



Research Article

Evaluation of aqueous extracts of *Azadirachta indica*, *Cassia occidentalis* and *Calotropis procera* on germination, growth parameters and fungal infections of cashew seedlings (*Anacardium occidentale* L.)

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Abstract

Anacardium occidentale L. is a plant of great economic importance, but its cultivation is confronted with problems of infection, growth and development due to the poor quality of the seeds, which are affected by fungal diseases that lead to crop losses. The objective of this study is to improve cashew nut production by treating the seeds with plant extracts before sowing. To achieve this objective, cashew seeds accessions in Cameroon and Chad were soaked in extracts of *Azadirachta indica*, *Calotropis procera* and *Cassia occidentalis* for 12 hours for treatment. These seeds were sown in pots containing black soil and sand as substrate to collect data on germination percentage, infection percentage and growth parameters (colar diameter, height, number of leaves, seedling vigour and plant vigour). The results showed that the seeds of the of the Koumra and Garoua accessions treated with *A. indica* extract at all concentrations showed 100 % germination and 0 % infection. Similarly, all seedlings from the seeds treated with the different plant extracts showed significantly higher crown diameters, heights, number of leaves and seedling vigour than seedlings from non-treated seeds. These results suggest the use of these plant extracts to reduce cashew seed infections and to enhance seed germination and seedling vigour.

1. Introduction

Cashew (*Anacardium occidentale* L.) is a perennial plant belonging to the Anacardiaceae family, native to Latin America, more precisely Brazil [1,2]. Its discovery was made by the Portuguese who introduced it to their colonies in Africa and Asia as a reforestation plant [3]. The plant is nowadays widely cultivated in many tropical and subtropical regions for its fruit, which is very rich in vitamins (vitamin A, C, B1, B2, B3, B5), carbohydrates, minerals (calcium, phosphorus, iron),

fiber, essential fatty acids and amino acids [4]. Formerly used as a protective species against bushfires around classified forests or as a windbreak around fields or to fight deforestation [5], today the plant is cultivated mainly for the exploitation of its nut and apple, which are traded extensively worldwide [6, 7].

Cashew is a fast-growing cash crop. This represents an opportunity for many African countries to earn

foreign exchange through the export of its raw nuts [8-10]. This is because the global demand for cashew nuts has been increasing over the years until today [11]. In addition, parts of the tree such as the bark, roots and leaves are used as antioxidant and anti-inflammatory in the treatment of certain diseases [12, 13]. The nut shell is used in the manufacture of many chemical products such as: inks, varnishes, insecticides, insulators [14, 11] and its kernel is used in the food industry especially in biscuit and pastry making and even in the preparation of certain dishes [15].

World cashew nut production in 2019 was estimated at 3.66 million tons and African production accounted for 60 % of world production, i.e. about 2.196 million tons. Ivory Coast was the world's largest producer with a production of 900 thousand tons [16]. Cameroon and Chad, despite their high potential, are not among the main cashew producing countries [7]. The governments of these two countries aim to be among the main cashew nut producing countries. Cameroon's ambition is to be among the main cashew nut producing countries by 2023. To this end, the Cameroonian government and numerous experts have initiated numerous projects in the Far North, North, Adamaoua and East regions with the aim of supporting farmers through advice and the distribution of cashew seeds or seedlings and agricultural inputs [11]. In Chad, the government has been implementing strategies to intensify cashew cultivation since 2000. These strategies include land distribution and capacity building of the farmers [17]. To be among the main cashew nut producing countries, the Cameroonian and Chadian governments should provide farmers with good quality seeds that can germinate and produce seedlings and uninfected plants with good vigour. Hence the need to check sanitary quality before sowing. It was reported that seed health is the most important parameter of seed quality because most pathogens initially present in the seed can lead to progressive disease development in the field and affect crop production [18]. Of the various factors that affect seed health, the most important are seed-borne pathogens that not only reduce the germination rate of the seed, but also the vigour of the seedlings and later the plants, resulting yield losses of more than 40

% [19]. These fungi have been also reported to cause fungal diseases such as: damping-off (*Fusarium* spp.), seed rot (*Pilgariella anacardii*), anthracnose (*Colletotrichum gloeosporioides*), pestalotiosis (*Pestalotia heterocornis*), powdery mildew (*Oidium anacardii*) [20-23].

