International Journal of Plant & Soil Science
4(2): 124-131, 2015; Article no.IJPSS.2015.013
ISSN: 2320-7035

SCIENCEDOMAIN ***international***
www.sciencedomain.org

Laboratory Assessment of the Levels of Resistance in Some Bean Varieties Infested with Bean Weevils (*Acanthoscelides obtectus and Zabrotes subfasciatus*)

Chancy B. Sibakwe[1] and Trust Donga[1*]

[1]Bunda College of Agriculture, University of Malawi, Box 219, Lilongwe, Malawi.

Authors' contributions

This work was carried out in collaboration between both authors. Author CBS designed and implemented the study. He performed the data collection, statistical analysis and wrote the draft of the manuscript. Author TD supervised the whole work and edited the final manuscript.

Article Information

DOI: 10.9734/IJPSS/2015/6806
Editor(s):
(1) Sławomir Borek, Faculty of Biology, Department of Plant Physiology, Adam Mickiewicz University, Poland.
Reviewers:
(1) Saad Farouk Mohamed Hussien, Agricultural Botany Department, Faculty of Agriculture, Mansoura University, Egypt.
(2) Anonymous, Gaziantep university, Turkey.
(3) Anonymous, Egerton University, Kenya.
(4) James D. Kelly, Dept. Plant, Soil and Microbial Sciences, Michigan State University, East Lansing MI, USA.
Complete Peer review History: http://www.sciencedomain.org/review-history.php?iid=680&id=24&aid=6288

Original Research Article

Received 11th September 2013
Accepted 12th March 2014
Published 30th September 2014

ABSTRACT

The main objective of the study was to assess levels of resistance of some selected bean (*Phaseolus vulgaris* L.) varieties to storage bruchids (Common bean weevil *Acanthoscelides obtectus* (Say) and Mexican weevil *Zabrotes subfasciatus*). Specifically it aimed at determining bruchids emergence and median development period, finding out the extent of damage caused by the bruchids on each variety and determining the weight loss incurred by each variety due to bruchids infestation. The experiment was laid out in a Complete Randomized Design (CRD) with four replicates. The study was conducted at Bunda college of Agriculture-Department of Crop and Soil Sciences-Entomology Laboratory. It was conducted from September 2012 to January 2013. Seven bean varieties, Kalima, Nasaka, BCB3, Sapelekedwa, BCB1, Bwenzilaana, and BCB2 were used as test materials. 100 g of bean seeds sample from each variety were put in kilner jars and were infested with 16 bruchids of common weevil and Mexican weevil, and it was carried in two phase; the first phase was used collect data for emerging insects and median development period;

**Corresponding author: E-mail: t.donga2012@gmail.com;*

and Second phase was used to collect other data parameters. The results at *P*=.05 only BCB2 was found to be significantly resistant to storage bruchids, Sapelekedwa was the only moderate resistant, Nasaka, BCB3 and Kalima were susceptible and finally Bwenzilaana and BCB1 were highly susceptible. The results also showed that the damage caused by the storage bruchids ranged from 20% in resistant varieties to 88% in high susceptible varieties while the weight loss ranged from 3% to 29%. It was concluded that the level of resistance among the seven varieties evaluated to two common bean weevils (*Acanthoscelides obtectus* and *Zabrotes subfasciatus*) differed significantly during the four and half month's storage period.

Keywords: Storage bruchids; DSI; percent damage and weight loss.

1. INTRODUCTION

Beans in Africa are regarded as the second most important source of human dietary protein and the third most important source of calories after maize and cassava [1]. The common bean *Phaseolus vulgaris* is one of the most important legumes grown by smallholder and large scale farmers in Malawi. Beans provide proteins to both poor and rich households.

Most farmers fail to realize the potential yield and full utilization of the produce due to many production constraints [2]. These are: inadequate production technologies; lack of information on suitable varieties to grow; poor rain distribution; diseases; and pest damage [3]. The latter factor is the most challenging when it comes to production and utilization of beans. Many farmers fail to utilize the harvest due to the damage caused by the storage insect pest [1]. In storage beans are mainly attacked by two types of bruchids; Common bean weevil, *Acanthoscelides obtectus* (Say) and Mexican weevil *Zabrotes subfasciatus.* These bruchids are reported to cause damage or losses of 40–100%, if the stored beans are not treated with chemicals like actellic dusts that control the storage pests [4].

