. 2013. Phenotyping common beans for adaptation to drought. *Frontiers in physiology* 4:35.

Bigirimana, J. and Höfte, M. 2001. Bean anthracnose: inoculation methods and influence of plant stage on resistance of *Phaseolus vulgaris* cultivars. *Journal of Phytopathology* 149(7 8):403-408.

Broughton, W.J., Hernandez, G., Blair, M., Beebe, S., Gepts, P. and Vanderleyden, J. 2003. Beans (*Phaseolus spp.*)–model food legumes. *Plant and soil* 252(1):55-128.

Buruchara, R., Estevez de Jensen, C., Godoy, G., Abawi, G., Pasche, J., Lobo Junior, M. and Mukankusi, C. 2015. A review of the root rot diseases of common bean with emphasis in Latin America and Africa, Conference paper, July 20-23, 2015, Protea Hotel, Kruger Gate Skukuza, South Africa.

Campbell, C.L. and Madden, L.V. 1990. Introduction to plant disease epidemiology. John Wiley & Sons, New York. 532pp.

CGIAR. 2017. Ten year research strategy for pulse crops. http://grainlegumes.cgiar.org/wp-content/uploads/2017/01/IYP_10-Year-Research-Strategy_Pulse-Crops_Final_Dec-8-20162.pdf Accessed 5th March 2018.

Correa, F.J. and Saettler, A.W. 1987. Angular leaf spot of red kidney beans in Michigan. *Plant disease* 71(10):915-918.

Ddamulira, G., Mukankusi, C., Ochwo-Ssemakula, M., Edema, R., Sseruwagi, P. and Gepts, P. 2014. Identification of new sources of resistance to angular leaf spot among Uganda common bean landraces. *Canadian Journal of Plant Breeding* 2(2):55-65.

De Queiroz, V.T., De Sousa, C.S., Costa, M.R., Sanglad, D.A., Arruda, K.M.A., De Souza, T.L.P.O., Ragagnin, V.A., De Barros, E.G. and Moreira, M.A. 2004. Development of SCAR markers linked to common bean angular leaf spot resistance genes. *Annual Report-Bean Improvement Cooperative* 47:237-238.

FAO. 2018. FAOSTAT: FAO Statistical Databases. Available online at: http://faostat.fao.org/

Garzón, L.N., Ligarreto, G.A. and Blair, M.W. 2008. Molecular marker-assisted backcrossing of anthracnose resistance into Andean climbing beans (*Phaseolus vulgaris* L.). *Crop Science* 48(2):562-570.

Harveson, R.M. and Schwartz, H.F. 2007. Bacterial diseases of dry edible beans in the Central High Plains. *Plant Health Progress* 8(1):35. doi:10.1094/PHP-2007-0125-01-DG

Jaetzold, R., Schmidt, H., Hornetz, B. and Shisanya, C. 2006. Farm Management Handbook of Kenya. Vol II, Natural conditions and farm management

information, 2nd Edition Part B Central Kenya. Subpart B2. Central Province.

Kilimo, Trust. 2012. Development of inclusive markets in agriculture and trade (DIMAT): The nature and markets of bean value chains in Uganda. United National Development Program (UNDP) Government of Uganda (GoU) Kilimo Trust, Kampala, Uganda, p.56.

Kimani, P.M., Buruchara, R., Muthamia, J., Mbikayi, N., Asfaw, A., Namayanja, A., Rachier, G., Otsyula, R. and Lunze, L. 2005a. Selection on marketable bean lines with improved resistance to angular leaf spot, root rots and yield for smallholder farmers in ECA. Presented at the Second Biotechnology, Breeding and Seed Systems Conference, January 2005, Rockefeller Foundation, Nairobi, Kenya, pp. 23-27.

Kimani, P.M., Buruchara, R., Ampofo, K., Pyndji, M., Chirwa, R. and Kirkby, R. 2005b. Breeding beans for smallholder farmers in Eastern, Central and Southern Africa: Constraints, achievements and potential. In: Pan-African Bean Research Network (PABRA) Millennium Workshop, 28 May-1 June, Arusha, Tanzania, pp. 11-28.

