Legume Science

WILEY

Legume Science Open Access

ORIGINAL ARTICLE OPEN ACCESS

Introgression and Stability of Common Bean Weevil (*Acanthoscelides obtectus* [Say]) Resistance in Diverse Market Classes From the Andean Gene Pool of Common Bean

Modreen Chinji[1] | Swivia Hamabwe[1] | Kuwabo Kuwabo[1] | Isabel Mugovu[1] | Rebeca Thole[1] | Maria Mazala[2] | Juan M. Osorno[2] | Phillip McClean[2] | Celestina Jochua[3] | Carlos Urrea[4] | Chikoti Mukuma[5] | Virginia Chisale[6] | Kelvin Kamfwa[1]

[1]Department of Plant Sciences, University of Zambia, Lusaka, Zambia | [2]Department of Plant Sciences, North Dakota State University, Fargo, North Dakota, USA | [3]Estacao Agraria de Chokwe, Instituto de Investigação Agraárias de Moçzambique (IIAM), Maputo, Mozambique | [4]Department of Agronomy and Horticulture, University of Nebraska, Panhandle Research, Extension and Education Center, Scottsbluff, Nebraska, USA | [5]Misamfu Research Station, Zambia Agricultural Research Institute, Misamfu, Zambia | [6]Chitedze Agricultural Research Station, Department of Agricultural Research and Technical Services (DARTS), Lilongwe, Malawi

Correspondence: Kelvin Kamfwa (kelvinkamfwa@gmail.com)

Received: 15 December 2023 | **Revised:** 22 February 2024 | **Accepted:** 23 February 2024

Funding: This work was funded in whole (or part) by the United States Agency for International Development (USAID) under Agreement #7200AA18LE00003 as part of Feed the Future Innovation Lab for Legume Systems Research.

Keywords: *Acanthoscelides obtectus* | APA locus | common bean | introgression | market class | *Phaseolus vulgaris* | resistance

ABSTRACT

The common bean weevil (*Acanthoscellides obtectus* [Say]) is a major post-harvest pest of common bean (*Phaseolus vulgaris* L.) in tropical regions. Developing and using weevil-resistant varieties is the most environmentally and cost-effective means of mitigating the losses caused by the common bean weevil. The arcelin–phytohemagglutinin–alpha-amylase (APA) locus, originally from tepary bean (*Phaseolus acutifolius* A. Gray), provides effective resistance against the common bean weevil. The APA locus is currently deployed in very limited market classes, and knowledge of the stability of its resistance across different market classes of common bean is limited. The objectives of this study were to (i) introgress the APA locus into selected market classes of Andean gene pool of common bean and (ii) determine the stability of APA-based resistance to *A. obtectus* (AO) in multiple market classes of common bean. A total of 571 $F_{5:7}$ breeding lines derived from crossing the weevil-resistant breeding line AO-1012-29-3-3A (AO-3A) possessing the APA locus with seven Andean genotypes belonging to five market classes were evaluated for resistance to AO. Of the 571 breeding lines screened, 16 were resistant, representing a low weevil resistance recovery rate of 2.8%. These lines are across diverse market classes, including those preferred in African countries. Of the 16 newly developed resistant breeding lines, six were more resistant to AO (scores ranging from 1–1.3) than AO-3A (score of 2), and these can be used for further genetic enhancement of common bean resistance to AO.

1 | Introduction

Common bean (*Phaseolus vulgaris* L.) is a major source of nutrition and food security for many households in Latin America and Africa (Petry et al. 2015; Uebersax et al. 2023). The common bean weevil (aka bruchid) (*Acanthoscellides obtectus* [Say]) and the Mexican weevil (*Zabrotes subfasciatus* [Boheman]) are important post-harvest pests of common bean responsible for

This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

© 2024 The Authors. *Legume Science* published by Wiley Periodicals LLC.

significant yield losses, and on average, these pests are estimated to cause 13% grain loss of common bean in developing countries (Kornegay and Cardona 1991). The geography of a particular location determines which of the two weevils is prevalent. *Z. subfasciatus* is prevalent at low altitudes and is favored by high temperatures while *A. obtectus* (AO) is prevalent at higher altitudes, where its life cycle is favored by cooler temperatures (Cardona et al. 1989; Howe and Currie 1964; Myers et al. 2001; Schoonhoven 1976). AO initial infestation of beans begins in the field and later progresses in storage, while *Z. subfasciatus* only attacks beans in storage (Schoonhoven 1976). In Africa, AO is more prevalent than *Z. subfasciatus*.

Depending on storage conditions, AO can cause seed yield losses of between 48% and 100% in storage (Slumpa and Ampofo 1991). Additionally, AO causes seed quality degradation and poor germination, product alteration through reduction of nutritional and aesthetic value, and cooking characteristics. It also affects taste/flavor (Kusolwa and Myers 2011; Mulungu et al. 2007). AO-damaged beans have significant price discounts, and farmers are forced to sell their beans within 2–3 months after harvesting at lower prices to avoid total grain loss or price reductions of AO-damaged seed (Mishili et al. 2011).

Weevils can be managed by use of chemicals, non-chemical treatment of grain, biological control, and improved post-harvest storage (Cardona 2004; Chireceanu et al. 2022; Mutungi et al. 2015); however, these methods are not always available and not sustainable (Blair et al. 2010), as resource-poor small-scale farmers produce most of the beans in Africa. For instance, some farmers in Eastern and Southern Africa use strategies such as solarization, sieving, admixing grain with ash, fine soil, granary hygiene, use of different storage methods and botanical products that exert some fumigant activity, contact toxicity or repellency, and oviposition-deterring properties to manage and control bean weevils (Abate and Ampofo 1996; Paul et al. 2009); however, the efficacies of these indigenous methods are low as there exists no standard application guidelines, and their use is only practical for preserving smaller quantities of seed (Stathers et al. 2008). Developing and using resistant cultivars is the most environmentally cost-effective management strategy for mitigating the losses caused by bean weevils.