Post-harvest losses of beans mostly occur during storage stage rather than during other stages like threshing or cleaning. There are several possible causes of these losses during storage: rotting due to exposure to moisture; theft; and storage pest attacks [5,6]. Pests which attack beans during storage are rodents and bruchids (A. *obtectus* and *Z. subfasciatus*). Bruchids attack is considered as the major cause of the losses and their levels of damage or loss depends on whether the stored beans are treated with chemicals or not.

Another factor that contributes to the level of damage or loss due to bruchids is the type of variety. The level of damage is different among varieties; this is due to physical characteristics such as seed size and seed coat and seed color, which different varieties possess [7,8]. The above physical characteristic of the seed are responsible for differences in susceptibility to bruchids among different bean varieties. Since the resistance or susceptibility depend on the type of variety, therefore it was necessary to carry out an experiment or a trial to assess the levels of resistance of some selected bean varieties under storage bruchids infestation.

2. MATERIALS AND METHODS

2.1 Area of Study

The experiment was conducted from September, 2012 to January, 2013 at the Department of Crop and Soil Sciences Entomology Laboratory of Bunda college of Agriculture, Lilongwe.

2.2 Experimental Materials

2.2.1 Bean samples

Bean seeds of seven released bean varieties commonly grown by farmers in Malawi were used. These varieties were: Kalima (an introduced line from CIAT), Nasaka, BCB3, Sapelekedwa, BCB1, Bwenzilaana, and BCB2 were used as test materials. These varieties were collected from Bean/Cowpea Collaborative Research Support Programme (B/C CRSP) of Bunda college of Agriculture. The seeds were dried up to 11-12% moisture content and were not previously treated with chemicals. The characteristics of the varieties are described in Table 1.

Table 1. Characteristics of the dry bean varieties tested in experiment for resistance to (*Z. subfasciatus and A. obtectus*)

Variety	Characteristics			
	Seed size	**Colour and shape**	**Growth habit**	**Maturity**
Kalima	Large seeded	Speckled red kidney	Dwarf	100-110 days
Nasaka	Large seeded	Tan seed coat kidney	Dwarf	100-110 days
Bwenzilaana	Medium	Roundish yellow seed coat	Dwarf	90-100 days
Sapelekedwa	Large seeded	Blue	Dwarf	77-100 days
BCB1	Medium seeded	Speckled	Dwarf	80-90 days
BCB2	Small seeded	Blown	Climber	90-100 days
BCB3	Medium seeded	Speckled	Climber	90-100 days

Source: Bean/Cowpea Collaborative Research Support Programme (B/C CRSP) (2010), Bunda College

2.2.2 Bean bruchids

Adult insects of *A. obtectus* and *Z. subfasciatus* species were collected from Chitedze Research Station (CRS) and students' research farm store room with naturally infested beans. A laboratory culture was then established at Entomology laboratory by allowing the collected samples of insects to lay eggs (oviposit) on Nasaka variety. The two bruchids species were maintained separately in the laboratory after collection. About 900 insects were reared in 0.5kg of Nasaka seed using 2L Kilner glass bottle jars. The jars were covered with muslin cloth.

2.3 Experimental Design

The trial had 7 treatments laid out in complete randomized design (CRD) and each treatment was replicated 4 times. The treatments were Kalima, Nasaka, BCB3, BCB2, BCB1, Bwenzilaana and Sapele kedwa. Seed samples were disinfested by freezing at temperature of -3 to 1°C for one week. This was done to ensure that any eggs or adult insects from the field were killed, as they might affect the results of the experiment. Seed samples were then removed from the deep freezer and placed in the room for conditioning for two weeks. No insect emerged from the disinfested seed at the end of the two weeks.

2.3.1 First phase of the experiment

Seed samples weighing 100g were placed in transparent plastic kilner jars. The bean samples in each bottle were infested with 16 randomly selected adult insects, but without sex determination. Out of 16 bruchids 8 were Mexican weevils and other 8 were common bean weevils. Weevils were allowed to lay eggs for ten days, then after they were removed. To prevent the bruchids from escaping, a muslin cloth was used to cover the mouth of each bottle and held in place by a rubber band.

2.3.2 Second phase of the experiment

The bean samples were re-infested in the same way as first phase and were not remove from the kilner jars. This was done to collect data for other parameters apart from those mentioned above.

