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VARIETAL RESISTANCE OF SOYBEAN TO *Callosobruchus maculatus* AND PROTECTION OF THE SOYBEAN SEEDS USING *Gnidia kraussiana* ROOT EXTRACTS

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ABSTRACT. Soybean (*Glycine max* L.) is a key source of plant protein and oil in sub-Saharan Africa, but its storage is affected by the cowpea seed beetle, *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae). The present study investigated the *C. maculatus* resistance of 10 soybean varieties in 2 agroecological zones of northern Cameroon and evaluated the protective efficacy of *Gnidia kraussiana* root extracts compared to neem seed oil on a susceptible variety. Significant varietal differences in *C. maculatus* susceptibility were observed, with resistance varying across locations. Only a subset of varieties (e.g., TGx1844-18E, SJ235, VT/SG Houla1, and Emgopa314) exhibited consistent resistance across both sites, and others showed location-dependent responses. Kodek/Houla1 displayed variable susceptibility and was classified as moderately susceptible in Maroua and moderately resistant in Ngaoundéré. Resistance was associated with seed biochemical traits. The soluble sugar content was positively correlated with susceptibility, whereas saponins were negatively correlated with susceptibility. Bioassays conducted on Kodek/Houla1 revealed that the hexane and acetone fractions of *G. kraussiana* root extracts caused high adult mortality, strongly reduced progeny emergence and seed damage, and prolonged protection during

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storage. At 5 g·kg^{-1}, both fractions almost completely inhibited beetle development and showed an efficacy comparable to neem seed oil. These findings suggest that integrating soybean varieties with stable resistance profiles and plant-based protectants, such as *G. kraussiana* extracts, could contribute to the environmentally sustainable management of *C. maculatus*. However, further studies including chemical characterisation of the active compounds are required.

Keywords: host biochemical defence; integrated pest management; plant-derived insecticides; postharvest protection; resistance screening.

INTRODUCTION

Soybean (*Glycine max* (L.) Merr.) is a major legume crop in sub-Saharan Africa due to its high protein (36–40%), oil (20%), and carbohydrate contents (Saha and Mandal, 2019). In Cameroon, 21,400 tonnes of soybean were produced in 2021 (MINEPAT, 2023), contributing to food security, livestock feed, and agro-industrial uses. However, postharvest losses during storage significantly reduce productivity and farmers' income.

The cowpea seed beetle, *Callosobruchus maculatus* (Fabricius) (Coleoptera: Chrysomelidae: Bruchinae), is one of the most destructive pests of stored legumes worldwide (Nwosu *et al*., 2020; Kalpna *et al*., 2022). Females lay eggs on the seed surface, and after hatching, the larvae penetrate the seed, where they complete development (Ashamo *et al*., 2021; Kalpna *et al*., 2022). This internal feeding habit reduces the seed weight and germination capacity and causes nutritional depletion and contamination, reducing its market and planting value (Ashamo *et al*., 2021; Asra *et al*., 2025).

Under favourable conditions, infestations can lead to severe quantitative and qualitative losses during prolonged storage, sometimes exceeding 50–60% in susceptible legume species (Tripathi *et al*., 2020). Although soybean is generally less susceptible than cowpea, significant damage still occurs, particularly in varieties with favourable nutritional profiles for larval development (Pessu and Umeozor, 2004; Pawlowski *et al*., 2021). In sub-Saharan Africa, which has a limited storage infrastructure, *C. maculatus* represents a major constraint to the expansion and profitability of soybean production systems (Loko *et al*., 2022).

Reliance on synthetic insecticides for control is widespread but problematic due to the cost, limited access in rural areas, and environmental and health concerns (Njume *et al*., 2019). Thus, safer and locally adaptable alternatives are needed. Integrated pest management offers viable solutions, particularly when it integrates host plant resistance with botanical insecticides. Resistance traits, such as seed hardness, coat thickness, tannins, and phenolics, can limit bruchid development (Allotey and Oyewo, 2005).

Gnidia kraussiana Meisn. belongs to the Thymelaeaceae family, which is known for the richness of its biologically active secondary metabolites, including terpenoids, flavonoids, and coumarins. Phytochemical analyses of *G. kraussiana* extracts have confirmed the presence of these compounds, which have been associated with insecticidal and biological activities (Kosini and Nukenine, 2017). In northern Cameroon, *G. kraussiana* is traditionally used by local farmers as a natural protectant of stored products, particularly in powdered form and applied to grains. Previous studies have demonstrated the insecticidal and repellent activities of *G. kraussiana* against storage pests, particularly bruchids, supporting its potential as a locally available and environmentally sustainable alternative to synthetic insecticides (Kosini *et al*., 2021). The present study advances existing knowledge by linking varietal resistance screening with targeted botanical protection in soybean, a system that remains poorly documented. This study evaluated the efficacy of *G. kraussiana* extracts on a highly susceptible soybean variety under realistic storage conditions and simultaneously examined the biochemical determinants of resistance across genotypes. This combined approach provides new insights into how host plant traits and botanical treatments can be strategically integrated for postharvest pest management.

Therefore, this study combined varietal resistance screening with the evaluation of *G. kraussiana* extracts to: i) assess resistance levels in 10 soybean varieties across 2 agroecological zones; ii) determine the role of seed physical and biochemical traits in varietal susceptibility; and iii) evaluate the insecticidal performance of *G. kraussiana* extracts compared to neem seed oil on the most susceptible variety. The aim was to propose practical, farmer-adapted strategies for reducing bruchid-induced postharvest losses in soybean.

MATERIALS AND METHODS

Experimental localities

This study was conducted in the Laboratory of Applied Zoology, Department of Biological Sciences, University of Ngaoundéré (Adamawa Region, Cameroon), and the Laboratory of Entomology in Maroua (Far North Region, Cameroon). Maroua (10.44° N, 14.25° E; 422 m above sea level) is located in the Sudano–Sahelian (SS) zone of Cameroon. The area is characterised by unimodal rainfall with a rainy season lasting 4 months (June–September) and transitional months of unreliable rainfall in May and October. The mean annual rainfall is approximately 834 mm, and the average annual temperature is 34.5°C (Tingem *et al*., 2008). Ngaoundéré (7.34° N, 13.57° E; 1104 m above sea level) lies in the Sudano–Guinean (SG) zone. Rainfall is also unimodal with a rainy season lasting 6 months (May–October). The annual average rainfall is 1514 mm, and the mean annual temperature is 29.0°C (Tingem *et al*., 2008).

Experimental conditions

Experiments were conducted under ambient laboratory conditions. In the Laboratory of Entomology (Maroua, SS zone), the mean temperature was 27.83 ± 2.06°C (22.00–32.00°C), and the mean relative humidity was 80.65 ± 6.13% (61.00–94.50%), under a 12:12 h light:dark photoperiod. In the Laboratory of Applied Zoology (Ngaoundéré, SG zone), the mean temperature was 22.57 ± 1.43°C (19.50–32.00°C), and the mean relative humidity was 82.01 ± 3.03% (55.50–88.50%) under a 12:12 h light:dark photoperiod.

Experiments were conducted under ambient laboratory conditions rather than in a controlled climate chamber to better simulate the real storage environments commonly encountered by smallholder farmers in northern Cameroon. This approach enhances the practical relevance and applicability of the results under field conditions, as temperature and relative humidity are not strictly regulated.

Insect rearing

Callosobruchus maculatus adults were collected from the Mokolo market (Mayo Tsanaga Division, Far North Cameroon). A total of 100 parent insects obtained from untreated, infested cowpea stocks were introduced into 500 g of sterilised cowpea seeds and maintained under ambient laboratory conditions following the method described by Kosini *et al*. (2019). The colony was reared for two successive generations, and newly emerged adults from the F_2 generation were used for all experiments. The use of F_2 individuals was intended to minimise potential field-related variability (e.g., prior insecticide exposure, host plant history, and environmental stress) and ensure a more homogeneous and laboratory-acclimated population with stable physiological and reproductive characteristics. This approach improves the experimental reproducibility and reduces bias associated with heterogeneous field-collected insects.