The control of these different fungal species, is done through seed treatment with chemical fungicides to protect the seeds and future seedlings and plants. It was reported by [24] showed that chemical fungicides based on Carbendazine and Prochloraz can reduce cashew seed-associated fungi and seed-borne fungi in seedlings. However, these chemicals lead to problems of toxicity for consumers due to the presence of their residues in agricultural products and environmental pollution [25]. Moreover, their abusive use leads to the development of resistance in certain fungi [26]. It is therefore necessary to explore other alternative control methods to chemical control. This alternative control can be directed towards the use of plant derivatives such as plant extracts. Plant extracts have the advantage of being not only available to farmers, but also non-toxic and easily biodegradable and therefore environmentally safe [27]. The work of [28], showed that treatment of cotton seeds with the aqueous extracts of *Azadirachta indica*, *Boswellia dalzielii* and *Cassia sieberiana* leaves before sowing, would reduce the incidence of seed-borne fungi to seedlings, improve seed germination rate and seedling vigour. The present study aims to improve cashew nut production through seed treatment with plant extracts before sowing.

2. Materials and methods

2.1 Preparation of plant extracts

The different plant extracts were prepared from the seeds of *Azadirachta indica*, the aerial parts of *Calotropis procera* (leaves, flowers and stem) and the young leaves of *Cassia occidentalis* collected very early in the morning in the outskirts of the city of Ndjamen, the political capital of Chad. These different organs were washed with tap water, dried separately in the shade for a fortnight and finely ground in a mill [29] before being transported to the University of Dschang in Cameroon, where the extractions were made. For the extractions, 100 g of powder of each plant was macerated in 500 mL of distilled water and the whole mixed. The mixture was filtered through Whatman

N°1 paper after 48 hours of maceration. The filtrates obtained were oven-dried at a temperature of 40° C [28].

2.2 Seed treatment of *Anacardium occidentale* and sowing

The seeds of *Anacardium occidentale* used in this study were made up of accessions collected and dried in Cameroon (Ngaoundéré, Garoua and Yagoua) and Chad (Koumra, Moundou and Sahr). These seeds were put in appropriate plastic bags, labelled and transported to the Phytopathology and Agricultural Zoology Research Unit (UR_PHYZA) of the University of Dschang.

Once at the research unit, these cashew seeds were washed separately with tap water, then disinfected in a 5 % sodium hypochlorite solution for 5 minutes, rinsed 3 times successively with distilled water to remove traces of the disinfectant and then placed on hydrophilic paper to remove excess water. The treatment consisted of macerating these different seeds in plant extracts at concentrations of 25, 50 and 75 mg/mL for 12 hours [30]. The seeds were macerated in distilled water or in a Monchamp fungicide at the manufacturer's dose (0.3 mg/mL) and served as negative and positive controls respectively. After treatment, these seeds were sown in 15 cm diameter and 25 cm high perforated plastic pots. Each pot was filled with a substrate consisting of black soil and sand in a ratio of 2:1. This substrate was previously sterilised in an autoclave at a temperature of 125° C for 1 hour [31]. These pots were placed in an open-air space. Maintenance consisted of watering or pulling weeds when necessary. This experiment was done in a completely randomised design with 3 replicates. The different data collected were the percentage of seed germination, the percentage of seedling infection and the agronomic parameters.

The germination of the seeds was monitored every day and the percentage of germination (PG) was calculated by the following formula:

$$PG (\%) = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds sown}} \times 100$$

The percentage of infection (PI) of emerged seedlings was calculated using the following formula:

$$PI (\%) = \frac{\text{Number of infected seedlings}}{\text{Total number of seedling}} \times 100$$

Growth parameters included: collar diameter, height, number of leaves and seedling vigour. Seedling neck diameter was measured with a slide caliper. Height measurements were made with a decimeter from the crown to the terminal bud. For the assessment of plant vigour (VP), it was the ratio of the average seedling height (SH) by the average seedling diameter (SD) [32].

$$VP = \frac{SH}{SD} \times 100$$

2.3 Statistical analyses

The collected data were subjected to analysis of variance (ANOVA). Means were separated using Duncan's Test at the $P \leq 0.05$. SPSS version 22.0 was used for this purpose.