2.4 Data Collection

The infested seed were kept for 12 days to allow for oviposition to take place. The following data was collected: Number of emerging insects, median development period and number of damaged and undamaged seed, number holes per seed, weight of damaged and undamaged seeds and weight of whole sample.

2.5 Resistance Classification

Resistance was measured by the number of holes, number of holes per seed, seed weight loss incurred due to damage caused by bruchids, the number of adult bruchids that emerged, and the period required for the insects to develop within the bean seeds.

Seed weight loss, was calculated as follows:

$$\%\ \textbf{weight loss} = \frac{(IGW - FGW)}{IGW\ of\ sample} \times 100$$

Where:

IGW = initial seed weight for the sample
FGW = final seed weight

The number of holes per seed (NHPS), was calculated as follows:

$$\textbf{NHPS} = \frac{NH}{Nd}$$

Where,

NHP = Number of Holes per Seed.
NH = Number of holes
Nd = Number of damaged seeds

The number of adult bruchid that emerged and the median development period were combined to calculate the susceptibility index (Dobie, 1974) for each variety using the equation:

$$DSI = \frac{\textbf{Loge Y}}{t} \times 100$$

Where;

Y = total number of adult bruchid emerged
t = median development period

Based on Dobie susceptibility Index, bean varieties were grouped into four resistance Classes (Table 2).

2.6 Data Analysis

Data was entered in Genstat and Microsoft excel and subjected to analysis of variance (ANOVA) procedure for calculating F probabilities and coefficient of variation for investigated variables. Least Significance difference (LSD) test was used to separate means.

3. RESULTS AND DISCUSSION

3.1 Dobie Index, Development Period and Bruchid Emergence

The seven varieties assessed under laboratory conditions showed significant variations in expression of resistance to bruchids. Significant differences (P = .05) were observed in DSI, bruchids emergence and development period (Table 3). The variety BCB2 performed better than the rest of the varieties (DSI 4.9). Sapelekedwa scored second with DSI of 9.4 and BCB1 performed poorly as it had DSI of 17.4. Bruchids in BCB2 had significant (P = .05) longer development period (45 days). On BCB1, bruchids had shorter period (27 days), but their differences were not significant with Kalima at P = .05. There were also significant differences on development period on Sapelekedwa, Nasaka, BCB3, and Bwenzilana at probability level of 5 %. There was high emergence of bruchids on BCB1 (110 bruchids) and low emergence on BCB2 (only 9 bruchids emerged in a period of 45 days). The results statistically showed significant differences.

A frequency distribution of the 7 varieties, based on the DSI, showed that 14% of the varieties tested were resistant, 14% moderate resistant, 43% susceptible and 29 % highly susceptible to bruchids (Fig. 1). Only BCB2 was in resistant category, Sapelekedwa in moderate resistant, Nasaka, BCB3 and Bwenzilana were susceptible and finally Kalima and BCB1 were in last category, highly susceptible.

3.2 Seed Damage Due to Insect Pest Infestation after 3.5 Month

In terms of numbers of holes, BCB1 had the highest number of holes (316 holes) caused by the bruchids as compared to the rest of the varieties (Table 4). In BCB2 there was low number of holes appeared (20 holes) and the other remaining varieties were between the two mentioned varieties and statistically similar. The number of holes per one seed ranged from 1-3 (Table 4). It was highly observed in Nasaka and Sapele kedwa (3 holes per seed), moderately observed in BCB3, Bwenzilana, Kalima and BCB1 (2 holes per seed) and finally in BCB2, there was low number of hole per seed (1 hole per seed). These differences were not significant. BCB1 suffered the highest seed damage, Table 4. Kalima (37%) was second from BCB1, while BCB2 suffered the least damage.

Table 2. Classification of resistance based on to DSI

Dobie Susceptibility Index (DSI)	Resistance class
0-5	Resistant
6-10	moderate resistant
11-15	Susceptible
16 and above	More Susceptible

3.3 Seed Damage Due to Insect Pest Infestation (after 4.5 Months)

3.3.1 Seed damage

BCB1 recorded the highest number of holes caused by bruchids (2397 holes) as compared to the rest of the varieties (Table 5). Holes per seed ranged from 1 to 7 (Table 5). Significant differences (P = .05) were observed in BCB2, while in other varieties; Nasaka and BCB1, Sapelekedwa and Bwenzilaana, BCB3 and Kalima. Percent seed damage ranged from 20% to 88 % (Table 5). BCB1 (86%) was second from Kalima; while in BCB2 (20%) there was low percent damage.