Plant materials and fractionated extracts

Gnidia kraussiana roots were harvested around Mogode in the Mayo-Tsanaga Division, Far-North region of Cameroon (latitude 10°36.25”N and longitude 13°34.46”E, altitude 1005 m above sea level [m.a.s.l]). The identity of the plant was confirmed at the Cameroon National Herbarium in Yaounde, where voucher specimens (Serial number: 38259/HNC) were kept. Harvested materials were washed, shade-dried, and ground until the powder passed through a 0.4-mm mesh sieve. Fractionated extracts were prepared by mixing 2300 g of plant powder with 7.5 L of hexane, stirring for 30 min, and leaving the mixture to stand for 24 h. The mixture was stirred again, and after 48 h, it was filtered through Whatman No. 1 filter paper. The residue was re-extracted using the same procedure, and the filtrates were combined. The remaining plant material was air-dried at room temperature for 10 h and sequentially extracted with acetone and methanol using the same procedure. Filtrates were concentrated separately using a rotary evaporator at 70°C (hexane), 60°C (acetone), and 65°C (methanol) at 120 rpm. Extracts were kept in amber bottles at 4°C and re-diluted to the required concentrations before use. Extract yields (w/w) were 9.729 g (0.423%), 17.412 g (0.757%), and 130.452 g (5.672%) for the hexane, acetone, and methanol fractions, respectively. Neem seed oil (*Azadirachta indica*) was extracted using the traditional kneading method as described by Nukenine *et al*. (2011). Neem kernels were manually pounded in a mortar to obtain a fine powder. Approximately 2.6 kg of the powder was moistened with 0.25 L of warm water in a clean plastic container and mixed thoroughly until a dough-like consistency was achieved. The resulting paste was kneaded and firmly pressed by hand in successive cycles to release the oil. Kneading and pressing were alternated for several minutes to extract the maximum amount of oil from the neem cake. The crude oil was decanted and stored in sealed bottles at 4°C until use in the bioassays. Neem seed oil was included as a positive control due to its well-documented insecticidal properties against storage pests, particularly *C. maculatus*, and its widespread use in smallholder storage systems as a reference botanical insecticide (Nukenine *et al*., 2011). Its inclusion enabled the comparison of the efficacy of tested plant extracts with a commonly used and scientifically validated botanical standard.

Soybean varieties

Ten soybean (*Glycine max*) varieties were supplied by the Institute of Agricultural Research for Development (IRAD), Garoua, Cameroon: TGx 713-09, SJ 235, VT/SG Houla1, TGx 1844-18E, TGx 1910-14F, TGx 849-294D, Emgopa314, Goiania, SK/Goiania, and Kodek/Houla1. Seeds were cleaned, disinfested at −20°C for 7 days, and equilibrated to room temperature before use.

Varietal resistance screening

For each variety, 50 g of seed was placed in a 500-mL glass jar. Ten pairs (♀/♂) of newly emerged *C. maculatus* were introduced and allowed to oviposit for six days, after which adults were removed. The emergence of F_1 adults was monitored from 40 days post-infestation until no emergence occurred for 5 consecutive days. Experiments were arranged in a completely randomised design with 5 replicates per variety (n = 5) and each replicate consisting of one experimental unit (50 g of seeds per jar). The recorded parameters included total F_1 emergence, median developmental period (MDP, days), and Dobie susceptibility index (SI), calculated as follows: $SI = (\ln(F_1)/MDP) \times 100$.

Varieties were classified following Dobie's scale (Dobie, 1974) as follows: resistant (SI ≤ 4), moderately resistant (4.1–6.0), moderately susceptible (6.1–8.0), susceptible (8.1–10), and highly susceptible (SI > 10).

Physical seed characteristics

The length, width, and thickness of 50 seeds per variety were measured using a Vernier calliper (± 0.05 mm). The derived physical parameters included the geometric mean diameter [$Dg = (L \times W \times T)^{1/3}$ (mm)], surface area [$S = \pi Dg^2$ (mm²)], sphericity ($Sp = Dg/L$), volume {$V = 0.25[\pi/6\ (L(W+T)^2)]$}, and mass of 100 seeds (Mohsenin, 1986; Jain and Bal, 1997; Unal *et al*., 2006; Mohammad *et al*., 2010; Noubissié *et al*., 2011).

Biochemical analyses

Proximate and secondary metabolite analyses were performed on six soybean genotypes selected from the available varieties based on their contrasting susceptibilities to *C. maculatus*, representing a susceptibility gradient and facilitating the identification of potential relationships between biochemical traits and susceptibility. Air-dried seeds (100 g per variety) were ground to a fine powder (150 µm) using a Wiley Mill (Scientific Equipment Works, Delhi, India) and stored in polyethylene bags at 4°C until analysis. The proximate composition was determined as follows: moisture and crude ash content according to AFNOR (1981, 1982); total carbohydrates and soluble sugars following Dubois *et al.* (1956); total proteins via the Kjeldahl method (AFNOR,1984; Devani *et al*., 1989); lipids according to UICPA (1979); and crude fibre using the Weende method (Wolff, 1968).

Secondary metabolites and allelochemicals were assessed as follows: total polyphenolics using Folin–

Ciocalteu reagent, i.e. gallic acid equivalents (Makkar *et al*., 1993); saponins following Harborne (1973); and phytates, tannins, flavonoids, alkaloids, steroids, and triterpenoids following Adeniyi *et al*. (2010). Quantitative data from proximate and secondary metabolite analyses were used to identify correlations with SI.

Bioassays on the susceptible variety

The most susceptible variety (Kodek/Houla1) was treated with *G. kraussiana* extracts. Application volumes of 0.2 and 1.0 mL of extract at a concentration of 0.25 $g{\cdot}mL^{-1}$ were applied to 50 g of seeds, corresponding to doses of 1 and 5 $g{\cdot}kg^{-1}$, respectively. These doses were calculated such that 0.2 mL of extract delivered 0.05 g of active material to 50 g of seeds (equivalent to 1 $g{\cdot}kg^{-1}$), and 1.0 mL delivered 0.25 g (equivalent to 5 $g{\cdot}kg^{-1}$). To standardise the total volume of liquid applied across treatments, an additional 0.8 mL of the corresponding solvent was added to the 0.2 mL treatment, ensuring that all experimental units received 1.0 mL of liquid. Control treatments included non-treated seeds, solvent-only seeds, and neem seed oil applied at equivalent doses. After application, the seeds were gently shaken to ensure uniform coating and air-dried for 2 h to allow complete solvent evaporation prior to insect introduction. All treatments were arranged in a completely randomised design with 4 replicates per treatment (n = 4) and each replicate consisting of one jar containing 50 g of treated seeds.

Adult toxicity

Each jar (50 g treated seed) was infested with 20 mixed-sex adults (1–2 days old). Mortality was recorded as the cumulative percentage per replicate (20 insects per unit); therefore, individual time-to-death data were not available, precluding the use of survival analysis. Mortality was corrected using the formula given by Abbott (1925).

Progeny suppression

After adult mortality assessment, jars were monitored for F_1 emergence until emergence ceased (5 consecutive days without emergence). The total F_1 emergence was recorded, and the percent of reproduction suppression compared to the controls was calculated.

Population increase, seed damage, and weight loss assay

The same samples used for first progeny production were used to evaluate the population increase and seed damage. After the first progeny were recorded, all insects (live and dead) as well as the seeds from each jar were kept on laboratory benches for four months, starting from the date of infestation. At the end of this period, the number of live and dead insects was recorded. The numbers of undamaged and damaged grains were separately counted and weighed from each jar. The final moisture content of seeds was also determined to correct the seed weight to the moisture.

The weight of additional moisture (WAM) was calculated as follows: $WAM = Wi \times (Mf - Mi) / (100 - Mf)$, where Wi is the initial seed weight (g), Mi is the initial seed moisture content (%), and Mf is the final seed moisture content (%).

The percent weight loss (PWL), corrected for both moisture and added insecticidal material, was calculated as $PWL = ((Wi - (Wf + WAM + Maad)) / Wi) \times 100$, where Wf is the final seed weight (g) measured after removing all insects, and Madd is the weight of any residual insecticidal material (g).

The percent damaged seeds (PDS) was expressed as PDS = (Nd / (Nd + Nu)) × 100, where Nd is the number of damaged seeds and Nu is the number of undamaged seeds.

Repellence assay

A linear olfactometer (30 cm length, 2 cm diameter) with a central hole was used following the description given by Kosini and Nukenine (2017). At each end, 10 g of treated or solvent-control seeds were placed. Twenty insects (≤ 48 h old) were introduced through the central hole and given 2 h to make a choice. Each treatment (1 and 5 g kg^{-1}) was replicated 5 times. The percentage of repellence (PR) was calculated using the formula given by Talukder and Howse (1994): PR = 2 × (C − 50), where C is the percentage of insects on the control side. The results were categorised using the Jilani and Su (1983) repellence scale (Classes 0–V).

Data analysis

SI data were analysed using a generalised linear model (GLM) with variety, site, and their interaction as fixed factors. A normal distribution with identity link function was specified. Mortality, progeny inhibition, and seed damage data were also analysed using GLMs with appropriate error distributions (binomial for proportion data and Poisson or Gaussian, where applicable) and corresponding link functions. Grain physicochemical traits used to compare genotype variability and weight loss were analysed using analysis of variance (ANOVA), and means were separated using Tukey's honestly significant difference (HSD) test at α = 0.05. Before ANOVA, data were checked for normality using the Shapiro–Wilk test and for homogeneity of variances using Levene's test. Relationships between biochemical traits and susceptibility were determined using correlation analysis within the Sudano–Guinean dataset only to avoid the confounding effects of environmental variation Spearman's rank correlation coefficient (ρ) was used due to the limited sample size (n = 6) and potential non-normal data distribution. This non-parametric approach is more appropriate for small sample sizes and does not assume linearity or normality of the variables. All statistical analyses were performed using IBM SPSS Statistics (IBM Corp., Armonk, NY, USA). The results are presented as the mean ± standard error (SE).