3. Results

3.1 Effect of the different plant extracts on cashew seed germination percentage and seedling infection percentage

The different plant extracts improved the germination percentage of cashew seeds by showing higher germination percentages than the negative controls (Table 1). *Azadirachta indica* extracts gave 100 % germination with seeds from the Koumra and Garoua accessions at all concentrations. The same percentage of germination (100 %) was obtained with seeds of the Sarh and Yagoua accessions on the one hand and the Moundou and Ngaoundéré accessions on the other hand, when treated with *A. indica* extracts at concentrations of 50 and 75 mg/mL respectively.

With *Calotropis procera* extracts, the Koumra accessions at all concentrations, as well as those of Sarh and Moundou from the 50 mg/mL concentration, all germinated (100 % germination). Seeds from the Ngaoundéré accessions showed germination percentages of 33.33, 66.66 and 100 % at concentrations of 25, 50 and 75 mg/mL respectively. The Garoua and Ngaoundéré accessions, when treated with *Cassia occidentalis* extracts at the concentration of 25 mg/mL, all germinated. Unlike the 50 and 75 mg/mL concentrations, where the germination percentages were 66.66 and 33.33% respectively. Seeds from the Moundou and Yagoua accessions treated with *C. occidentalis* extracts, at all concentrations, showed 100 % germination. The aqueous extracts of the three plants reduced the percentage of infections in cashew seedlings (Table 2).

Table 1: Effect of plant extracts on the percentage of germination (%) of *Anacardium occidentale* L. seeds

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	66.66 ^b	33.33 ^b	33.33 ^c	66.66 ^b	66.66 ^b	66.66 ^b
25 mg/mL	100 ^a	33.33 ^b	66.66 ^b	100 ^a	66.66 ^b	66.66 ^b
50 mg/mL	100 ^a	33.33 ^b	100 ^a	100 ^a	66.66 ^b	100 ^a
75 mg/mL	100 ^a	100 ^a	100 ^a	100 ^a	100 ^a	100 ^a
T+	33.33 ^c	100 ^a	100 ^a	100 ^a	100 ^a	66.66 ^b
<i>Calotropis procera</i>						
T-	66.66 ^b	33.33 ^c	33.33 ^c	66.66 ^b	66.66 ^b	66.66 ^a
25 mg/mL	100 ^a	66.66 ^b	66.66 ^b	100 ^a	33.33 ^c	33.33 ^c
50 mg/mL	100 ^a	100 ^a	100 ^a	100 ^a	66.66 ^b	66.66 ^a
75 mg/mL	100 ^a	100 ^a	100 ^a	66.66 ^b	100 ^a	66.66 ^a
T+	33.33 ^c	100 ^a	100 ^a	100 ^a	100 ^a	66.66 ^a
<i>Cassia occidentalis</i>						
T-	66.66 ^b	33.33 ^b	33.33 ^c	66.66 ^b	66.66 ^b	66.66 ^b
25 mg/mL	100 ^a	100 ^a	66.66 ^b	100 ^a	100 ^a	100 ^a
50 mg/mL	100 ^a	100 ^a	100 ^a	66.66 ^b	66.66 ^b	100 ^a
75 mg/mL	66.66 ^b	100 ^a	100 ^a	33.33 ^c	33.33 ^c	100 ^a
T+	33.33 ^c	100 ^a	100 ^a	100 ^a	100 ^a	66.66 ^b

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$.

T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

Accessions from Yagoua treated with *Azadirachta indica* and *Calotropis procera* extracts; as well as those from Moundou treated with *A. indica* and *Cassia occidentalis* extracts and accessions from Garoua (0 %). While their different negative controls showed a percentage of infection of 11.11 %. The Koumra accessions treated with *A. indica* and *C. procera* extracts, at all concentrations, showed seedlings with 0 % infection. The positive and negative controls of this accession showed identical percentages of infection (22.22 %).

3.2 Effect of the different plant extracts on the growth parameters of cashew seedlings

Table 3 presents the effects of the extracts of the three plants on the collar diameter of the seedling. Seedlings from Moundou and Ngaoundéré seeds treated with *Azadirachta indica* extracts at concentrations of 50 and 75 mg/mL and Monchamp (positive control) showed significantly identical collar diameters to each other and significantly higher than the other seedlings according to the Duncan $P \leq 0.05$ test. With seedlings from Moundou, these values were 0.80 and 0.91 cm respectively for seedlings whose seeds were treated with *A. indica* extracts at concentrations of 50 and 75 mg/mL. The positive control seedlings showed a collar diameter of 0.82 cm. For *Calotropis procera* extracts, seedlings from Gaoua seeds treated with 50

mg/mL showed a collar diameter of 1.14 cm. This collar diameter was greater than that of other seedlings from the same accession. Seedlings from the Moundou accession, with extracts from the same plant, at the concentration of 75 mg/mL, showed the largest collar diameter (0.95 cm). The smallest was obtained with seedlings whose seeds were treated with 50 mg/mL of *C. procera* extracts (0.63 cm).