Table 3. The DSI, development period and number of bruchids emerged on each variety

Variety	Development period	Bruchid emerged	DSI
BCB2	45^{a}	9^{f}	4.9
Sapelekedwa	38^{b}	36^{e}	9.4
Nasaka	36^{bc}	51^{d}	11.0
BCB3	33^{cd}	64^{c}	12.7
Bwenzilaana	32^{cd}	60^{c}	12.8
Kalima	28^{e}	91^{b}	16.3
BCB1	27^{e}	110^{a}	17.4
Grand mean	34	63	12.3
F(SIG)	.001	.001	.001
LSD	4	14	0.8
CV (%)	7.6	15.7	4.6%

Note: Means annotated by the same letter (s) are significantly the same

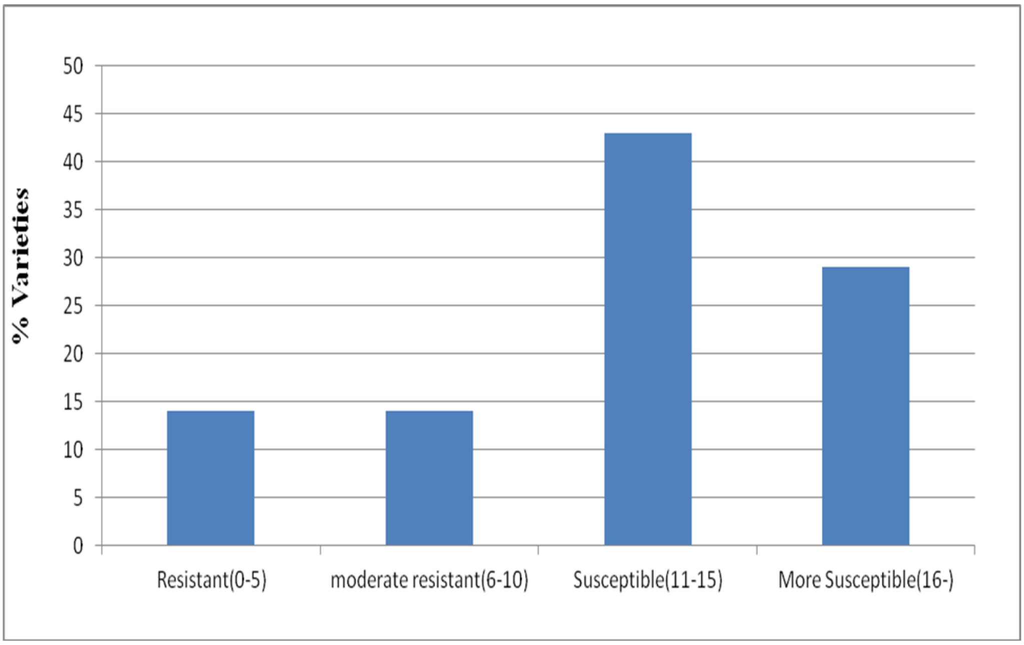

Fig. 1. Frequency distribution of 7 bean varieties for resistance to bruchids under laboratory infestation

Table 4. Extent of damages caused by bruchids after 3.5 months

Variety	Number of undamaged seed	Number of damaged seed	Number of holes	Holes per seed	Percent damage
BCB2	607	15	20^{c}	1	2^{b}
Sapele kedwa	164	95	247^{ab}	3	37^{a}
Nasaka	137	91	256^{ab}	3	40^{a}
BCB3	168	92	201^{ab}	2	36^{a}
Bwenzilaana	203	96	177^{bc}	2	32^{a}
Kalima	137	78	207^{ab}	2	37^{a}
BCB1	236	171	316^{a}	2	42^{a}
Grand mean	236	91	204	2	32
F(SIG)	.001	.001	.013	.424	.024
LSD	53	51	191	1	23
CV (%)	15	38	56	39	47