RESULTS

Varietal resistance of soybean to *Callosobruchus maculatus*

F_1 adult emergence, MDP, and SI varied among soybean varieties and between the two experimental sites (*Table 1a*). These descriptive results highlight substantial variability in the response of soybean genotypes to infestation by *C. maculatus*. F_1 emergence differed across varieties and sites. In Maroua, emergence ranged from 1 individual in TGx713-09 to 26 in Kodek/Houla1, whereas in Ngaoundéré, it ranged from 1 individual in TGx1844-18E to 41 in Kodek/Houla1 (*Table 1a*). Similarly, the MDP tended to be longer in Ngaoundéré than in Maroua, suggesting that environmental conditions influenced insect development.

The patterns observed in the descriptive data were supported by the

GLM (*Table 1b*), which showed a significant effect of site on SI. The positive parameter estimate for Maroua (B = 1.921, $p < 0.001$) indicates that susceptibility was significantly higher in Maroua than in Ngaoundéré, confirming the influence of environmental conditions.

Based on the SI presented in *Table 1a*, varietal responses to *C. maculatus* varied among genotypes and between locations. TGx1844-18E, SJ235, VT/SG Houla1, and Emgopa314 were consistently resistant across both sites. Several varieties showed location-dependent responses. Goiania, TGx1910-14F, and TGx849-294D were moderately resistant in Maroua but resistant in Ngaoundéré, whereas TGx713-09 exhibited the opposite pattern, showing resistance in Maroua and moderate resistance in Ngaoundéré. SK/Goiania remained moderately resistant at both locations. In contrast, Kodek/Houla1 showed variable susceptibility, ranging from moderately susceptible in Maroua to moderately resistant in Ngaoundéré.

These differences were further supported by the GLM (*Table 1b*), which demonstrated that SI was significantly affected by both varietal and biological factors. Several varieties, including TGx1844-18E ($p = 0.004$), SJ235 ($p = 0.029$), Kodek/Houla1 ($p < 0.001$), SK/Goiania ($p = 0.035$), VT/SG Houla1 ($p < 0.001$), and TGx713-09 ($p < 0.001$), showed a significantly lower susceptibility compared to the reference variety (TGx849-294D). Among these, TGx713-09 exhibited the strongest reduction in susceptibility, which is consistent with its very low progeny emergence, as reported in *Table 1a*.

Biological covariates also played a significant role. The number of progeny was positively and strongly associated with SI (B = 0.146, $p < 0.001$), indicating that increased reproductive success led to higher susceptibility. In contrast, MDP was not significantly associated with SI ($p = 0.119$), suggesting that variation in the development time alone did not explain susceptibility differences.

Overall, *Table 1a* and *Table 1b* suggest that susceptibility to *C. maculatus* is shaped by both genotype and environmental conditions, with reproductive performance acting as a key driver of variation among soybean varieties.

Physical and biochemical determinants of susceptibility

Physical properties of soybean varieties

Significant differences ($p < 0.001$) were observed among soybean varieties for all measured physical parameters, including 100-grain mass, geometric diameter, degree of sphericity, volume, and geometric surface area (*Table 2*). The grain mass varied among varieties, ranging from 11.4 g in SJ235 to 15.0 g in Emgopa314.

Similarly, the geometric diameter ranged from 5.7 (SJ235) to 6.4 mm (TGx713-09). TGx713-09 exhibited the highest values for both volume (141.4 mm^3) and geometric surface area (113.8 mm^2), whereas SJ235 and TGx849-294D showed comparatively lower values for these parameters. Despite this significant variability in physical traits, no clear patterns linking seed size or shape to susceptibility levels were observed. Varieties with larger seeds or greater surface areas were not consistently more or less susceptible to infestation.

Table 1a – Mean (± S.E.) adult emergence, developmental period, and susceptibility index of *Callosobruchus maculatus* in soybean grains at Maroua and Ngaoundéré

Variety	Mean number of emerged adult		Mean developmental period (Days)		Index of susceptibility	
	Sites		Sites		Sites	
	Maroua	Ngaoundere	Maroua	Ngaoundere	Maroua	Ngaoundere
Emgopa314	8 ± 2	5 ± 0	51.0 ± 1.4	75.6 ± 1.8	4.0 ± 0.3^{RR}	2.0 ± 0.1^{RR}
Goiania	10 ± 1	20 ± 1	48.0 ± 0.7	77.0 ± 0.6	4.7 ± 0.2^{MR}	3.9 ± 0.1^{RR}
Kodek/Houla1	26 ± 2	41 ± 2	48.2 ± 0.4	66.8 ± 0.6	6.8 ± 0.1^{MS}	5.6 ± 0.1^{MR}
SJ235	5 ± 0	19 ± 1	51.6 ± 0.8	76.2 ± 1.0	3.2 ± 0.1^{RR}	3.9 ± 0.0^{RR}
SK/Goiania	21 ± 1	29 ± 2	48.6 ± 0.7	73.6 ± 0.5	6.3 ± 0.2^{MR}	4.6 ± 0.1^{MR}
TGx1844-18^{E}	4 ± 0	1 ± 0	49.2 ± 0.2	41.0 ± 0.0	2.8 ± 0.2^{RR}	0.6 ± 0.3^{RR}
TGx1910-14^{F}	7 ± 1	8 ± 1	46.4 ± 1.0	75.6 ± 0.8	4.1 ± 0.2^{MR}	2.7 ± 0.1^{RR}
TGx713-09	1 ± 0	36 ± 1	45.6 ± 1.3	72.4 ± 0.5	0.4 ± 0.4^{RR}	5.0 ± 0.1^{MR}
TGx849-294^{D}	23 ± 3	12 ± 1	49.2 ± 0.7	73.6 ± 0.3	6.3 ± 0.2^{MR}	3.4 ± 0.1^{RR}
VT/SG Houla1	3 ± 0	20 ± 2	53.2 ± 0.9	76.4 ± 0.6	2.0 ± 0.3^{RR}	3.9 ± 0.1^{RR}
Mean	11 ± 1	19 ± 2	49.1 ± 0.4	70.8 ± 2.1	4.1 ± 0.3^{MR}	3.6 ± 0.2^{RR}

RR = resistant, MR = moderately resistant, MS = moderately susceptible

Table 1b – Parameter estimates from a generalised linear model (GLM) assessing the effects of site, soybean variety, and biological parameters on the susceptibility index of soybean to *Callosobruchus maculatus*

Predictor	B (Estimate)	Std. Error	Wald Chi-Square	df	p-value
(Intercept)	0.752	0.46	2.628	1	0.105
Maroua	1.921	0.1707	126.769	1	0.000
Ngaoundéré	0[a]	-	-	-	-
TGx1844-18^{E}	-0.804	0.2781	8.356	1	0.004
Emgopa314	-0.221	0.2571	0.741	1	0.389
SJ235	-0.533	0.2447	4.749	1	0.029
Kodek/Houla1	-1.000	0.2751	13.222	1	0.000
SK/Goiania	-0.520	0.2469	4.429	1	0.035
VT/SG Houla1	-1.059	0.2461	18.518	1	0.000
Goiania	-0176	0.2409	0.531	1	0.466
TGx1910-14^{F}	0.050	0.2536	0.039	1	0.844
TGX713-09	-2.292	0.2404	90.963	1	0.000
TGx849-294^{D}	0[a]	-	-	-	-
Number of progeny	0.146	0.0081	328.598	1	0.000
Median Developmental Period (MDP)	0.009	0.0059	2.435	1	0.119

a: Reference category. Ngaoundéré and TGx849-294^{D} were used as reference levels for sites and varieties, respectively

Table 2 – Physical properties (mean ± S.E.) of soybean varieties

Variety	Parameters				
	100 GM (g)	GD (mm)	D.Sph.	Vol. (mm^3)	GS (mm^2)
Emgopa314	15.0 ± 0.1^{a}	6.3 ± 0.0^{ab}	0.9 ± 0.0^{a}	135.7 ± 2.6^{ab}	113.5 ± 1.5^{ab}
Goiania	12.0 ± 0.0^{e}	6.0 ± 0.0^{def}	0.8 ± 0.0^{b}	111.9 ± 1.6^{def}	98.5 ± 0.9^{c}
Kodek/Houla1	11.9 ± 0.1^{ef}	6.0 ± 0.1^{d}	0.8 ± 0.0^{bc}	117.4 ± 2.7^{d}	100.5 ± 1.5^{c}
SJ235	11.4 ± 0.1^{f}	5.7 ± 0.0^{g}	0.8 ± 0.0^{bc}	100.3 ± 2.2^{g}	90.7 ± 1.4^{c}
SK/Goiania	12.0 ± 0.1^{de}	6.0 ± 0.0^{de}	0.8 ± 0.0^{bc}	115.9 ± 2.1^{de}	99.9 ± 1.2^{c}
TGx1844-18E	12.7 ± 0.1^{cd}	6.1 ± 0.0^{cd}	0.8 ± 0.0^{c}	119.6 ± 2.5^{cd}	101.2 ± 1.4^{c}
TGx1910-14F	11.8 ± 0.2^{ef}	6.2 ± 0.0^{bc}	0.8 ± 0.0^{bc}	129.4 ± 2.6^{bc}	107.4 ± 1.5^{b}
TGx713-09	13.6 ± 0.2^{b}	6.4 ± 0.0^{a}	0.8 ± 0.0^{bc}	141.4 ± 2.7^{a}	113.8 ± 1.4^{a}
TGx849-294D	14.5 ± 0.1^{a}	5.8 ± 0.0^{fg}	0.8 ± 0.0^{b}	103.7 ± 2.7^{fg}	93.5 ± 1.4^{de}
VT/SG Houla1	12.9 ± 0.2^{c}	5.8 ± 0.0^{efg}	0.8 ± 0.0^{b}	106.4 ± 2.2^{efg}	95.1 ± 1.5^{cde}
F -value	92.89^{***}	31.95^{***}	13.32^{***}	33.62^{***}	33.25^{***}