With *Cassia occidentalis* extracts, the positive control seedlings showed the largest collar diameter (1.87 cm), followed by seedlings from seeds treated with 50 mg/mL (0.97 cm) and 25 mg/mL (0.79 cm). Seedlings from the Sarh and Ngaoundéré accessions at all concentrations, as well as their controls (positive and negative), showed significantly identical collar diameters. The values of the different collar diameters ranged from 0.63 cm to 0.92 cm.

All plant extracts generally resulted in seedlings with greater heights than the negative control seedlings (Table 4). Seedlings from the Moundou accessions treated with *Azadirachta indica* extracts at all concentrations showed greater heights than the positive (9 cm) and negative (6 cm) controls. These values were 11 cm, 16 cm and 15.33 cm for seedlings whose seeds were treated with *A. indica* extracts at concentrations of 25, 50 and 75 mg/ml respectively.

Table 2: Effect of plant extracts on the percentage of infection (%) of *Anacardium occidentale* L. seedlings

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	22.22 ^a	11.11 ^a	22.22 ^a	11.11 ^a	0 ^b	11.11 ^a
25 mg/mL	0 ^b	0 ^b	11.11 ^b	0 ^b	0 ^b	0 ^b
50 mg/mL	0 ^b	0 ^b	0 ^c	0 ^b	0 ^b	0 ^b
75 mg/mL	0 ^b	0 ^b	11.11 ^b	0 ^b	0 ^b	0 ^b
T+	22.22 ^a	0 ^b	22.22 ^a	0 ^b	22.22 ^a	0 ^b
<i>Calotropis procera</i>						
T-	22.22 ^a	11.11 ^b	22.22 ^a	11.11 ^a	0 ^a	11.11 ^a
25 mg/mL	0 ^b	0 ^c	0 ^c	0 ^b	0 ^a	0 ^b
50 mg/mL	0 ^b	22.22 ^a	11.11 ^b	0 ^b	0 ^a	0 ^b
75 mg/mL	0 ^b	0 ^c	11.11 ^b	0 ^b	0 ^a	0 ^b
T+	22.22 ^a	0 ^c	22.22 ^a	0 ^b	22.22 ^b	0 ^b
<i>Cassia occidentalis</i>						
T-	22.22 ^a	11.11 ^a	22.22 ^a	11.11 ^b	0 ^c	11.11 ^a
25 mg/mL	22.22 ^a	0 ^b	0 ^c	33.33 ^a	11.11 ^b	11.11 ^a
50 mg/mL	0 ^b	0 ^b	0 ^c	0 ^c	0 ^c	0 ^b
75 mg/mL	0 ^b	0 ^b	11.11 ^b	0 ^c	0 ^c	0 ^b
T+	22.22 ^a	0 ^b	22.22 ^a	0 ^c	22.22 ^a	0 ^b

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$. T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

Table 3: Effect of plant extracts on the collar diameter (cm) of *Anacardium occidentale* L.