Within common bean, resistance to AO and *Z. subfaciatus* was discovered first in wild beans from Mexico (Schoonhoven, Cardona, and Valor 1983). This resistance was initially attributed to the insecticidal activity of seed storage proteins known as arcelins (Osborn et al. 1988). Arcelins and two other related seed proteins, phytohemagglutinin and alpha-amylase, are encoded by the APA (arcelin, phytohemagglutinin, and alpha-amylase) locus, which is a multi-gene family locus on common bean chromosome Pv04. The alpha-amylase affects bruchid development, but the arcelins are more effective (Cardona et al. 1989). Previous breeding efforts introgressed the arcelins from the wild common beans into cultivated beans, resulting in the development of weevil-resistant varieties of common bean (Kornegay and Cardona 1991; Kornegay, Cardona, and Posso 1993; Mbogo, Davis, and Myers 2009). However, this introgressed resistance was more effective against the Mexican bean weevil than the common bean weevil (Myers et al. 2001; Osborn et al. 1988). The tepary bean (*P. acutifolius*) accession G40199 is highly resistant to the common bean weevil (Goossens et al. 2000), causing adult mortality, reduced adult emergency, and prolonged larval development (Shade, Pratt, and Pomeroy 1987). The arcelins encoded by the APA locus appear to be the source of this resistance (Kusolwa and Myers 2011). Resistance from tepary bean has been introgressed into common bean, resulting in the development of common bean weevil resistant germplasm, mainly dark-red kidneys, such as AO-1012-29-3-3A (AO-3A) (Kusolwa et al. 2016).

Previous quantitative trait locus (QTL) studies have provided insights into the genetic architecture of AO resistance. Blair et al. (2010) mapped the APA locus and the associated Arc 1 allele on Pv04 in a mapping population where RAZ 106, a bruchid resistant wild accession, was used as a parent. Kamfwa et al. (2018) mapped the APA locus on Pv04 and reported additional resistance QTL on Pv06 using an Andean population of recombinant inbred lines derived from a cross of AO-3A and Solwezi. Li et al. (2022) also mapped one QTL for bruchid resistance on Pv06 using a bruchid-resistant black bean. However, this QTL was in a different physical position to that of Kamfwa et al. (2018).

Since its introgression from tepary, deployment of the APA-based resistance has been limited to a few market classes such as dark-red kidneys (AO-1012-29-3-3A and SUA-Red), light-red kidney bean (SUA-Karanga), and red-speckled kidney bean (SUA-Rose) (Kusolwa et al. 2016). Knowledge on the stability of the APA-based resistance in varied market classes of common bean is limited.

The objectives of this study were to (i) introgress the APA locus into selected market classes of Andean gene pool of common bean and (ii) determine the stability of APA-conditioned resistance to AO in multiple market classes of common bean.

2 | Materials and Methods

2.1 | Plant Materials

In this study, 571 $F_{5:7}$ breeding lines were evaluated for AO resistance. These breeding lines came from seven breeding populations derived from crosses of AO-resistant parent AO-1012-29-3-3A (AO-3A) with seven AO-susceptible Andean parents. AO-3A is a dark-red kidney bush (type I) breeding line that was developed and released cooperatively by the Sokoine University of Agriculture, Oregon State University, USDA-ARS, and the University of Puerto Rico (Kusolwa et al. 2016). The genetic basis of AO resistance in AO-3A is the APA locus that was introgressed from the wild tepary bean genotype G40199 (Kusolwa et al. 2016). The APA locus was later mapped on chromosome Pv04 (Kamfwa et al. 2018). AO-3A possesses the genes *I* and *bc-1*^{2}, which confers resistance to bean common mosaic virus and bean common mosaic necrotic virus. Furthermore, AO-3A is resistant to some races of *Colletotrichum lindemuthianum*, causative pathogen of anthracnose (Mungalu et al. 2020) and to aphids (Zimba et al. 2022).

The ADP genotypes were selected from the Andean Diversity Panel (Cichy et al. 2015) based on superior agronomic performance and to have a representation of the major market classes preferred in the three Southern African countries (Malawi, Mozambique, and Zambia). The seven AO-susceptible Andean parents were ADP1 (Rozi Koko), ADP466 (Mwezi Moja),

ADP725, ADP763 (Punda), ADP770, Lusaka, and Kabulangeti. Rozi Koko is a large-seeded and red-mottled variety from Tanzania. Mwezi Moja is a purple-speckled variety from Kenya. ADP725 is a CIAT breeding line with a cranberry (aka sugar) seed type and kidney seed shape. ADP 763 is a purple-speckled landrace from Tanzania. ADP770 is a Canadian breeding line with a cranberry seed type. Kabulangeti (purple-speckled seed) and Lusaka (Manteca yellow seed color) are both landraces from Zambia. The resistant parent AO-3A was crossed to the seven susceptible parents at the University of Zambia (UNZA) to develop seven populations of $F_{5:7}$ RILs using the single seed descent method. The number of RILs for each of the seven populations is shown in Table 1.