***: $p < 0.001$; Means within the same column followed by the same letter do not differ significantly ($p < 0.05$; Tukey's test). Com: commodity, GM: grain mass, GD: geometric diameter, D.Sph: degree of sphericity, Vol.: volume, GS: geometric surface

Biochemical composition of soybean varieties

The biochemical composition of soybean varieties differed significantly ($p < 0.001$) for most parameters, except moisture content, which remained similar across genotypes (7.6–8.9 g/100 g) (*Table 3*). The ash content was highest in TGx718-09 (6.0 g/100 g) and lowest in Emgopa314 (3.6 g/100 g). Total sugars ranged from 23.7 g/100 g in TGx718-09 to 33.4 g/100 g in TGx1844-18E. The soluble sugar content varied, being highest in susceptible variety Kodek/Houla1 (6.3 g/100 g), intermediate in Goiania (5.7 g/100 g), and very low (≈1 g/100 g) in resistant genotypes, such as TGx718-09, TGx1844-18E, and VT/SG Houla1.

The crude fibre content was greatest in VT/SG Houla1 (22.4 g/100 g), followed by TGx1844-18E (19.5 g/100 g), and the lowest value was found in TGx718-09 (14.3 g/100 g). The fat content also varied significantly, with Emgopa314 having the highest content (20.1 g/100 g) and VT/SG Houla1 (17.4 g/100 g) and TGx1844-18E (18.1 g/100 g) having the lowest. Protein levels were narrowly distributed but significantly different, ranging from 33.6 g/100 g (VT/SG Houla1) to 35.8 g/100 g (TGx718-09). Secondary metabolites also varied between varieties. Saponins were most abundant in VT/SG Houla1 (88.2 mg/g) and Emgopa314 (61.5 mg/g), and Kodek/Houla1, TGx718-09, and TGx1844-18E had lower levels (36–46 mg/g). The total polyphenols were abundant in VT/SG Houla1 (15.7 mg/g) and Goiania (14.6 mg/g) but very low in Kodek/Houla1 and Emgopa314 (1.6–1.7 mg/g). Qualitative screening confirmed that tannins, flavonoids, alkaloids, steroids, and terpenoids were generally present, often in greater abundance in resistant genotypes than in susceptible ones.

Overall, susceptible variety Kodek/Houla1 was distinguished by its high soluble sugar content and low polyphenol content, whereas resistant varieties tended to combine a low soluble sugar content with higher contents of crude fibre, saponins, and polyphenolic compounds.

Correlation between biochemical constituents and susceptibility

Correlation analysis revealed significant associations between several biochemical traits and SI (*Table 4*). SI was positively correlated with the moisture content ($\rho = 0.57$, $p < 0.01$), total carbohydrate content ($\rho = 0.64$, $p < 0.01$), soluble sugar content ($\rho = 0.52$, $p < 0.05$), and protein content ($\rho = 0.57$, $p < 0.01$). These positive relationships indicate that higher nutrient availability enhances insect development and increases susceptibility. In contrast, saponins showed a significant negative correlation with susceptibility ($\rho = -0.49$, $p < 0.05$), suggesting the protective role of these compounds against *C. maculatus*. The total fibre also exhibited a negative but non-significant association with susceptibility. Other parameters, including the ash, fat, and total polyphenol contents, did not show significant correlations with SI. Taken together, these results suggest that susceptibility is positively associated with nutritional components that favour insect development and that certain secondary metabolites, particularly saponins, contribute to resistance.

Table 3 – Chemical composition (mean ± S.E.) of resistant (TGx718-09, VT/SG Houla1, TGx1844-18E, and Emgopa314) and moderately resistant (Goiania and Kodek/Houla1) soybean varieties to *Callosobruchus maculatus*

MA	**Variety**						
	Kodek/ Houla1	**Emgopa 314**	**Goiania**	**TGx 718-09**	**TGx 1844-18^{E}**	**VT/SG Houla1**	***F*-value**
MC^{1}	8.8 ± 0.9^{A}	8.9 ± 0.7^{A}	8.8 ± 0.5^{A}	8.0 ± 0.1^{A}	8.4 ± 0.2^{A}	7.6 ± 0.5^{A}	2.883^{ns}
As^{1}	5.1 ± 0.0^{B}	3.6 ± 0.3^{C}	5.4 ± 0.1^{B}	6.0 ± 0.2^{A}	5.4 ± 0.1^{B}	5.4 ± 0.4^{B}	42.813^{***}
TS^{1}	32.9 ± 1.4^{A}	29.5 ± 1.7^{B}	29.3 ± 0.8^{B}	23.7 ± 0.8^{C}	$33,4 \pm 0.8^{A}$	29.7 ± 0.7^{B}	28.87^{***}
SS^{1}	6.3 ± 0.1^{A}	1.4 ± 0.1^{C}	5.7 ± 0.4^{B}	1.0 ± 0.0^{C}	1.1 ± 0.0^{C}	1.1 ± 0.1^{C}	597.58^{***}
CF^{1}	16.0 ± 1.1^{C}	17.4 ± 1.0^{BC}	17.4 ± 1.0^{BC}	14.3 ± 1.2^{C}	19.5 ± 0.6^{AB}	22.4 ± 1.7^{A}	18.07^{***}
Fat^{1}	18.1 ± 0.0^{BC}	20.1 ± 0.1^{A}	18.9 ± 0.0^{B}	19.0 ± 0.2^{B}	18.1 ± 0.4^{C}	17.4 ± 0.2^{C}	44.701^{***}
Pro^{1}	35.1 ± 0.1^{C}	34.9 ± 0.0^{D}	35.4 ± 0.1^{B}	35.8 ± 0.1^{A}	33.7 ± 0.1^{E}	33.6 ± 0.0^{E}	1052.00^{***}
Sap^{2}	45.2 ± 5.0^{C}	61.5 ± 5.5^{B}	42.2 ± 5.4^{C}	36.1 ± 5.4^{C}	60.7 ± 5.4^{B}	88.2 ± 6.5^{A}	34.85^{***}
TPP^{2}	1.7 ± 0.2^{E}	1.6 ± 0.2^{E}	14.6 ± 0.6^{B}	13.2 ± 0.2^{C}	3.1 ± 0.1^{D}	15.7 ± 0.7^{A}	964.10^{***}
Tan	-	-	+	+	-	+	
Flav	++++	+++	+++	+	++++	+	
Alk	++++	++++	++++	++++	++++	+	
Ster	++	++	+++	++	+++	+++	
Terp	++	++	+++	++	+++	+++	

ns: not significative; ***: $p < 0.001$; Means in a row followed by the same letter(s) do not differ significantly at the 5% level by Tukey's test. MA: main analysis, MC: moisture content, As: ash, TS: total sugar, SS: soluble sugar, CF: crud fiber, Pro: protein, Sap: saponins, TPP: total polyphenolic compounds, Tan: tannins, Flav: flavonoids, Alk: alkaloids, Ster: steroids, Terp: terpenoids, 1: in g/100 g dried matter, 2: in mg/1 g dried matter. Qualitative test: – = absent; + = present but not abundant; ++ = moderately abundant; +++ = abundant; ++++ = very abundant

Table 4 – Correlation between biochemical constituents of soybean varieties and the *Callosobruchus maculatus* susceptibility index (SI)

Biochemical constituents								
MC	**Ash**	**TC**	**S**	**Prot**	**Fat**	**TF**	**Sap**	**TPP**
0.57^{**}	0.40^{ns}	0.64^{**}	0.52^{*}	0.57^{**}	0.07^{ns}	-0.40^{ns}	-0.49^{*}	0.38^{ns}

ns: not significant; *: $p < 0.05$; **: $p < 0.01$; MC, moisture content; TC, total carbohydrates; S, soluble sugar; Prot, proteins; CF, crude fibre; Sap, saponins; TPP, total polyphenols

Phytochemical constituents of *Gnidia kraussiana* extracts

Phytochemical screening revealed differences among solvent fractions (*Table 5*). Acetone and methanol extracts contained abundant phenolic compounds, tannins, flavonoids, and cardiac glycosides, whereas hexane extracts were rich in terpenoids. Alkaloids and saponins were detected in the acetone and methanol extracts but were absent in the hexane extract.