Kiryowa, M.J., Ebinu, A., Kyaligonza, V., Nkalubo, S.T., Paparu, P., Mukankusi, C. and Tukamuhabwa, P. 2016. Pathogenic variation of *Colletotrichum lindemuthianum* causing anthracnose of beans (*Phaseolus vulgaris*) in Uganda. *International Journal of Phytopathology* 5(3):89-98.

Leitich, R.K., Arinaitwe, W., Mukoye, B., Omayio, D.O., Osogo, A.K., Were, H.K., Muthomi, J.W., Otsyula, R.M. and Abang, M.M. 2016. Mapping of Angular Leaf Spot Disease Hotspot Areas in Western Kenya Towards Its Management. *American Journal of Applied Scientific Research* 2(6):75-81.

Mahuku, G., Buruchara, R., Navia, M. and Otsyula, R. 2005. A gene that confers resistance to *Pythium* root rot in common bean: Genetic characterization and development of molecular markers. CIAT poster Publication. Retrieved from http://webapp.ciat.cgiar.org/ipm/pdfs /poster_aps_ 2005_mahuku.pdf

Mahuku, G.S., Henríquez, M.A., Montoya, C., Jara, C., Teran, H. and Beebe, S. 2011. Inheritance and development of molecular markers linked to angular leaf spot resistance genes in the common bean accession G10909. *Molecular Breeding* 28(1):57-71.

Miklas, P.N., Kelly, J.D., Beebe, S.E. and Blair, M.W. 2006. Common bean breeding for resistance against biotic and abiotic stresses: from classical to MAS breeding. *Euphytica* 147(1-2):105-131.

Mondo, M.J., Kimani, P.M. and Narla, R.D. 2018. Marker-assisted gamete selection for multiple disease resistance and grain yield in inter-gene pool and inter-racial populations of common bean. *African Journal of Rural Development* 3 (4): 931-941.

Mondo, J.M., Kimani, P.M. and Narla, R.D. 2019a. Genotype x environment interactions on seed yield of inter-racial common bean lines in Kenya. *World Journal of Agricultural Research* 7(3):76–87.

Mondo, J.M., Kimani, P.M. and Narla, R.D. 2019b. Yield stability in small red inter-racial common bean lines in Kenya. *African Crop Science Journal* 27(3):351–374.

Mukankusi, C. 2008. Improving resistance to Fusarium root rot [*Fusariun solani* (mart) Sacc. f. sp. *phaseoli* (Burkholder) W.C Snyder & H.E Hans] in common beans (*Phaseolus vulgaris* L.). PhD Thesis, University of KwaZulu-Natal, Pietermaritzburg, South Africa.

Mukankusi, C. 2015. Breeding for root rot resistance in East and Central Africa. http://arsftfbean.uprm.edu/bean/wp-content/uploads/2015/09/BDW-Day2-Clare-Mukankusi.pdf

Mukankusi, C., Derera, J., Melis, R., Gibson, P.T. and Buruchara, R. 2011. Genetic analysis of resistance to Fusarium root rot in common bean. *Euphytica* 182(1):11.

Mukankusi, C.M., Amongi, W., Sebuliba, S., Musoke, S. and Acam, C. 2018. Characterisation of *Phaseolus coccineus* interspecific germplasm accessions for disease resistance, grain market class and yield attributes. *African Crop Science Journal* 26(1):117-135.

Musyimi, A.S. 2014. Marker-Assisted Gamete Selection for Multiple Disease Resistance in Andean Bean Genotypes and Characterization of *Colletotrichum lindemuthianum* in Kenya. M.Sc. diss., University of Nairobi, Kenya, p. 234.

Mwaipopo, B., Nchimbi-Msolla, S., Njau, P., Tairo, F., William, M., Binagwa, P., Kweka, E., Kilango, M. and Mbanzibwa, D. 2017. Viruses infecting common bean (*Phaseolus vulgaris* L.) in Tanzania: A review on molecular characterization, detection and disease management options. *African Journal of Agricultural Research* 12(18):1486-1500.

Mwang'ombe, A.W., Wagara, I.N., Kimenju, J.W. and Buruchara, R.A. 2007. Occurrence and Severity of Angular leaf spot of common bean in Kenya as influenced by geographical location, altitude and agroecological zones. *Plant Pathology Journal* 6(3):235-241.