2.2 | Phenotypic Screening for *A. obtectus*

A spontaneous colony of AO was obtained from old stocks of beans from the UNZA Bean Breeding program in 2020. The colony was multiplied, raised, and maintained in the insectarium at UNZA on susceptible lines ADP 33 (Kijivu), ADP 466 (Mwezi Moja), and ADP 763 (Punda) following previously described protocol (Kamfwa et al. 2018; Kusolwa and Myers 2011). To multiply the colony, approximately 300 adult weevils were infested on 400-g seed in 500-mL plastic rearing jars that were covered with an insect net to allow for ventilation. Screening of breeding lines and their parents for AO resistance was conducted concurrently at the two locations, UNZA in Lusaka and Zambia Agricultural Research Institute (ZARI) in Kasama in the northern part of Zambia using the same genotypes and same population of weevils. This double screening was conducted to avoid escapes (false negatives). Further, the lines that were resistant in the initial screening were re-screened to confirm their resistance. Notably, insect resistance is more difficult to evaluate due to the high chances for escape in the field or artificial testing (Yencho, Cohen, and Byrne 2000). Screening at each location was conducted following previously described protocols (Kamfwa et al. 2018; Kusolwa and Myers 2011). At each location, a completely randomized design was used with two replications. The landraces Lusaka and Kabulangeti beans were susceptible checks, while AO-3A was a resistant check. The experimental units were 250-mL plastic jars. The seed used in the experiment was put in a freezer maintained at −20°C for 4–5 days to kill off any eggs that might have been carried from the field. This was done to ensure that the number of perforations and the number of damaged seed scored at 45 and 60 days were as a result of the artificial infestation and not the natural infestation from the field. Seeds were removed from the freezer and left at room temperature to acclimate for 24 h. After that, 10 seeds of each line were placed in separate 250-mL plastic jars, and then 10 unsexed adult weevils were introduced into the jars. The lids of the jars were cut open on top, and nets were placed on to allow for ventilation but prevent escape of adult weevils. The jars were then placed in the insectarium, maintaining a temperature of 28°C for 60 days. Weevil damage on the seed was assessed at 30, 45, and 60 days after infestation. At 30 days, the initial 10 adult weevils infested on the seed would have died, and these dead weevils were removed from the jars. Removing the 10 weevils used in the infestation helped in knowing the total number of new adults that emerged. At 45 and 60 days, weevil resistance of a particular line was assessed based on the total number of perforations on the 10 seeds and percentage of damaged seed using the International Center for Tropical Agriculture (CIAT) scoring system. This system uses a scale of 1–5 based on the number of perforations on the seed, where 1 = *zero perforations*, 2 = *one to five perforations*, 3 = *6 to 10 perforations*, 4 = *11 to 15 perforations*, and 5 = ≥*16 perforations*. Counting the number of seeds with perforations out of the 10 seeds was used to assess percentage of seed damaged (Kamfwa et al. 2018). A seed with at least one perforation caused by a weevil was considered damaged. Further, soft seeds when pinched with fingers were also deemed damaged. Genotypes with scores of ≤2 at 45 days after infestation were considered to have some useful level of resistance, whereas those that maintained the score of ≤2 at 60 days after infestation are deemed to have high levels of resistance.

TABLE 1 | Pedigree information and total number of breeding lines from each of the seven breeding populations evaluated for common bean weevil resistance in the insectarium at the University of Zambia and Misamfu.

Population code/ pedigree	Number of lines	Number of resistant lines
AO-3A/ADP 763 (Punda)	241	7
AO-3A/ADP 1 (Rozi Koko)	54	5
AO-3A/ADP 770	47	0
AO-3A/ Kabulangeti	65	0
AO-3A/ADP 466 (Mwezi Moja)	34	0
AO-3A/Lusaka	106	3
AO-3A/ADP 725	24	1
Total	571	16

2.3 | Data Analysis

Statistical analyses for seed perforations score and percentage of seed damaged were conducted in R version 4.2.1. Normality tests indicated that the percentage seed perforation score was not normally distributed; therefore, it was transformed using Box Cox transformation. Transformed data was used in the analysis of variance (ANOVA) conducted following the mixed model:

$$Y = \mu + G + L + G * L + E$$

where Y was the response variable, for example, seed perforation score; G was the fixed, variable effect of genotype; L was the random effect of location; G*L was the random effect of the interaction between genotype and location; and E was the random error.

3 | Results

3.1 | Evaluation of the 571 Breeding Lines for Resistance to Common Bean Weevil

ANOVA results for the 571 breeding lines indicated a significant genotypic effect on both seed perforation score ($p < 0.05$) and percentage of seed damaged ($p < 0.05$). The effect of location and

its interaction with the genotype was not significant ($p > 0.05$). Seed perforation score ranged from 1–5, with a mean of 3.92 for the 571 breeding lines. Seed perforation scores were skewed to susceptibility (Figure 1). The percentage of damaged seeds ranged from 0% to 100%, and the population average was 56.10%. Screening results of individual populations are presented next.

3.2 | AO-3A/ADP763 Population

Of the 241 lines from AO-3A/ADP763 population, one line (3A/ADP763-86a) had a score of one, while 18 (7.5%) lines had scores of less or equal to two. Parent AO-3A held its resistance with a score of two, while parent ADP 763 was highly susceptible (score of 5) (Figure 2A). Out of 241 lines screened for percentage seed damage, 24 lines had seed damage ranging from 0% to 10%. Parents ADP763 had over 90% of its seed damaged while AO-3A had only 20%. This population gave the highest number (7) of resistant lines in comparison to the other six populations. This could be attributed to the large population size and, to some extent, possible existence of minor QTL for resistance from the supposedly susceptible parent (Kamfwa et al. 2018).

3.3 | AO-3A/ADP1 Population

Of the 54 lines from AO-3A/ADP1 population, five (9.2%) lines were resistant with scores of one, while three lines had less or equal to two. Most of the lines were susceptible, including the two parents (score > 2) (Figure 2B). Out of 54 lines screened for percentage seed damage, seven lines had less than 10% of seed damage. Parents ADP1 had 100% of its seed damaged, while AO-3A had 50% of its seed damaged. The percentage of seed damage ranged from 0% to 100% for the breeding lines. This is the population that gave the second largest number (5) of resistant lines.

3.4 | AO-3A/ADP725 Population

Of the 24 lines from AO-3A/ADP 725 population screened, only one line (4.8%) was resistant, with a score of one. Both parents, AO-3A and ADP 725, had susceptible scores of 4.5 and 5, respectively. Most lines were skewed towards susceptibility (Figure 2C). One line (3A/ADP725-27) out of 24 lines screened had less than 10% of its seed damaged. Both parents had over 70% of their seeds damaged.