Table 5 – Phytochemical analyses of *Gnidia kaussiana* extracts

Chemical	HE	AE	ME
Total phenolic compounds	–	+++	+++
Alkaloids	–	++	+
Saponins	–	–	+
Tannins	–	+	++
Flavonoids	–	++	+
Steroids	–	–	–
Terpenoids	+++	+	+
Cardiac glycosides	–	++	+++

HE, hexane extract; AE, acetone extract; ME, methanol extract; – = absent; + = present but not abundant; ++ = moderately abundant; +++ = abundant

Insecticidal activity of *Gnidia kraussiana* extracts against *Callosobruchus maculatus*

The corrected cumulative mortality of *C. maculatus* adults increased as a function of the extract type, concentration, and exposure duration (*Table 6a*). No mortality was recorded in the non-treated control. Neem seed oil, used as a positive control, exhibited moderate activity, with mortality reaching 17.4 and 48.0% after 7 days at 1 and 5 $g \cdot kg^{-1}$, respectively.

In contrast, *G. kraussiana* extracts induced higher mortality, with differences among solvent types. The hexane extract showed the highest efficacy, producing rapid and near-complete mortality. At 1 $g \cdot kg^{-1}$, mortality reached 83.0% after 7 days, while complete mortality (100%) was achieved at 5 $g \cdot kg^{-1}$ by day 3 and maintained thereafter. The acetone extract displayed intermediate activity, with mortality reaching 59.4 and 89.7% at 1 and 5 $g \cdot kg^{-1}$, respectively, after 7 days. The methanol extract was comparatively less effective, with a maximum mortality of 35.2 and 46.9% at 1 and 5 $g \cdot kg^{-1}$, respectively. Overall, mortality followed a consistent efficacy gradient, showing dose- and time-dependent responses: hexane extract > acetone extract > methanol extract > neem seed oil.

These descriptive patterns were strongly supported by the GLM (*Table 6b*), which identified the extract type, exposure period, and concentration as significant determinants of adult mortality. Compared to neem seed oil (reference category), all *G. kraussiana* extracts significantly increased mortality, with the largest effect observed for the hexane extract (B = 40.869, $p < 0.001$), followed by the acetone (B = 22.867, $p < 0.001$) and methanol extracts (B = 7.760, $p = 0.007$). Exposure time had a significant effect with early time points (days 1–4) associated with significantly lower mortality compared to day 7, as indicated by negative parameter estimates. This could be associated with a progressive accumulation of mortality over time, consistent with the temporal trends observed in *Table 6a*. In contrast, differences among later time points (days 5–7) were not statistically significant, suggesting that mortality approached a plateau under most treatments. The concentration exerted a strong positive effect on mortality (B = 8.637, $p < 0.001$), demonstrating that increasing the dose significantly enhanced the insecticidal activity across all extract types.

Overall, the combined evidence from *Table 6a* and *Table 6b* indicate that the insecticidal efficacy of *G. kraussiana* is driven by a synergistic interaction between solvent extraction, dosage, and exposure duration. The superior performance of the hexane extract suggests that most bioactive compounds are predominantly non-polar, and the lower efficacy of the methanol extract indicates a reduced contribution of polar constituents to acute toxicity.

Table 6a – Corrected cumulative mortality (Mean ± SE) of *Callosobruchus maculatus* in soybean seeds (variety: Kodek/Houla1) treated with solvent extracts from *Gnidia kaussiana*

Product	Conc. (g·kg^{-1})	1	2	3	4	5	6	7
NSO	0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	1	0.0 ± 0.0	1.3 ± 1.3	2.6 ± 1.5	4.0 ± 1.3	7.2 ± 1.5	8.6 ± 1.7	17.4 ± 0.5
	5	2.6 ± 1.5	6.5 ± 1.3	15.8 ± 0.3	15.7 ± 2.0	15.7 ± 1.3	41.4 ± 1.1	48.0 ± 3.3
G. kaussiana-Hex	0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	1	6.3 ± 2.4	20.5 ± 3.5	70.1 ± 2.5	74.8 ± 3.6	77.5 ± 2.7	77.3 ± 2.5	83.0 ± 2.4
	5	48.2 ± 3.5	87.2 ± 3.4	100.0 ± 0.0	100.0 ± 0.0	100.0 ± 0.0	100.0 ± 0.0	100.0 ± 0.0
G. kaussiana-Acet	0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	1	2.6 ± 1.5	13.0 ± 1.4	23.7 ± 1.6	39.5 ± 1.1	46.5 ± 1.3	50.1 ± 2.0	59.4 ± 2.2
	5	21.7 ± 2.2	35.0 ± 0.7	43.3 ± 1.8	71.0 ± 3.6	85.9 ± 2.9	85.6 ± 3.0	89.7 ± 3.7
G. kaussiana-MeOH	0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	1	1.3 ± 1.3	16.5 ± 1.2	22.1 ± 1.1	23.5 ± 1.8	28.6 ± 2.6	28.6 ± 2.6	35.2 ± 1.8
	5	3.8 ± 1.3	20.3 ± 2.1	24.6 ± 0.9	28.7 ± 1.9	32.6 ± 1.7	38.1 ± 2.6	46.9 ± 1.8

Conc.: Concentration, NSO: neem seed oil, *G. kaussiana*-Hex., *G. kaussiana*- Acet., *G. kaussiana*–methanol are extracts from *G. kaussiana* obtained using hexane, acetone, and methanol solvents, respectively

Table 6b – Generalised linear model (GLM) parameter estimates for the effects of *Gnidia kraussiana* extracts, neem seed oil, exposure period (days after infestation), and concentration on the adult mortality of *Callosobruchus maculatus*

Predictor	B (Estimate)	Std. Error	Wald Chi-Square	df	p-value
(Intercept)	4.822	3.3356	2.290	1	0.148
Hexane Extract	40.869	2.8633	203.735	1	0.000
Acetone Extract	22.867	2.8633	63.781	1	0.000
Methanol Extract	7.760	2.8633	7.345	1	0.007
Neem Seed Oil	0^a	-	-		
1 Day After Infestation	-32.772	3.7877	74.861	1	0.000
2 Days After Infestation	-23.293	3.7877	37.817	1	0.000
3 Days After Infestation	-14.784	3.7877	15.235	1	0.000
4 Days After Infestation	-10.216	3.7877	7.275	1	0.007
5 Days After Infestation	-7.119	3.7877	3.533	1	0.060
6 Days After Infestation	-4.261	3.7877	1.265	1	0.261
7 Days After Infestation	0^a	-	-		-
Concentration	8.637	0.4686	339.714	1	0.000

a: Reference category. Neem seed oil and 7 days after infestation were used as reference levels for the extract type and exposure period, respectively

Effects of *Gnidia kraussiana* extracts on progeny production and seed protection

The effects of *G. kraussiana* extracts on F_1 progeny emergence, progeny inhibition, seed damage, and weight loss are presented in *Table 7a*. In the non-treated controls (0 g·kg^{-1}), high levels of progeny emergence were recorded across all treatments (31.5–37.0 individuals) and accompanied by substantial seed damage (25.9–29.6%) and weight loss (2.6–2.9%), indicating severe infestation by *C. maculatus*.

The application of treatments at 1 and 5 g·kg^{-1} significantly reduced progeny emergence and associated damage. Neem seed oil completely suppressed progeny emergence at both doses, resulting in 100% inhibition and near-total seed protection, with damage reduced to ≤ 1.0% and weight loss to ≤ 0.1%. A similar pattern was observed for the hexane extract, which also achieved complete inhibition of progeny emergence (100%) at both concentrations, with negligible seed damage and no measurable weight loss.

The acetone extract exhibited dose-dependent efficacy. At 1 g·kg^{-1}, progeny emergence was reduced to 3.3 individuals, corresponding to 92.0% inhibition, and complete suppression (100%) was achieved at 5 g·kg^{-1}. This reduction was accompanied by a decrease in seed damage and weight loss. In contrast, the methanol extract was less effective with partial inhibition observed at 1 g·kg^{-1} (35.9%) and substantial but incomplete inhibition at 5 g·kg^{-1} (89.9%). Residual seed damage and weight loss remained higher under methanol treatment compared to other extracts. ANOVA indicated that weight loss was significantly affected by treatments for all extracts ($p < 0.01$), with treated seeds showing lower losses compared to controls. The GLM of progeny inhibition (*Table 7b*) revealed that the concentration had a strong and highly significant positive effect (B = 14.713, $p < 0.001$), indicating that the increasing dose significantly enhanced the inhibitory activity. In contrast, the extract type did not show a statistically significant effect compared to the reference treatment (neem seed oil), suggesting comparable efficacy among treatments when considering the concentration. Similarly, the GLM for seed damage (*Table 7c*) showed that the concentration had a significant negative effect (B = –4.029, $p < 0.001$), indicating that increasing doses reduced seed damage. The extract type did not significantly influence seed damage compared to neem seed oil, supporting the dominant role of concentration in determining the treatment efficacy.