Namayanja, A., Msolla, S.N., Buruchara, R. and Namusoke, A. 2014. Genetic Analysis of Resistance to Pythium Root Rot Disease in Common Bean (*Phaseolus vulgaris* L.) Genotypes. *Journal of Crop Improvement* 28(2):184-202.

Njuguna, S.M. 2014. Marker-assisted gamete selection for multiple disease resistance in Mesoamerican bean genotypes and race typing of angular leaf spot pathogen in Kenya. M.Sc. diss., University of Nairobi, Kenya, p. 246.

Nzungize, J., Gepts, P., Buruchara, R., Male, A., Ragama, P., Busogoro, J.P. and Baudoin, J.P., 2011a. Introgression of Pythium root rot resistance gene into Rwandan susceptible common bean cultivars. *African Journal of Plant Science* 5(3):193-200.

Nzungize, J., Gepts, P., Buruchara, R., Buah, S., Ragama, P., Busogoro, J.P. and Baudoin, J.P., 2011b. Pathogenic and molecular characterization of Pythium species inducing root rot symptoms of common bean in Rwanda. *African Journal of Microbiology Research* 5:1169-1181.

Obala, J., Mukankusi, C., Rubaihayo, P.R., Gibson, P. and Edema, R. 2012. Improvement of resistance to Fusarium root rot through gene pyramiding in common bean. *African Crop Science Journal* 20(1):1-13.

Odogwu, B.A., Nkalubo, S.T., Mukankusi, C., Odong, T., Awale, H.E., Rubaihayo, P. and Kelly, J.D. 2017. Phenotypic and genotypic screening for rust resistance in common bean germplasm in Uganda. *Euphytica* 213(2):49.

Okii, D., Mukankusi, C., Sebuliba, S., Tukamuhabwa, P., Tusiime, G., Talwana, H., Odong, T., Namayanja, A., Paparu, P., Nkalubo, S. and Otim, M. 2018. Genetic variation, Heritability estimates and GXE effects on yield traits of Mesoamerican common bean (*Phaseolus vulgaris* L) germplasm in Uganda. *Plant Genetic Resources* 16(3):237-248.

Okii, D., Tukamuhabwa, P., Tusiime, G., Talwana, H., Odong, T., Mukankusi, C., Male, A., Amongi, W., Sebuliba, S., Paparu, P. and Nkalubo, S. 2017. Agronomic qualities of genetic pyramids of common bean developed for multiple-disease-resistance. *African Crop Science Journal* 25(4):457-472.

Omunyin, M.E., Gathuru, E.M. and Mukunya, D.M. 1995. Pathogenicity groups of bean common mosaic virus isolates in Kenya. *Plant Disease* 79:985-989.

Ongom, P.O., Nkalubo, S.T., Gibson, P.T., Mukankusi, C.M. and Rubaihayo, P.R.

2012. Evaluating genetic association between Fusarium and Pythium root rots resistances in the bean genotype RWR 719. *African Crop Science Journal* 20(1):31-39.

Otsyula, R.M. 2010. Nature of genetic control and inheritance of resistance to *Pythium* root rot in bean genotypes. PhD Thesis, Makerere University, Kampala, Uganda.

Otsyula, R.M., Buruchara, R.A., Mahuku, G. and Rubaihayo, P. 2003. Inheritance and transfer of root rot (*Pythium*) resistance to bean genotypes. http://ciat-library.ciat.cgiar.org/Articulos_Ciat/ipm/pdfs/reuben_2.pdf.

Ronner, E., Descheemaeker, K., Almekinders, C.J., Ebanyat, P. and Giller, K.E. 2018. Farmers' use and adaptation of improved climbing bean production practices in the highlands of Uganda. *Agriculture, Ecosystems & Environment* 261:186-200.

Schoonhoven, A.V. and Pastor-Corrales, M.A. 1987. Standard system for the evaluation of bean germplasm. *CIAT, Cali, Colombia*, *56*.

Sharma, P.N., Pathania, A., Kapil, R., Sharma, P., Sharma, O.P., Patial, M. and Kapoor, V. 2008. Resistance to bean common mosaic potyvirus strains and its inheritance in some Indian landraces of common bean. *Euphytica* 164(1):173-180.