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	0.59±0.11 ^c	0.65±0.21 ^b	0.82±0.37 ^a	0.92±0.21 ^a	0.71±0.36 ^{ab}	0.71±0.11 ^{ab}
25 mg/mL	0.84±0.17 ^b	0.67±0.16 ^b	0.84±0.21 ^a	0.85±0.18 ^{ab}	0.63±0.09 ^b	0.78±0.21 ^{ab}
50 mg/mL	0.96±0.28 ^b	0.80±0.28 ^{ab}	0.76±0.20 ^a	0.79±0.08 ^b	0.8±0.05 ^a	0.91±0.11 ^a
75 mg/mL	0.66±0.53 ^{bc}	0.91±0.12 ^a	0.70±0.43 ^a	0.81±0.07 ^b	0.82±0.16 ^a	0.69±0.08 ^b
T+	1.87±0.21 ^a	0.82±0.19 ^{ab}	0.79±0.48 ^a	0.89±0.47 ^{ab}	0.84±0.18 ^a	0.82±0.38 ^{ab}
<i>Calotropis procera</i>						
T-	0.59±0.11 ^c	0.65±0.21 ^{bc}	0.82±0.37 ^{ab}	0.92±0.21 ^b	0.71±0.36 ^a	0.71±0.11 ^b
25 mg/mL	0.81±0.13 ^b	0.74±0.15 ^{abc}	0.79±0.37 ^{ab}	0.61±0.02 ^c	0.75±0.80 ^a	0.89±0.04 ^a
50 mg/mL	0.76±0.35 ^{bc}	0.63±0.18 ^c	0.82±0.04 ^a	1.14±0.09 ^a	0.87±0.43 ^a	0.75±0.29 ^{ab}
75 mg/mL	0.85±0.20 ^b	0.95±0.12 ^a	0.45±0.19 ^b	0.76±0.18 ^{bc}	0.76±0.28 ^a	0.75±0.18 ^{ab}
T+	1.87±0.21 ^a	0.82±0.19 ^{ab}	0.79±0.48 ^{ab}	0.89±0.47 ^{ab}	0.84±0.18 ^a	0.82±0.38 ^{ab}
<i>Cassia occidentalis</i>						
T-	0.59±0.11 ^c	0.65±0.21 ^b	0.82±0.37 ^a	0.92±0.21 ^a	0.71±0.36 ^{ab}	0.71±0.11 ^{ab}
25 mg/mL	0.79±0.25 ^{bc}	0.7±0.37 ^{ab}	0.79±0.37 ^a	0.88±0.55 ^a	0.54±0.21 ^b	0.94±0.02 ^a
50 mg/mL	0.97±0.08 ^b	0.71±0.11 ^{ab}	0.91±0.32 ^a	0.79±0.92 ^a	0.71±0.38 ^{ab}	0.96±0.04 ^a
75 mg/mL	0.68±0.28 ^{bc}	0.76±0.18 ^{ab}	0.86±0.74 ^a	0.63±0.38 ^a	0.73±0.18 ^{ab}	0.78±0.16 ^{ab}
T+	1.87±0.21 ^a	0.82±0.19 ^a	0.79±0.48 ^a	0.89±0.47 ^a	0.84±0.18 ^a	0.82±0.38 ^{ab}

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$. T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

The seedlings of the Sarh and Ngaoundéré accessions from seeds treated with 50 mg/mL of *A. indica* extracts showed greater heights than those of other seedlings of the same accessions. These heights were 16.33 cm and 16 cm respectively for the seedlings of the Sarh and Ngaoundéré accessions. The seedlings of the Garoua accession, at the concentration of 75 mg/mL, showed the greatest height (16.67 cm).

With *Calotropis procera* extracts, seedlings from the Moundou and Sarh accessions, those from the 25 mg/mL treatment showed the greatest height. These heights were 18 cm and 14.33 cm respectively. On the other hand, with the Yagoua accession, the positive control seedlings showed the greatest height (16.33 cm) followed by the negative control seedlings (13.5 cm). When the seeds were treated with *Cassia occidentalis* extracts, the Moundou, Ngaoundéré and

Table 4: Effect of plant extracts on the height (cm) of *Anacardium occidentale* L. seedlings