Overall, *Table 7a*, *Table 7b* and *Table 7c* suggest that both *G. kraussiana* extracts and neem seed oil effectively suppress progeny development and protect soybean seeds from damage, with efficacy primarily driven by the concentration. Among the extracts, the hexane fraction exhibited the highest performance, achieving complete protection at both tested doses, whereas the methanol extract showed comparatively lower efficacy.

Repellence

Repellence values of *G. kraussiana* extracts and neem seed oil against *C. maculatus* are presented in *Table 8*. At 1 g·kg^{-1}, neem seed oil showed a negative repellence value (–28.3%), indicating that it was attractive rather than repellent to the insects (Class 0). At the higher dose (5 g·kg^{-1}), neem seed oil shifted to moderate repellence (34.9%, Class II). In contrast, all *G. kraussiana* fractions were repellent even at 1 g·kg^{-1}. The methanol fraction produced the strongest repellence (39.9%, Class II), followed by acetone (37.5%, Class II) and hexane fractions (21.6%, Class II). Increasing the dose to 5 g·kg^{-1} enhanced repellence, with the hexane fraction reaching 57.7% (Class III) and acetone 41.8% (Class III), while the methanol fraction remained moderately repellent (22.7%, Class II).

Overall, *G. kraussiana* extracts demonstrated consistent repellence, whereas neem seed oil was inconsistent, showing attractiveness at a low dose but moderate repellence at higher dose.

Table 7a – Effects of two doses of neem seed oil and *Gnidia kraussiana* fractionated extracts on F_1 progeny emergence, inhibition of progeny production, seed damage, and weight loss of soybean infested by *Callosobruchus maculatus*

Product/ Dose (g·kg^{-1})	Number of F_1 progeny emerged	Inhibition of progeny production relative to control (%)	Damage	Seed weight loss (%)
		Seed oil		
0	36.3 ± 6.5	0.0 ± 0.0	28.1 ± 2.4	2.7 ± 0.2^{a}
1	0.0 ± 0.0	100.0 ± 0.0	1.0 ± 0.4	0.1 ± 0.1^{b}
5	0.0 ± 0.0	100.0 ± 0.0	0.5 ± 0.3	0.0 ± 0.0^{b}
F-value	-	-	-	159.93***
		Hexane		
0	37.0 ± 4.9	0.0 ± 0.0	25.9 ± 4.6	2.9 ± 0.8^{a}
1	0.0 ± 0.0	100.0 ± 0.0	0.1 ± 0.1	0.0 ± 0.0^{b}
5	0.0 ± 0.0	100.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0^{b}
F-value	-	-	-	14.47***
		Acetone		
0	31.5 ± 0.7	0.0 ± 0.0	28.1 ± 2.4	2.7 ± 0.2^{a}
1	3.3 ± 1.3	92.0 ± 3.2	1.2 ± 0.8	0.4 ± 0.2^{b}
5	0.0 ± 0.0	100.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0^{b}
F-value	-	-	-	94.09***
		Methanol		
0	35.0 ± 4.6	0.0 ± 0.0	29.6 ± 2.3	2.6 ± 0.5^{a}
1	22.0 ± 2.2	35.9 ± 3.6	2.2 ± 1.4	0.4 ± 0.2^{b}
5	4.0 ± 2.4	89.9 ± 5.8	3.4 ± 2.8	0.5 ± 0.3^{b}
F-value	-	-	-	12.29**

**: p < 0.01; ***: p < 0.001; Means in a column followed by the same letter(s) do not differ significantly at the 5% level by Tukey's test

Table 7b – Generalised linear model (GLM) parameter estimates for the inhibitory effects of *Gnidia kraussiana* extracts and neem seed oil on *Callosobruchus maculatus*

Predictor	B (Estimate)	Std. Error	Wald Chi-Square	df	p-value
(Intercept)	37.241	10.5301	12.508	1	0.000
Hexane Extract	-5.199	13.5141	0.000	1	1.000
Acetone Extract	-2.680	13.5141	0.039	1	0.843
Methanol Extract	-11.770	13.5141	0.759	1	0.384
Neem Seed Oil	0^{a}	-	-	-	-
Concentration	14.713	2.2118	44.249	1	0.000

a: Reference category. Neem seed oil was used as the reference treatment

Table7c – Generalised linear model (GLM) parameter estimates for the effects of *Gnidia kraussiana* extracts and concentration on seed damage in soybean infested by *Callosobruchus maculatus*

Predictor	B (Estimate)	Std. Error	Wald Chi-Square	df	p-value
(Intercept)	17.434	3.2185	29.340	1	0.000
Hexane Extract	-0.727	4.1306	0.031	1	0.860
Acetone Extract	0.412	4.1306	0.010	1	0.921
Methanol Extract	1.382	4.1306	0.112	1	0.738
Neem Seed Oil	0[a]	-	-	-	-
Concentration	-4.029	0.6760	35.525	1	0.000

a: Reference category. Neem seed oil was used as the reference treatment

Table 8 – Mean percentage of repellence (PR) of three *Gnidia kaussiana* fractions against *Callosobruchus maculatus*

Product	repellency values	Class	Interpretation
	NSO		
1	-28.30 ± 5.59	0	Attractive
5	34.88 ± 8.25	II	Moderately repellent
	G. kaussiana-Hex		
1	21.61 ± 3.73	II	Moderately repellent
5	57.66 ± 8.38	III	Averagely repellent
	G. kaussiana-Acet		
1	37.49 ± 3.84	II	Moderately repellent
5	41.78 ± 5.34	III	Averagely repellent
	G. kaussiana-MeOH		
1	39.88 ± 6.40	II	Moderately repellent
5	22.68 ± 3.67	II	Moderately repellent

DISCUSSION

This study suggests that the susceptibility of soybean to *C. maculatus* is primarily influenced by internal biochemical traits and that environmental conditions significantly modulate the expression of this susceptibility. In addition, the results showed that protection of a highly susceptible genotype can be improved through the application of botanical extracts derived from *G. kraussiana*.

Unlike previous studies that have examined either host plant resistance or botanical insecticides separately, the present study integrated both approaches in a single soybean storage system, providing a more comprehensive understanding of their complementary roles. Together, these findings support an integrated framework combining host-mediated antibiosis and plant-derived insecticidal activity under storage conditions.

Environmental and seed trait determinants of varietal resistance

Although significant variation was observed in seed mass and geometric parameters, these physical traits did not consistently explain differences in susceptibility among soybean varieties.

Early work has suggested that mechanical characteristics, such as seed coat hardness may influence bruchid penetration (Dobie, 1974), and similar effects have been reported in legumes (Allotey and Oyewo, 2005).

However, more recent studies have emphasised the predominant role of internal chemical composition in determining larval survival and development (Loko *et al*., 2022; Swamy *et al*., 2020).

These findings support this interpretation. By linking these biochemical traits to observed susceptibility patterns across multiple soybean genotypes, this study reveals the chemical basis of resistance in soybean, a crop for which such integrative analyses remain limited. Biochemical analyses were conducted on a subset of six varieties selected to represent the range of susceptibility based on the availability of plant material. Although this targeted approach enhances the biological relevance of comparisons, it may limit the generalisability of the observed relationships.

The correlation analysis conducted under Sudano–Guinean conditions suggests that the soluble sugar content is positively associated with susceptibility. This relationship may be explained by the reliance of early larval stages on readily available carbohydrates before the full activation of proteolytic systems (Swamy *et al*., 2020). Seeds with a higher soluble sugar content may facilitate larval establishment and increase adult emergence. Similar nutrient performance relationships have been reported in other bruchid–legume systems (Kpoviessi *et al*., 2021; Loko *et al*., 2022). However, this relationship should be cautiously interpreted given the limited number of varieties analysed (n = 6), which may influence the correlation strength (Cohen, 1988).

In contrast, polyphenolic compounds showed a negative association with susceptibility. These compounds are known to bind proteins, inhibit digestive enzymes, and reduce the nutrient assimilation efficiency of herbivorous insects (Singh *et al*., 2021). In bruchids, whose larvae develop within a single seed, such biochemical constraints directly reduce growth and survival. Comparable relationships between the phenolic content and resistance have been reported in cowpea and related legumes (Kosini *et al*., 2019; Kpoviessi *et al*., 2021).

Saponins, which were present at higher concentrations in some resistant genotypes, may contribute antibiosis effects. These compounds interact with membrane sterols and may disrupt gut integrity, impairing nutrient absorption (Hazzam *et al*., 2025). Overall, these results indicate that resistance is chemically multifactorial rather than controlled by a single defensive compound, consistent with broader plant–insect interaction theory (War *et al*., 2012). This multifactorial perspective, supported by simultaneous biochemical and biological data, reinforces the novelty of this study in moving beyond single trait explanations of resistance.

The same batch of soybean seeds was used across experimental sites, indicating that the biochemical composition remained constant. Therefore, differences in susceptibility between the Sudano–Sahelian and Sudano–Guinean zones are attributable to environmental conditions. Temperature and relative humidity are known to strongly influence the development rate, fecundity, and survival of *C. maculatus* (Pawlowski *et al*., 2021). The significant site effect revealed by GLM may indicate the importance of environmental modulation.