Note: Means annotated by the same letter (s) are significantly the same

Table 5. Extent of damages caused by bruchids after 4.5 months

Variety	Number of undamaged seed	Number of damaged seed	Number of holes	Holes per seed	Percent damage
BCB2	499	124	151^{c}	1^{c}	20^{b}
Sapele kedwa	68	191	924^{b}	5^{b}	73^{a}
Nasaka	43	186	1346^{b}	7^{a}	82^{a}
BCB3	64	196	1196^{b}	6^{ab}	76^{a}
Bwenzilaana	80	220	1018^{b}	5^{b}	73^{a}
Kalima	26	189	1203^{b}	6^{ab}	88^{a}
BCB1	56	351	2397^{a}	7^{a}	86^{a}
Grand mean	119	208	1196	5	71
F(SIG)	.001	.001	.001	.001	.001
LSD	59	60	685	1.5	21
CV (%)	34	20	39	19.8	20

Note: Means annotated by the same letter (s) are significantly the same

4.4 Seed Weight Loss

Seeds weight loss ranged from 3% to 29 % (Table 6). BCB1 (29 %) and 26 % Kalima suffered the greatest weight loss.

Overall, only BCB2 was resistant to the bean weevils, several factors may account for the observed resistance in this variety. First, physical characteristics such as grain size and weight have been reported confer resistance in certain crops. According to Bean/Cowpea Collaborative Research Support Programme (CRSP), BCB2 is regarded as a small seeded variety as it has a seed weight of less than 25g. Small seeded varieties are more resistant as compared to medium seeded and large seeded varieties [9]. The small seeded varieties have seed coat which is rough and hard, this increase the resistance to pest [10]. Hence, few insects are able to penetrate the seed coat. This is evidenced by few reported holes per grain as large seeded varieties have been reported to be more prone to insect burrowing than in small seeded varieties [11].

White colored Chickpeas (*Cicer arietinum*) were found to be vulnerable to storage pest [8]. In Nigeria, black coloured Bambara (*Vigna subterranean*) nuts supported more bruchids than other seed coat colours [12]. This however, whether this was at work in the present experiment was could not be ascertained for the bean varieties used in this experiment.

Insect developmental period is longer in resistant varieties [11,13]. These results are in agreement with previous findings in Malawi [7]. Varieties with short development period of bruchids have implications to growers or anyone associated with beans, they shorten the storage period by increases post-harvest losses. As such they require immediate treatment with an effective insecticide. In the absence of any control strategy*, A. obtectus* and *Z. subfasciatus* are capable of causing up to 100% damage on stored bean seeds [14]. This increases cost of production.

Biochemical characteristics play a great role in host plant resistance. The crop may exude chemicals which repel the insects. Some secondary plant metabolites deter insects from feeding, mating and/or oviposition, Studies by Ishimoto et al. [15] found that transgenic Azuki beans (*Vigna angularis*) expressing αAI gene completely blocked bruchid development [16]. A protein called arcelin in the bean cotyledons was reported to be the source of resistance responsible for the resistance of wild common beans in Mexico [17]. On the contrary, an arcelin-5 variant *P. vulgaris* accession G02771A was found not to be the source of resistance [18]. However, for beans mechanisms of bruchid resistance in the field may be different from that in the laboratory [7]. Detailed studies on biochemical and physical characteristics of the seeds underlining the resistance need to be carried out.

Table 6. Response of different bean varieties to bean weevil infestation

Variety	Weight of undamaged	Weight of damaged	Initial weight of seeds	Final weight of seeds	Actual weight loss %
BCB2	84[b]	13	100	97	3[c]
Sapelekedwa	27[a]	56	100	84	16[ab]
Nasaka	19[a]	62	100	81	19[ab]
BCB3	24[a]	54	100	77	23[ab]
Bwenzilaana	25[a]	50	100	75	25[ab]
Kalima	24[a]	50	100	74	26[ab]
BCB1	18[a]	53	100	71	29[a]
Grand mean	30	50	100	80	20
F(SIG)	.001	.001		.01	.01
LSD	20	16		13	13
CV (%)	47	22		11	45

Note: Means annotated by the same letter (s) are significantly the same

4. CONCLUSION

The level of resistance among the seven varieties evaluated to two common bean weevils (*Acanthoscelides obtectus and Zabrotes subfasciatus*) differed significantly during the four and half month's storage period. Only BCB2 was resistant to the two weevils. Sapele kedwa was also the only moderate resistant, Nasaka, BCB3 and Bwenzilaana were susceptible and Kalima and BCB1 were highly susceptible.