3.5 | AO-3A/Kabulangeti Population

A total of 65 breeding lines from AO-3A/Kabulangeti (KAB) population was evaluated, and all were susceptible, with scores ranging from 3 to 5 (Figure 2D). The parent A0-3A had a score of 2, while KAB had a score of 4. Out of 65 lines screened for percentage seed damage, none of the lines had less than 20% of their seeds damaged. The parent KAB had 42.5% of its seed damaged, while AO-3A had 25%. The percentage of damaged seeds ranged from 32.5% to 97.5% for the breeding lines. This population had no resistant lines obtained, and it could be attributed to the genetic background of the susceptible Kabulangeti parent, which could have affected the introgression of the APA locus into the breeding lined that were developed from this cross (Bornowski et al. 2023).

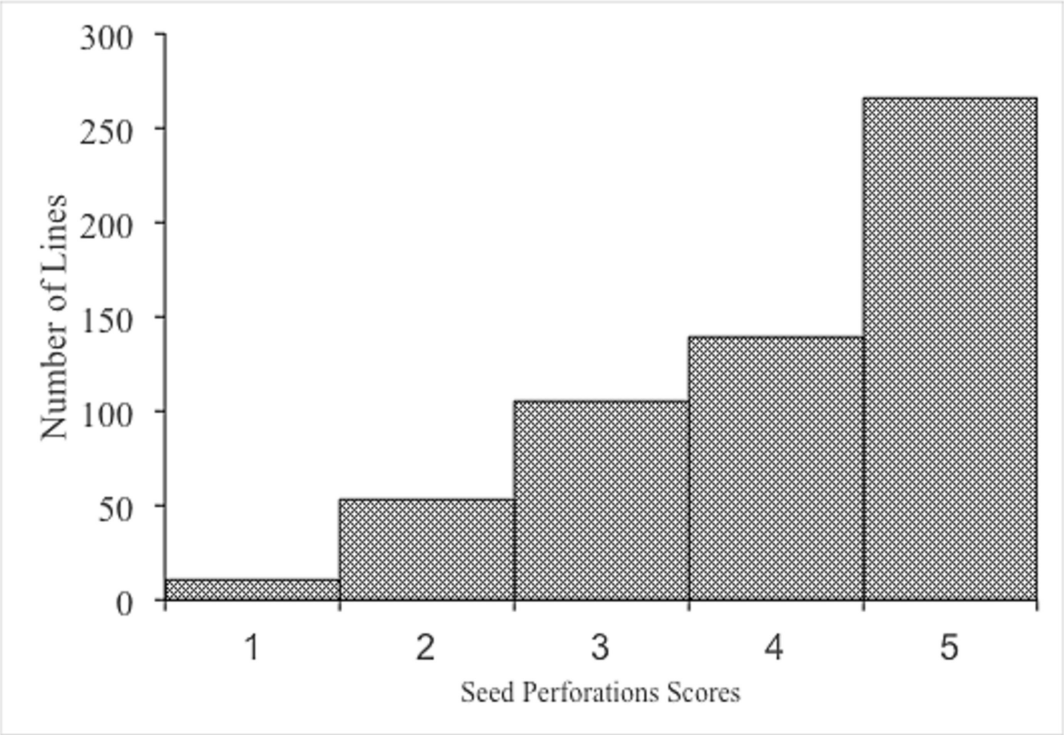


FIGURE 1 | Frequency distribution graph of seed perforations scores for 571 breeding lines evaluated for resistance to *A. obtectus* at 60 days post-infestation in the insectarium at the University of Zambia and Misamfu.