Although varietal rankings were relatively consistent, differences in magnitude indicate that resistance is dependent on the environment. This supports the concept of genotype × environment interactions in host–insect systems (Fox *et al*., 2003), suggesting that resistance should not be considered a fixed trait. By combining multi-site evaluation with biochemical profiling, this study provides additional insights into how environmental conditions modulate chemically mediated resistance in soybean storage systems.

Toxicological performance of *Gnidia kraussiana* extracts

Botanical treatments applied to the susceptible genotype demonstrated strong insecticidal activity. Hexane and acetone extracts produced rapid mortality, high F_1 progeny suppression, and reduced seed damage. Similar insecticidal effects of *G. kraussiana* against bruchids have been reported in stored legumes (Kosini and Nukenine, 2017; Kosini *et al*., 2021), supporting the reproducibility of these findings.

The present study extends these findings by demonstrating the practical effectiveness of these extracts in a soybean storage context, which has been comparatively less investigated. The GLM confirmed that extract type, concentration, and exposure time significantly influenced mortality. The positive effect of concentration indicates a clear dose–response relationship, which is consistent with previous studies on botanical insecticides (Isman, 2020). The temporal pattern reflects the cumulative mortality dynamics typical of plant-derived insecticides (Regnault-Roger *et al*., 2012).

The higher efficacy of hexane and acetone extracts suggests that active compounds are mainly non-polar or moderately polar. These fractions are typically rich in terpenoids, alkaloids, and phenolic compounds, which act through neurotoxicity, respiratory inhibition, or metabolic disruption (Isman, 2020; Talukder and Howse, 1994). The observed rapid mortality suggests that contact and fumigant effects play a major role.

In contrast, the methanol extract was less effective, indicating that highly polar compounds alone may not provide sufficient toxicity. This solvent-dependent variation in efficacy has been widely reported and reflects differences in the extraction efficiency of bioactive compounds (Rohimatun *et al*., 2024).

The strong inhibition of the higher efficacy of hexane and acetone extracts suggests that bioactive compounds may be predominantly non-polar or moderately polar. These fractions are often associated with compounds, such as terpenoids, alkaloids, and phenolic constituents, which have been reported to have insecticidal effects through neurotoxicity, respiratory inhibition, or metabolic disruption mechanisms (Isman, 2020; Talukder and Howse, 1994). However, the present study did not include chemical characterisation (e.g., gas chromatography–mass spectrometry); therefore, the specific compounds responsible for the observed activity cannot be identified with certainty.

The strong inhibition of progeny emergence may be associated with a combination of adulticidal and larvicidal effects. However, because oviposition and egg viability were not measured separately, the underlying mechanisms cannot be distinguished. Similar patterns have been reported in studies on botanical insecticides (Kosini and Nukenine, 2017). Complete progeny suppression observed in the susceptible soybean variety highlights the potential of botanical extracts as a complementary strategy when host plant resistance is limited. Progeny emergence likely results from combined adulticidal and larvicidal effects.

Persistence and storage protection

A notable finding is the persistence of protective effects over four months of storage. Although non-treated seeds showed substantial damage and weight loss, treated seeds particularly those exposed to hexane and acetone extracts at higher concentrations remained effectively protected. This long-term protection under ambient storage conditions provides practical relevance, as it reflects real smallholder storage environments rather than controlled laboratory conditions.

Although botanical insecticides are often considered short-lived, certain compounds, particularly terpenoids and phenolics, may exhibit prolonged activity under storage conditions (Alves *et al*., 2024; Regnault-Roger *et al*., 2012). The storage environment, which characterised by reduced light exposure and limited air exchange, may contribute to compound stability.

Implications for integrated pest management

The results support the development of an integrated pest management strategy combining host plant resistance and botanical insecticides. Biochemical resistance mechanisms reduce host suitability, and plant-derived compounds provide additional mortality pressure. Although botanical treatments were only evaluated only on a highly susceptible variety, the combined findings of this study provide complementary evidence suggesting that these approaches can be effectively integrated.

Thus, the novelty of this study lies in linking the biochemical determinants of varietal resistance with the targeted use of botanical extracts as a compensatory protection strategy for susceptible genotypes rather than treating these approaches as independent control options. This combination may reduce the risk of resistance development due to the complex mixture of bioactive compounds (Alves *et al*., 2024; Isman, 2020). This approach is particularly relevant for smallholder storage systems in sub-Saharan Africa where access to synthetic insecticides is limited (Njume *et al*., 2019).

The use of locally available plant resources, such as *G. kraussiana*, offers a sustainable and environmentally safer alternative. However, because botanical treatments were only evaluated on a susceptible genotype, potential interactions between host resistance and botanical protection remain hypothetical and require further investigation. Future studies should test this integrated approach across multiple resistance categories to validate its applicability.

This study has several limitations that should be acknowledged. First, the chemical composition of *G. kraussiana* extracts was not characterised using analytical techniques, such as gas chromatography–mass spectrometry, limiting the identification of the compounds responsible for the observed insecticidal activity. Second, variability in varietal responses between locations indicates that resistance expression is influenced by environmental conditions, highlighting potential genotype–environment interactions. Third, some soybean varieties exhibited inconsistent responses across sites, limiting their classification into a single resistance category and affecting their general applicability. Finally, oviposition and egg viability were not separately assessed, preventing a precise distinction between adulticidal and developmental treatment effects.

To strengthen our causal inference and mechanistic understanding, future studies should consider measuring larval digestive enzyme activity in resistant versus susceptible genotypes, quantifying oxidative stress responses in insects exposed to botanical extracts, conducting chemical profiling (e.g., gas chromatography–mass spectrometry) of active fractions, and testing botanical extracts across multiple resistance categories.

CONCLUSIONS

This study suggests that the *C. maculatus* resistance of soybean is influenced by intrinsic biochemical traits and that the effective protection of susceptible varieties can be achieved through the application of *G. kraussiana* root extracts under storage conditions. Significant genotypic variation was observed among the 10 soybean varieties evaluated across 2 agroecological zones of northern Cameroon.

Several varieties consistently expressed resistant or moderately resistant responses, whereas Kodek/Houla1 was the most susceptible. Physical seed characteristics showed limited explanatory power, whereas the biochemical composition, particularly soluble sugars and polyphenolic compounds, emerged as key determinants of susceptibility.

The positive correlation between soluble sugars and susceptibility, coupled with the negative association between polyphenols and SI, suggests that host nutritional suitability and defensive secondary metabolites jointly regulate bruchid performance. Therefore, resistance appears to be chemically mediated and multifactorial rather than primarily structural.

The botanical protection of the susceptible genotype proved highly effective. The hexane and acetone fractions of *G. kraussiana* induced rapid adult mortality, completely suppressed progeny emergence at 5 $g \cdot kg^{-1}$, and prevented seed damage and weight loss over four months of storage. Their performance equalled or exceeded that of neem seed oil. The strong adulticidal, progeny-inhibiting, and repellent activities suggest the involvement of non-polar and moderately polar bioactive phytochemicals acting through multiple modes of action.

Overall, the findings support the following practical integrated strategy for soybean storage protection in tropical smallholder systems: (i) preferential cultivation and storage of resistant varieties, and (ii) targeted botanical treatment of susceptible genotypes using locally available plant resources. Such an approach reduces reliance on synthetic insecticides, enhances environmental safety, and aligns with sustainable postharvest pest management principles.

Future research should focus on the chemical characterisation of active fractions, the elucidation of insect physiological responses to resistant seed biochemistry, and the evaluation of combined varietal–botanical strategies under on-farm storage conditions.

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REFERENCES

Abbott, W.S. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* **1925**, *18*, 265-267. https://doi.org/10.1093/jee/18.2.265a

Adeniyi, S.A.; Orjiekwe, C.L.; Ehiagbonare, J.E.; Arimah, B.D. Preliminary phytochemical analysis and insecticidal activity of ethanolic extracts of four tropical plants against beans weevil. *International Journal of the Physical Sciences* **2010**, *5*, 753-762.

Allotey, J.; Oyewo, E.O. Some aspects of the biology and control of *Callosobruchus maculatus* on stored soybean varieties. *African Journal of Food, Agriculture, Nutrition and Development* **2005**, *4*(2), 1-11. https://doi.org/10.4314/ajfand.v4i2.19160

Alves, D.R.; Andrade, J.C.A.; Nobre, D.A.C. Natural compounds and their potential use in stored grain treatment: A review. *Revista Brasileira de Engenharia Agrícola e Ambiental* **2024**, *29*, e279969. https://doi.org/10.1590/1807-1929/agriambi.v29n1e279969

Ashamo, M.O.; Ileke, K.D.; Ogungbite, O.C. Entomotoxicity of some agro-wastes against cowpea bruchid, *Callosobruchus maculatus* (Fab.) [Coleoptera: Chrysomelidae] infesting cowpea seeds in storage. *Heliyon* **2021**, *7* (6), e07202. https://doi.org/10.1016/j.heliyon.2021.e07202

Asra, A.; Syahrani, W.; Maharani, B.; Al Atsariyah, L.; Joshi, R.C.; Brugman, E.; Fernando, I.; Mario, M.B. Biology of *Callosobruchus maculatus* (Fabricius) (Coleoptera: Chrysomelidae) on two commercial beans in Makassar, Indonesia. FAO AGRIS – International System for Agricultural Science and Technology 2025. https://agris.fao.org/search/en/providers/125622/records/6943d3c5f2f3b9724407b686 (accessed at 20 March 2025).