Sicard, D., Buchet, S., Michalakis, Y. and Neema, C. 1997. Genetic variability of *Colletotrichum lindemuthianum* in wild populations of common bean. *Plant Pathology* 46(3):355-365.

Sichilima, T., Mapemba, L. and Tembo, G., 2016. Drivers of dry common beans trade in Lusaka, Zambia: A trader's perspective. *Sustainable Agriculture Research* 5(2):15.

Singh, S.P. 1994. Gamete selection for simultaneous improvement of multiple traits in common bean. *Crop Science* 34(2):352-355.

Singh, S.P. 2013. Common bean improvement in the twenty-first century (Vol. 7). Springer Science & Business Media.

Singh, S.P. and Schwartz, H.F. 2010. Breeding common bean for resistance to diseases: a review. *Crop Science* 50(6):2199-2223.

Singh, S.P., 2001. Broadening the genetic base of common bean cultivars. *Crop Science* 41(6):1659-1675.

Singh, S.P., Cardona, C., Morales, F.J., Pastor-Corrales, M.A. and Voysest, O. 1998. Gamete selection for upright carioca bean with resistance to five diseases and a leafhopper. *Crop Science* 38(3):666-672.

Singh, S.P., Terán, H., Lema, M., Dennis, M.F., Hayes, R. and Robinson, C. 2008. Breeding for slow-darkening, high-yielding, broadly adapted dry bean pinto 'Kimberly'and 'Shoshone'. *Journal of Plant Registrations* 2(3):180-186.

Singh, S.P., Terán, H., Muñoz, C.G. and Osorno, J.M. 2002. Selection for seed yield in Andean intra-gene pool and Andean× Middle American inter-gene pool populations of common bean. *Euphytica* 127(3):437-444.

Spence, N. 2003. Characterization and epidemiology of root rot diseases caused by *Fusarium* and *Pythium sp.* in beans in Uganda. Final technical report, R7568 (ZA0373). Horticultural Research International, Wellesbourne, Warwick, CV 35, 9EF UK.

Strausbaugh, C.A., Myers, J.R., Forster, R.L. and McClean, P.E. 1999. bc-1 and bc-u—two loci controlling bean common mosaic virus resistance in common bean are linked. *Journal of the American Society for Horticultural Science* 124(6):644-648.

Terán, H. and Singh, S.P. 2009. Gamete selection for improving physiological resistance to white mold in common bean. *Euphytica* 167(3):271-280.

Valentini, G., Gonçalves-Vidigal, M.C., Cregan, P.B., Song, Q. and Pastor-Corrales, M.A.

2017. Simple sequence repeat DNA markers linked with genes for resistance to major diseases of common bean. Publications from USDA-ARS / UNL Faculty. 1659.

Verma, P. and Gupta, U.P. 2010. Immunological detection of bean common mosaic virus in French bean (*Phaseolus vulgaris* L.) leaves. *Indian Journal of Microbiology* 50(3):263-265.

VSN International. 2013. Genstat for Windows (15th Edition), Oxford, UK.

Wagara, I.N., Mwang'ombe, A.W. and Siboe, G.M. 1999. Colonisation of resistant and susceptible bean tissues by *Phaeoisariopsis griseola* (Sacc) Ferr. *African Crop Science Journal* 7(4):523-530.

Welsh, W., Bushuk, W., Roca, W. and Singh, S.P. 1995. Characterization of agronomic traits and markers of recombinant inbred lines from intra-and interracial populations of *Phaseolus vulgaris* L. *Theoretical and Applied Genetics* 91(1):169-177.

White, J.G. 1988. Studies on the biology and control of cavity spot of carrots. *Annals of Applied Biology* 113(2):259-268.

Winham, D.M., Hutchins, A.M., Thompson, S.V. and Dougherty, M.K. 2018. Arizona registered dietitians show gaps in knowledge of bean health benefits. *Nutrients* 10(1):52.

Wortmann, C.S. 1998. Atlas of common bean (*Phaseolus vulgaris* L.) production in Africa (No. 297). CIAT, Cali, Colombia.