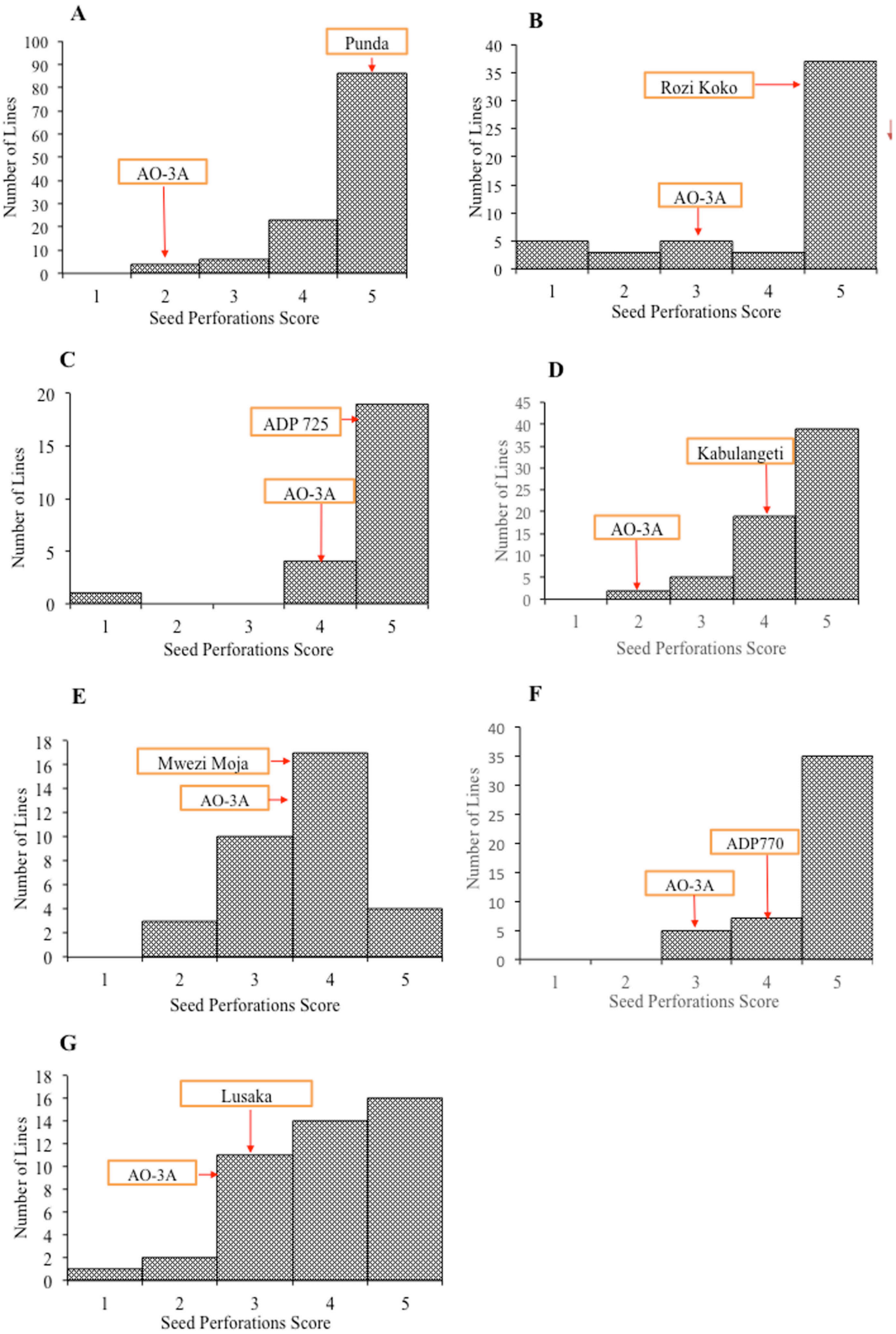


FIGURE 2 | Frequency distribution graph of seed perforations scores of seven breeding populations evaluated for resistance to *A. obtectus* at 60 days post-infestation in the insectarium at the University of Zambia and Misamfu.

3.6 | AO-3A/ADP466 Population

A total of 34 breeding lines was evaluated, of which three (8.8%) lines were resistant with scores of less or equal to two. There were no lines without perforations (Figure 2E). Both parents had scores of 4. For the percentage of seed damage, out of 34 lines screened, there were no lines with less than 10% of seed damage. Both parents A0-3A and ADP 466 had 50% of their seeds damaged. Percent of damage ranged from 20% to 80% for the breeding lines.

3.7 | AO-3A/ADP770 Population

All 47 breeding lines from AO-3A/ADP770 population were susceptible, with scores ranging from 3 to 5 (Figure 2F). The scores for parents A0-3A and ADP 770 were 3 and 4, respectively. For the percentage of seed damage, out of 47 lines screened, none of the lines had less than 20% of their seeds damaged. The parent ADP 770 had 65.5% of its seed damaged, while AO-3A had 27.5%. The percentage of damage ranged from 25% to 97.5% for the breeding lines. This population as well did not produce any resistant line, and it might be as a result of difficulties in introgressing the APA locus into the genetic background of cranberry (ADP 770) (Bornowski et al. 2023); also the smaller population size could have affected the recovery rate of resistant lines (Kamfwa et al. 2018).

3.8 | AO-3A/LSK Population

A total of 106 breeding lines was evaluated from AO-3A/Lusaka (LSK) population, of which two (1.9%) lines were resistant with a score of one, while 20 lines had scores of less or equal to two (Figure 2G). Parent AO-3A had a score of 3, while LSK had a score of 4. For the percentage of seed damage, out of 106 lines screened, one line had none of its seeds damaged (AO-3A-LSK-3), while 13 lines had seed damage ranging from 0% to 10%. The parent Lusaka had 45% of its seed damaged, while AO-3A had 12.5%. The percentage of damage ranged from 0% to 100% for the breeding lines. This population gave three resistant lines, and this could be as a result of the large population size and contribution of resistant alleles at the minor-QTL effects.

3.9 | Resistant Breeding Lines Identified From the 571 Breeding Lines

When screening results of the seven populations were aggregated, 16 breeding lines out of the 571 evaluated were identified as resistant to AO (Table 2). These 16 had seed perforation scores of less or equal to two (Figure 3). The percentage of seed damaged for the 16 lines ranged from 0% to 50%. These 16 lines are in diverse market classes (Table 2).

TABLE 2 | Market classes of 16 breeding lines identified as resistant to *A. obtectus* from screenings conducted at the University of Zambia and Misamfu.

Breeding line name	Seed color/market class
AO-3A-ADP 763-10	Grey speckled
AO-3A-ADP 763-46	Grey speckled
AO-3A-ADP 763-66	Tan
AO-3A-ADP 763-86	Dark red speckled
AO-3A-ADP 763-109	Dark red speckled
AO-3A-ADP 763-24	Grey speckled
AO-3A-ADP 763-45	Dark red speckled
AO-3A-ADP 1-2	Red mottled
AO-3A-ADP 1-13	Purple mottled
AO-3A-ADP 1-28	Purple mottled
AO-3A-ADP 1-47	Tan
AO-3A-ADP 1-51	Tan
AO-3A-LSK-3	Specked
AO-3A-LSK-11	Yellow
AO-3A-ADP 725-27	Cranberry
LSK-41-50-42	Brown

4 | Discussion

A. obtectus is the most important post-harvest pest of common bean. The APA locus on chromosome Pv04, originally derived from *P. acutifolius*, provides resistance to the common bean weevil. The APA locus has only been deployed in limited market classes of common bean such as dark-red kidneys, AO-1012-29-3-3A, SUA-Red, SUA-Karanga (light red kidney bean), and SUA-Ros (red-speckled kidney bean) (Kusolwa et al. 2016); however, there are reports of loss of weevil resistance of progenies during introgression through backcrossing of arcelins from the wild bean into cultivated bean (Myers et al. 2001; Osborn et al. 1988). The stability of resistance conditioned by APA locus in genetic backgrounds of various common bean market classes has been yet to be studied. This study explored the stability of the APA locus resistance in genetic backgrounds of varied market classes of common bean.