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	6.00±1.04 ^c	9.00±2.24 ^c	6.12±0.58 ^c	10.00±2.43 ^b	7.52±1.43 ^c	13.50±2.81 ^{ab}
25 mg/mL	11.00±5.50 ^{ab}	15.00±2.66 ^{ab}	13.00±0.21 ^c	11.67±1.15 ^b	11.63±1.25 ^b	13.33±2.30 ^{ab}
50 mg/mL	16.00±1.00 ^a	16.00±1.03 ^a	16.33±1.53 ^a	11.00±2.65 ^b	16.00±1.65 ^a	13.00±1.02 ^b
75 mg/mL	15.33±2.00 ^a	17.00±0.77 ^a	14.00±0.26 ^b	16.67±2.66 ^a	10.67±0.58 ^b	15.67±2.31 ^{ab}
T+	9.00±2.24 ^b	14.00±0.77 ^b	8.33±1.93 ^d	7.50±1.41 ^c	11.27±1.51 ^b	16.33±1.81 ^a
<i>Calotropis procera</i>						
T-	6.00±1.04 ^c	9.00±2.24 ^c	6.12±0.58 ^d	10.00±2.43 ^{ab}	7.52±1.43 ^b	13.50±2.81 ^{ab}
25 mg/mL	13.33±2.52 ^{ab}	18.00±1.93 ^a	14.33±1.07 ^a	12.37±0.15 ^a	7.67±1.15 ^b	8.50±1.93 ^c
50 mg/mL	16.00±4.16 ^a	10.00±0.13 ^c	13.33±1.62 ^{ab}	9.50±2.75 ^{bc}	11.65±2.65 ^a	13.00±1.26 ^b
75 mg/mL	16.33±2.00 ^a	11.67±3.61 ^{bc}	13.00±0.51 ^b	10.39±0.41 ^b	13.07±1.18 ^a	12.00±1.53 ^b
T+	9.00±2.24 ^b	14.00±0.77 ^b	8.33±1.93 ^c	7.50±1.41 ^c	11.27±1.51 ^a	16.33±1.81 ^a
<i>Cassia occidentalis</i>						
T-	6.00±1.04 ^c	9.00±2.24 ^c	6.12±0.58 ^b	10.00±2.43 ^b	7.52±1.43 ^c	13.50±2.81 ^{ab}
25 mg/mL	15.00±1.20 ^a	12.00±2.51 ^{bc}	12.00±2.18 ^a	11.67±1.15 ^{ab}	11.17±1.15 ^b	11.5±2.51 ^b
50 mg/mL	9.67±1.00 ^b	14.67±1.93 ^b	13.67±1.53 ^a	12.75±2.05 ^a	12.05±1.65 ^b	16.00±1.20 ^a
75 mg/mL	15.00±1.53 ^a	18.00±1.73 ^a	12.00±1.69 ^a	13.67±0.43 ^a	14.23±0.69 ^a	16.67±2.36 ^a
T+	9.00±2.24 ^b	14.00±0.77 ^b	8.33±1.93 ^b	7.50±1.41 ^c	11.27±1.51 ^b	16.33±1.81 ^a

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$. T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

Table 5: Effect of plant extracts on leaf number of *Anacardium occidentale* L. seedlings

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	7.00±2.08 ^c	5.00±1.24 ^c	8.67±1.15 ^c	5.00±2.89 ^{bc}	8.00±0.62 ^b	11.33±5.51 ^{ab}
25 mg/mL	9.50±1.50 ^{bc}	12.67±1.53 ^b	9.00±2.29 ^c	4.00±2.89 ^c	8.00±2.73 ^{ab}	8.67±5.03 ^{ab}
50 mg/mL	10.67±0.15 ^b	16.00±1.07 ^a	15.00±1.06 ^a	11.33±1.53 ^a	5.00±0.21 ^c	10.50±2.02 ^{ab}
75 mg/mL	13.33±0.93 ^a	15.00±2.13 ^{ab}	14.00±0.31 ^b	8.33±1.53 ^b	7.33±0.31 ^b	12.53±1.31 ^a
T+	10.00±1.53 ^{bc}	16.13±0.82 ^a	9.00±3.20 ^c	6.00±1.53 ^{bc}	11.67±1.65 ^a	10.67±2.81 ^{ab}
<i>Calotropis procera</i>						
T-	7.00±2.08 ^b	5.00±1.24 ^c	8.67±1.15 ^b	5.00±2.89 ^c	8.00±0.62 ^b	11.33±5.51 ^{ab}
25 mg/mL	12.33±0.51 ^a	13.33±2.52 ^{ab}	11.67±2.88 ^a	9.00±0.58 ^{ab}	6.33±1.08 ^c	10.00±6.92 ^b
50 mg/mL	11.00±1.73 ^a	9.67±2.16 ^b	9.50±1.73 ^{ab}	10.67±2.31 ^a	10.00±1.04 ^a	16.00±2.50 ^a
75 mg/mL	11.67±2.08 ^a	15.00±2.00 ^a	10.00±0.12 ^a	9.50±1.29 ^{ab}	8.33±2.11 ^{ab}	9.50±1.36 ^b
T+	10.00±1.53 ^{ab}	16.13±0.82 ^a	9.00±3.20 ^{ab}	6.00±1.53 ^c	11.67±1.65 ^a	10.67±2.81 ^b
<i>Cassia occidentalis</i>						
T-	7.00±2.88 ^{ab}	5.00±1.24 ^b	8.67±1.15 ^b	5.00±2.89 ^b	8.00±0.62 ^b	11.33±5.51 ^a
25 mg/mL	9.33±2.08 ^{ab}	3.00±1.20 ^b	15.00±0.90 ^a	10.00±4.93 ^a	11.33±1.05 ^a	9.00±6.81 ^{ab}
50 mg/mL	8.00±1.46 ^{ab}	16.00±1.04 ^a	15.33±2.14 ^a	12.33±3.79 ^a	8.00±1.51 ^b	9.67±2.30 ^{ab}
75 mg/mL	7.33±1.09 ^b	16.33±1.53 ^a	8.00±3.47 ^b	13.00±2.35 ^a	8.50±0.93 ^b	10.00±1.53 ^a
T+	10.00±1.53 ^a	16.13±0.82 ^a	9.00±3.20 ^b	6.00±1.53 ^b	11.67±1.65 ^a	10.67±2.81 ^a