Cohen, J. Statistical power analysis for the behavioral sciences, 2nd ed. Lawrence Erlbaum Associates, 1988.

Devani, M.B.; Shishoo, J.C.; Shal, S.A.; Suhagia, B.N. Spectrophotometrical method for determination of nitrogen in Kjeldahl digest. *Journal of the Association of Official Analytical Chemists* **1989**, *72*, 953-956. https://doi.org/10.1093/jaoac/72.6.953

Dobie, P. The laboratory assessment of the inherent susceptibility of maize varieties to post-harvest infestation by *Sitophilus zeamais* Motsch. (Coleoptera, Curculionidae). *Journal of Stored Products Research* **1974**, *10*, 183-197. https://doi.org/10.1016/0022-474X(74)90006-X

Dubois, M.; Gilles, K.A.; Hamilton, J.K.; Roberts, P.A.; Smith, F. Colorimetric method for determination of sugars and related substances. *Analytical Chemistry* **1956**, *28*, 350-356.

Fox, C.W.; Bush, M.L.; Wallin, W.G. Maternal age affects offspring lifespan of the seed beetle, Callosobruchus maculatus. *Functional Ecology* **2003**, *15*(6), 811-820. https://doi.org/10.1111/j.1365-2435.2003.00799.x

Hazzam, K.E.; Mhada, M.; Bakrim, W.B.; Zhar, H.; Bchitou, R.; El Alaoui, M. Antinutritional and insecticidal potential of Chenopodium quinoa saponin extract against *Tribolium castaneum* (Herbst) and its action mechanism. *Scientific Reports* **2025**, *15*, 6829. https://doi.org/10.1038/s41598-025-90952-x

Jilani, G.; Su, H.C.F. Laboratory studies on several plant materials as insect repellants for protection of cereal grains. *Journal of Economic Entomology* **1983**, *76*, 154-157. https://doi.org/10.1093/jee/76.1.154

Kalpna, Hajam, Y.A.; Kumar, R. Management of stored grain pest with special reference to *Callosobruchus maculatus*, a major pest of cowpea: A review. *Heliyon* **2022**, *8*(1), e08703. https://doi.org/10.1016/j.heliyon.2021.e08703

Kosini, D.; Nukenine, E.N. Bioactivity of Novel Botanical Insecticide From *Gnidia kaussiana* (Thymeleaceae) Against *Callosobruchus maculatus* (Coleoptera: Chrysomelidae) in Stored *Vigna subterranea* (Fabaceae) Grains. *Journal of Insect Science* **2017**, *17*(1), 31. https://doi.org/10.1093/jisesa/iex004

Kosini, D.; Nukenine, E.N.; Agbor, G.A.; Tchinda, A.T.; Abdou, J.P.; Yaya, J.A.G.; Kowa, T.K. Fractionated Extracts From *Gnidia kraussiana* (Malvales: Thymeleaceae) as Bioactive Phytochemicals for Effective Management of *Callosobruchus maculatus* (Coleoptera: Chrysomelidae) in Stored *Vigna unguiculata* (Fabales: Fabaceae) Seeds. *Journal of Insect Science* **2021**, *21*(1), 14. https://doi.org/10.1093/jisesa/ieab006

Kosini, D.; Nukenine, E.N.; Saidou, C.; Noubissié, J.T.; Dolinassou, S. Environmental and seed traits influencing cowpea resistance to *Callosobruchus maculatus*. *Bulletin of Entomological Research* **2019**, *109*, 595-603. https://doi.org/10.1017/S0007485318000962

Kpoviessi, A.D.; Agbahoungba, S.; Agoyi, E.E.; Nuwamanya, E.; Assogbadjo,

A.E.; Chougourou, D.C.; Adoukonou-Sagbadja, H. Primary and secondary metabolite compounds in cowpea seeds resistant to the cowpea bruchid [*Callosobruchus maculatus* (F.)] in postharvest storage. *Journal of Stored Products Research* **2021**, *93*, 101858. https://doi.org/10.1016/j.jspr.2021.101858

Loko, Y.L.E.; Toffa, J.; Orobiyi, A.; Dassou, G.A.; Okpeicha, R.; Gavoedo, D.; Dansi, A. Effects of Seed Physical Characteristics of Benin Soybean Germplasm on their Resistance to *Callosobruchus maculatus* Fabricius (Coleoptera: Bruchidae). *Sarhad Journal of Agriculture* **2022**, *38*(4), 1468-1477. https://doi.org/10.17582/journal.sja/2022/38.4.1468.1477

MINEPAT. Report on the competitiveness of the Cameroonian economy (RCC 2023). https://minepat.gov.cm/wp-content/uploads/2020/07/RCC-2023-EN.pdf (accessed 15 December 2023).

Njume, A.C.; Kamanga, B.M.; Widodo, S.; Purwanto, Y.A. Improving postharvest handling of soybean (Glycine Max (L.) Merrill) in Cameroon. *Journal of Applied Physical Sciences* **2019**, *5*, 48-59. https://doi.org/10.20474/japs-5.2.2

Nwosu, L.C.; Azeez, O.M.; Eluwa, A.N.; Petgrave, G.M. Two stubborn storage insect pests, *Callosobruchus maculatus* and Sitophilus zeamais: Biology, food security problems and control strategies. *International Research Journal of Insect Sciences* **2020**, *5*(1), 7-13. https://doi.org/10.18488/journal.106.2020.51.7.13

Pawlowski, M.L.; Lagos-Kutz, D.M.; da Fonseca Santos, M.; Lee, N.; Chigeza, G.; Nachilima, C.; Botan Francischini, J.H.M.B.; Hartman, G.L. Potential threat of bruchids on soybean production in sub-Saharan Africa. *Plant Health Progress* **2021**, *22*, 86-91. https://doi.org/10.1094/PHP-10-20-0093-MR

Pessu, P.O.; Umeozor, O.C. Susceptibility of cowpea and soybean to *Callosobruchus maculatus*. *Indian Journal of Agricultural Research* **2004**, *38*, 102-106.

Rohimatun, R.; Dewi Nur Aisyah, M.; Rismayani, R.; Rizal, M.; Noveriza, R.; Mardiningsih, T.L. Evaluation of citronella oil nanoemulsion formulation against the insect-stored pest *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). *Journal of Plant Protection Research* **2024**, *64*, 288-297. https://doi.org/10.24425/jppr.2024.151256

Regnault-Roger, C.; Vincent, C.; Arnason, J.T. Essential oils in insect control: low-risk products in a high-stakes world. *Annual Review of Entomology* **2012**, *57*, 405-424. https://doi.org/10.1146/annurev-ento-120710-100554

Saha, A.; Mandal, S. Nutritional benefit of soybean and its research advances. *Sustainable Food Production* **2019**, *5*, 6-16. https://doi.org/10.18052/www.scipress.com/SFP.5.6

Singh, S.; Kaur, I.; Kariyat, R. The Multifunctional Roles of Polyphenols in Plant-Herbivore Interactions. *International Journal of Molecular Sciences* **2021**, *22*(3), 1442. https://doi.org/10.3390/ijms22031442

Swamy, S.V.S.G.; Raja, D.S.; Wesley, B.J. Susceptibility of stored chickpeas to bruchid infestation as influenced by physico-chemical traits of the grains. *Journal of Stored Products Research* **2020**, *87*, 101583. https://doi.org/10.1016/j.jspr.2020.101583

Talukder, F.A.; Howse, P.E. Repellent, toxic and protectant effects of

Aphanamixis polystachya extracts. *Journal of Chemical Ecology* **1994**, *20*, 899-908. https://doi.org/10.1007/BF02059586

Tingem, M.; Rivington, M.; Bellocchi, G.; Azam-Ali, S.; Colls, J. Effects of climate change on crop production in Cameroon. *Climate Research* **2008**, *36*, 65-77. https://doi.org/10.3354/cr00733

Tripathi, K.; Prasad, T.V.; Bhardwaj, R.; Jha, S.K.; Semwal, D.P.; Gore, P.G.; Sharma, P.K.; Bhalla, S. Evaluation of diverse germplasm of cowpea [Vigna unguiculata (L.) Walp.] against bruchid [Callosobruchus maculatus (Fab.)] and correlation with physical and biochemical parameters of seed. *Plant Genetic Resources: Characterization and Utilization* **2020**, *18*(3), 120-129. https://doi.org/10.1017/S1479262120000180

War, A.R.; Paulraj, M.G.; Ahmad, T.; Buhroo, A.A.; Hussain, B.; Ignacimuthu, S.; Sharma, H.C. Mechanisms of plant defense against insect herbivores. *Plant Signaling & Behavior* **2012**, *7*(10), 1306-1320. https://doi.org/10.4161/psb.21663

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