A total of 571 $F_{5:7}$ breeding lines from seven breeding populations derived from crosses of AO-3A with seven Andean varieties of common bean genotypes was evaluated for resistance to AO. Of the 571 lines screened, 16 (2.8%) lines were resistant to AO. Six resistant breeding lines had higher resistance to AO with scores ranging from 1 to 1.3 than that of AO-3A, which had a score of 2. The 16 resistant breeding lines were in variable market classes, such as red mottled, purple mottled, grey speckled, tan, yellow, brown, cranberry, and dark red speckled, indicating the stability of APA-conditioned resistance in varied genetic backgrounds of common bean. The recovery rate of resistance in the breeding lines was generally very low. In fact, there were no resistant lines for some of the crosses. It is difficult to ascertain the reason/s for this low recovery rate. The low recovery rate of AO resistance in progenies is not unique to this study and has previously been reported. Kamfwa et al. (2018) screened 210 RILs derived from the cross of AO-3A and Solwezi for resistance to AO, and only 15 were resistant, representing a low resistance recovery rate of 7.1%. Kornegay and Cardona (1991) reported a low recovery rate of resistant progenies in a cross between resistant wild common bean and cultivated common bean. Bornowski et al. (2023) phenotyped 157 tepary diversity panel (TDP) (37 cultivated, 6 weedy, and 114 wild) lines for seed damage, and only two lines showed lower percentage of seed damage at 45 DAI, which later showed 60% damaged seed, representing a low recovery rate. The low recovery rate of resistant lines in segregating populations suggests that weevil resistance is a complex trait that is polygenically controlled (Bornowski et al. 2023). The breeding implication of the low recovery rate is that breeding programs seeking to breed for resistance to AO may require large breeding population sizes to increase the chances of recovering resistant progenies. It was interesting, however, to notice that some crosses provided more resistant lines than other populations, suggesting a possible role of

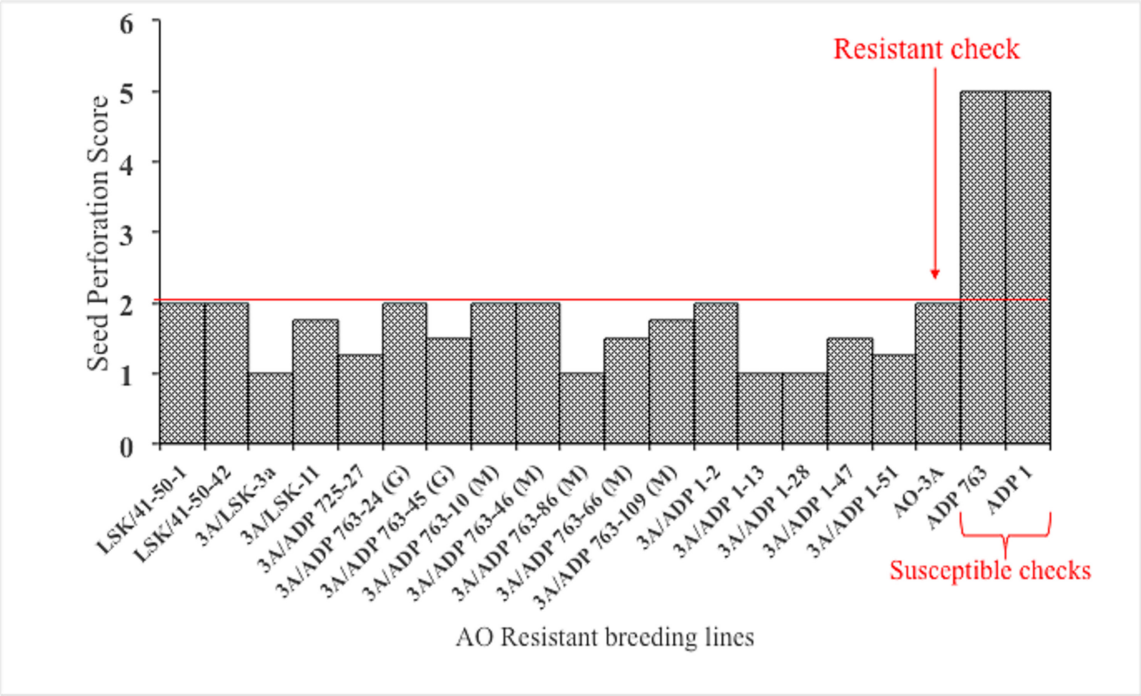


FIGURE 3 | Seed perforation scores of 16 resistant breeding lines and checks assessed at 60 days post infestation with *A. obtectus*.

genetic background in the recovery rate of the APA-conditioned AO resistance. Further, some crosses provided lines with higher resistance levels than the resistant parent AO-3A. This transgressive segregation could be attributed to the possible existence of minor QTL for resistance in the supposedly susceptible parents. Kamfwa et al. (2018) reported minor-effect QTL on Pv06, and the susceptible parent Solwezi contributed the resistance allele at that QTL. It is plausible that the parents ADP1, ADP763, and Lusaka, which produced the lines with superior AO resistance to that of AO-3A, might possess resistance alleles at the minor-effect QTL for AO resistance. The APA locus, possibly acting additively with the minor-effect QTL, could have been the basis of the observed higher AO resistance for some of the breeding lines than the parent AO-3A. The breeding lines identified to be more resistant than the parent AO-3A offer an opportunity for further genetic enhancement of common bean for resistance to AO. The 16 resistant lines identified in this study are in variable market classes. This variability offers an opportunity to circumvent the breeding challenge of various seed colors in progenies from crosses between parents of different seed colors. Breeding programs seeking to improve red-mottled can use the red-mottled resistant lines. This way, the breeder does not have to be concerned about some progenies possessing AO resistance but having undesirable seed color.

Insect resistance is notably more difficult to evaluate due to the high chances for escape in the field or artificial testing and the need to maintain live colonies, specialized staff, and appropriate infestation conditions (Yencho, Cohen, and Byrne 2000). This was also observed in this study as more than one screening was done to confirm if the lines that showed resistance in the initial screening were truly resistant. The other challenges of phenotyping for AO is the long duration of the assay (60 days) and significant effect of rearing environment on successful infestation (Mazaheri 2018; Singh and Schwartz 2010; Tigist 2020).

It is worth noting the challenges that are associated with conventional phenotypic screening for insect resistance using insect infestation, which is time consuming, coupled with high chances of escapes in artificial testing and the need for specific environmental conditions (Mazaheri 2018; Singh and Schwartz 2010; Tigist 2020). Therefore, developing and using molecular markers is the efficient way to reduce the long period for phenotypic screening for *A. obtectus* and speed up the breeding process for the development of bruchid-resistant varieties.