CONSENT

Both authors declare that this was a student work and as registered members of Bunda College had access to the laboratory research of different selected bean varieties for publication of this study report.

ETHICAL APPROVAL

This project did not involve the use of human or animal subjects.

ACKNOWLEDGEMENTS

The main and corresponding author would like to thank the Bunda College of Agriculture for providing the financial support on this study. A vote of thanks goes to the following; Nyokase Kanyoza, Stewart Chauluka, Harry Manyamba, Calvin Msauka, Lumbani Banda, Esther Ngwira, Peter Mpanang'ombe, Kareem Longwe, Walusako Kayinga and Precious Mtalimanja

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Abate T, Ampofo J. Insect pests of beans in Africa: Their ecology and management. Annual Review of Entomology. 1996;41:45-73.
2. Dent D. Insect Pest Management. 2nd edition. UK: CABI; 2000.
3. Denning G, Kabambe P, Sanchez P, Malik A, Flor R. Input subsidies to improve smallholder maize productivity in Malawi: toward an African green revolution. PLoS Biol. 2009;7(1):118-123.
4. MoAFS. Guide to agricultural production and natural resources management in Malawi. Malawi Government: Lilongwe: Malawi; 2004.
5. Jones A, Phaseolus bean: Post-harvest operations. (D. Mejia, and Lewis, Eds.) CIAT: Cal: Colombia; 1999.
6. Mlangiza C. Evaluation of pigeon pea varieties for resistance to pigeon pea bruchid (weevil) *Callobruchus maculatus.* Bachelors project report, University of Malawi: Lilongwe; 2006. (in press).
7. Kananji G. A Study of Bruchid Resistance and Its Inheritance in Malawian Dry Bean Germplasm: PhD thesis in plant breeding. University of KwaZulu-Natal: South Africa; 2007.
8. Sarwar M. Assessment of resistance to the attack of bean beetle Callosobruchus maculatus (Fabricius) in chickpea genotypes on the basis of various parameters during storage. Songklanakarin J. Sci. Technol. 2012;3(34):287-291.
9. Mei L, Cheng X, Wang L, Liu C, Sun L, Xu N, Liu C. Relationship between bruchids resistance and seed mass in mangbean

based on QTL analysis. NRC. 2009;52(8):589-591.
10. Edge O. Recommendation for Bean production. Bunda College Of Agriculture: Lilongwe; 1981.
11. Bell A, Muck O, Mutlu P, Schneider H. Integrated Post-Harvest Protection.Is worth its money! The cost-effectiveness of systems-oriented measures in post-harvest protection control of the Largere Borer Prostephanus trunctus. GTZ publication; 1999.
12. Dyton, M. Field infestation of different bean varieties at different time by storage insect pests at Bunda College. Unpublished Bachelors project report, University of Malawi. Lilongwe; 1998.
13. Echezona BC, Amuji CF, Eze S. Evaluation of some accessions of bambara groundnut (*Vigna subterranean* l. Verdc) for resistance to bruchid infestation based on grain source and seed coat colour. Journal of Plant Protection Research. 2013;53(3):210-214.
14. Ahmed, B., and Yusuf, A. Host plant resistant: A variable non –chemical and environmentally friendly strategy of controlling stored products-A review. Emir. J. Food Agric. 2007;19(1):01-12.
15. Porca M. The influence of chemical composition of different origin beans (Phaseolus vulgaris L.) on tolerance to the bean weevil (*Acanthoscelides obtectus* Say) stroke. Journal of Central European Agriculture. 2003;4:1-10.
16. Ishimoto M, Sato T, Chrispeels MJ, Kitamura K. Bruchid resistance of transgenic azuki bean expressing seed α-amylase inhibitor of common bean. Entomologia Experimentalis et Applicata. 1996;79:309–315.
17. Osborn TC, Alexander DC, Sun SSM, Cardona C, Bliss FA. Insecticidal activity and lectin homology of arcelin seed protein. Science. 1988;240:207-210.
18. Goossens A, Quintero C, Dillen W, De Rycke R, Valor JF, De Clercq K, Van Montagu M, Cardona C. Analysis of bruchid resistance in the wild common bean accession G02771: no evidence for insecticidal activity of arcelin 5. Journal of Experimental Botany. 2000;51(348):1229-1236.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history.php?iid=680&id=24&aid=6288