Currently, the 16 resistant breeding lines in this study were shared with collaborators in Malawi, Mozambique, and the United States for agronomic performance and cooking time evaluation and molecular marker validation.

5 | Conclusions

This study has identified 16 resistant lines from the 571 breeding lines evaluated for AO resistance. These resistant breeding lines are in variable market classes/seed colors, and some have higher AO resistance than the resistant parent AO-3A. These resistant lines offer the bean breeding community an opportunity for further genetic enhancement of weevil resistance to mitigate the post-harvest losses caused by AO. Line AO-3A-ADP1-13 showed higher breed resistance to AO, faster cooking, has superior agronomic performance, and could be a possible candidate for use as a parental line in the breeding program or released as a variety.

Author Contributions

Modreen Chinji: Formal analysis; Investigation; Methodology; Writing – original draft. **Swivia Hamabwe:** Methodology. **Kuwabo Kuwabo:** Methodology. **Isabel Mugovu:** Methodology. **Rebeca Thole:** Methodology. **Maria Mazala:** Methodology. **Juan Manuel**

Osorno: Conceptualization; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Supervision; Writing – original draft; Writing – review and editing. **Phillip McClean:** Conceptualization; Funding acquisition. **Celestina Jochua:** Conceptualization; Funding acquisition. **Carlos Urrea:** Conceptualization; Funding acquisition. **Chikoti Mukuma:** Methodology. **Virginia Chisale:** Conceptualization; Funding acquisition. **Kelvin Kamfwa:** Conceptualization; Formal analysis; Methodology; Project administration; Funding acquisition; Supervision; Writing – original draft; Writing – review and editing.

Acknowledgments

This work was funded in whole (or part) by the United States Agency for International Development (USAID) under Agreement #7200AA18LE00003 as part of Feed the Future Innovation Lab for Legume Systems Research. Any opinions, findings, conclusions, or recommendations expressed here are those of the authors alone.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author.

References

Abate, T., and J. K. O. Ampofo. 1996. "Insect Pests of Beans in Africa: Their Ecology and Management." *Annual Review of Entomology* 41, no. 1: 45–73.

Blair, M. W., C. Muñoz, H. F. Buendía, J. Flower, J. M. Bueno, and C. Cardona. 2010. "Genetic Mapping of Microsatellite Markers around the Arcelin Bruchid Resistance Locus in Common Bean." *Theoretical and Applied Genetics* 121, no. 2: 393–402.

Bornowski, N., J. P. Hart, A. V. Palacios, et al. 2023. "Genetic Variation in a Tepary Bean (*Phaseolus acutifolius* A. Gray) Diversity Panel Reveals Loci Associated with Biotic Stress Resistance." *The Plant Genome* 16, no. 3: e20363.

Cardona, C. 2004. *Crop Post-Harvest: Science and Technology*. Durables: Blackwell Science.

Cardona, C., C. E. Posso, J. Kornegay, J. Valor, and M. Serrano. 1989. "Antibiosis Effects of Wild Dry Bean Accessions on the Mexican Bean Weevil and the Bean Weevil (Coleoptera: Bruchidae)." *Journal of Economic Entomology* 82, no. 1: 310–315.

Chireceanu, C., P. Cardei, A. Geicu, I. Florescu, F. Burnichi, and M. Călin. 2022. "Insecticidal Effect of Pătârlagele Diatomaceous Earth Against *Acanthoscelides obtectus* Adults." *Romanian Agricultural Reasearch* 39: 649–659.

Cichy, K. A., T. G. Porch, J. S. Beaver, et al. 2015. "A *Phaseolus vulgaris* Diversity Panel for Andean Bean Improvement." *Crop Science* 55, no. 5: 2149–2160.

Goossens, A., C. Quintero, W. Dillen, et al. 2000. "Analysis of Bruchid Resistance in the Wild Common Bean Accession G02771: No Evidence for Insecticidal Activity of Arcelin 5." *Journal of Experimental Botany* 51, no. 348: 1229–1236.

Howe, R. W., and J. E. Currie. 1964. "Some Laboratory Observations on the Rates of Development, Mortality and Oviposition of Several Species of Bruchidae Breeding in Stored Pulses." *Bulletin of Entomological Research* 55, no. 3: 437–477.

Kamfwa, K., J. S. Beaver, K. A. Cichy, and J. D. Kelly. 2018. "QTL Mapping of Resistance to Bean Weevil in Common Bean." *Crop Science* 58, no. 6: 2370–2378.

Kornegay, J., C. Cardona, and C. E. Posso. 1993. "Inheritance of Resistance to Mexican Bean Weevil in Common Bean, Determined by Bioassay and Biochemical Tests." *Crop Science* 33, no. 3: 589–594.

Kornegay, J. L., and C. Cardona. 1991. "Inheritance of Resistance to *Acanthoscelides obtectus* in a Wild Common Bean Accession Crossed to Commercial Bean Cultivars." *Euphytica* 52, no. 2: 103–111.

Kusolwa, P. M., and J. R. Myers. 2011. "Seed Storage Proteins ARL2 and its Variants from the Apalocus of Wild Tepary Bean G40199 Confers Resistance to *Acanthoscellides obtectus* When Expressed in Common Beans." *African Crop Science Journal* 19, no. 4: 255–265.

Kusolwa, P. M., J. R. Myers, T. G. Porch, Y. Trukhina, A. González-Vélez, and J. S. Beaver. 2016. "Registration of AO-1012-29-3-3A Red Kidney Bean Germplasm Line With Bean Weevil, BCMV, and BCMNV Resistance." *Journal of Plant Registrations* 10, no. 2: 149–153.

Li, X., Y. Tang, L. Wang, Y. Chang, J. Wu, and S. Wang. 2022. "QTL Mapping and Identification of Genes Associated With the Resistance to *Acanthoscelides obtectus* in Cultivated Common Bean Using a High-Density Genetic Linkage Map." *BMC Plant Biology* 22, no. 1: 1–15.

Mazaheri, L. I. 2018. "Development of a Molecular Marker to Track APA G40199 Introgression in Common Bean for Bruchid Resistance." Master's Thesis, North Dakota State University.

Mbogo, K. P., J. Davis, and J. R. Myers. 2009. "Transfer of the Arcelin-Phytohaemagglutinin-α Amylase Inhibitor Seed Protein Locus From Tepary Bean (*Phaseolus acutifolius* A. Gray) to Common Bean (*P. vulgaris* L.)." *Biotechnology* 8, no. 3: 285–295.

Mishili, F. J., A. Temu, J. Fulton, and J. Lowenberg-DeBoer. 2011. "Consumer Preferences as Drivers of the Common Bean Trade in Tanzania: A Marketing Perspective." *Journal of International Food & Agribusiness Marketing* 23, no. 2: 110–127.

Mulungu, L. S., E. N. Luwondo, S. O. W. M. Reuben, and R. N. Misangu. 2007. "Effectiveness of Local Botanicals as Protectants of Stored Beans (*Phaseolus vulgaris* L.) Against Bean Bruchid (*Zabrotes subfasciatus* Boh) (Genera: Zabrotes. Family: Bruchidae)." *Journal of Entomology* 4, no. 3: 210–217.

Mungalu, H., M. Sansala, S. Hamabwe, et al. 2020. "Identification of Race-Specific Quantitative Trait Loci for Resistance to *Colletotrichum lindemuthianum* in an Andean Population of Common Bean." *Crop Science* 60, no. 6: 2843–2856.

Mutungi, C., H. D. Affognon, A. W. Njoroge, J. Manono, D. Baributsa, and L. L. Murdock. 2015. "Triple-Layer Plastic Bags Protect Dry Common Beans (*Phaseolus vulgaris*) against Damage by *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae) During Storage." *Journal of Economic Entomology* 108, no. 5: 2479–2488.

Myers, J. R., J. Davis, D. Kean, S. Nchimbi-Msolla, and R. Misangu. 2001. "Backcross Breeding to Introduce Arcelin Alleles Into Improved African Bean Cultivars." In *Bean/Cowpea Collaborative Research Support Program. East Africa Proceedings: Bean Seed Workshop Arusha, Tanzania January*, 12–14.

Osborn, T. C., D. C. Alexander, S. S. M. Sun, C. Cardona, and F. A. Bliss. 1988. "Insecticidal Activity and Lectin Homology of Arcelin Seed Protein." *Science* 240, no. 4849: 207–210.

Paul, U. V., J. S. Lossini, P. J. Edwards, and A. Hilbeck. 2009. "Effectiveness of Products from Four Locally Grown Plants for the Management of *Acanthoscelides obtectus* (Say) and *Zabrotes subfasciatus* (Boheman) (Both Coleoptera: Bruchidae) in Stored Beans Under Laboratory and Farm Conditions in Northern Tanzania." *Journal of Stored Products Research* 45, no. 2: 97–107.

Petry, N., E. Boy, J. P. Wirth, and R. F. Hurrell. 2015. "The Potential of the Common Bean (*Phaseolus vulgaris*) as a Vehicle for Iron Biofortification." *Nutrients* 7, no. 2: 1144–1173.

Schoonhoven, A. 1976. "Pests of Stored Beans and Their Economic Importance in Latin America." In *Proceedings of the Symposium on*

Tropical Stored Product Entomology, 15th International Congress of Entomology, Washington, DC. 19–27 Aug. 1976. Entomol. Soc. Am., 691–698. College Park, MD.

Schoonhoven, A. V., C. Cardona, and J. Valor. 1983. "Resistance to the Bean Weevil and the Mexican Bean Weevil (Coleoptera: Bruchidae) in Noncultivated Common Bean Accessions." *Journal of Economic Entomology* 76, no. 6: 1255–1259.

Shade, R. E., R. C. Pratt, and M. A. Pomeroy. 1987. "Development and Mortality of the Bean Weevil, *Acanthoscelides obtectus* (Coleoptera: Bruchidae), on Mature Seeds of Tepary Beans (*Phaseolus acutifolius*) and Common Beans (*Phaseolus vulgaris*)." *Environmental Entomology* 16, no. 5: 1067–1070.

Singh, S., and H. Schwartz. 2010. "Breeding Common Bean for Resistance to Insect Pests and Nematodes." *Canadian Journal of Plant Science* 91, no. 2: 239–250.

Slumpa, S., and J. K. O. Ampofo. 1991. "Evaluation of Different Methods for the Control of Bean Bruchid (*Acanthoscelides obtectus*)." *Annual Report of Bean Improvement Cooperative* 34: 66–67.

Stathers, T. E., W. Riwa, B. M. Mvumi, et al. 2008. "Do Diatomaceous Earths Have Potential as Grain Protectants for Small-Holder Farmers in Sub-Saharan Africa? The Case of Tanzania." *Crop Protection* 27, no. 1: 44–70.

Tigist, S. G. 2020. "Common Bean (*Phaseolus vulgaris* L.) and the Bean Bruchid (*Zabrotes subfasciatus*): A Review." *International Journal of Research in Agriculture and Forestry* 7, no. 11: 21–31.

Uebersax, M. A., K. A. Cichy, F. E. Gomez, et al. 2023. "Dry Beans (*Phaseolus vulgaris* L.) as a Vital Component of Sustainable Agriculture and Food Security—A Review." *Legume Science* 5, no. 1: e155.

Yencho, G. C., M. B. Cohen, and P. F. Byrne. 2000. "Applications of Tagging and Mapping Insect Resistance Loci in Plants." *Annual Review of Entomology* 45, no. 1: 393–422.

Zimba, K. J., P. H. Sohati, K. Munyinda, K. Kamfwa, J. M. Roberts, and T. W. Pope. 2022. "Evaluation of Resistance to Black Bean Aphid (*Aphis fabae*) in Selected Varieties and Mutant Genotypes of Common Bean (*Phaseolus vulgaris* L.)." *Annals of Applied Biology* 181, no. 3: 298–308.