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$. T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

Yagoua accessions showed the greatest heights at the 75 mg/mL concentration. These heights were 18 cm, 14.23 cm and 16.67 cm respectively. With the seedlings from the Koumra accession, the greatest height was 15 cm. This height was obtained at concentrations of 25 and 75 mg/mL. Seedlings from seeds treated with extracts of all three plants had a higher number of leaves than the negative control plants (Table 5). Seedlings from the Koumra and Yagoua accessions

whose seeds were previously treated with *Azadirachta indica* extracts at a concentration of 75 mg/mL, showed 13.33 and 12.55 leaves respectively. These leaf numbers were higher than those of the other seedlings. The seedlings of the Sarh and Garoua accessions, at the concentration of 50 mg/mL with the same plant, showed a greater number of leaves (15 leaves for Sarh and 11.33 for Garoua) than the other seedlings. Seedlings from the Koumra, Sarh and Ngaoundéré

Table 6. Effect of plant extracts on vigour of *Anacardium occidentale* L. seedlings

Concentration	Koumra	Moundou	Sarh	Garoua	NGaoundéré	Yagoua
<i>Azadirachta indica</i>						
T-	10.16±0.64 ^d	13.84±0.05 ^d	15.24±0.65 ^c	6.50±0.29 ^e	15.49±0.75 ^b	18.77±0.40 ^a
25 mg/mL	19.05±0.18 ^b	23.88±0.12 ^a	21.41±0.20 ^a	19.23±0.08 ^a	11.9±0.24 ^d	16.67±0.14 ^b
50 mg/mL	15.96±0.15 ^c	18.75±1.09 ^b	11.42±0.10 ^e	16.13±0.13 ^c	13.34±0.34 ^c	17.21±0.54 ^b
75 mg/mL	20.19±0.12 ^a	19.78±0.04 ^b	18.13±0.30 ^b	17.13±0.12 ^b	18.29±0.16 ^a	12.32±0.25 ^c
T+	5.88±0.13 ^e	17.07±0.79 ^c	11.90±0.17 ^d	9.34±0.03 ^d	14.59±0.08 ^b	16.88±0.40 ^b
<i>Calotropis procera</i>						
T-	10.16±0.64 ^d	13.84±0.05 ^c	15.24±0.65 ^c	6.50±0.29 ^e	15.49±0.75 ^b	18.77±0.40 ^a
25 mg/mL	19.75±0.15 ^b	13.51±0.12 ^d	15.45±0.09 ^c	17.44±0.10 ^a	12.00±0.24 ^d	15.85±0.10 ^d
50 mg/mL	21.48±1.17 ^a	18.52±0.05 ^a	23.33±0.07 ^a	11.34±0.09 ^c	12.06±0.20 ^d	16.00±0.10 ^c
75 mg/mL	17.65±0.02 ^c	12.63±0.09 ^e	19.40±0.26 ^b	15.63±0.29 ^b	17.19±0.40 ^a	15.33±0.31 ^e
T+	5.88±0.13 ^e	17.07±0.79 ^b	11.90±0.17 ^d	9.34±0.03 ^d	14.59±0.08 ^c	16.88±0.40 ^b
<i>Cassia occidentalis</i>						
T-	10.16±0.64 ^d	13.84±0.05 ^e	15.24±0.65 ^a	6.50±0.29 ^e	15.49±0.75 ^b	18.77±0.40 ^b
25 mg/mL	12.24±0.24 ^c	20.91±0.20 ^c	12.63±0.23 ^c	11.39±0.26 ^c	22.06±0.10 ^a	17.02±0.28 ^c
50 mg/mL	16.67±0.2 ^a	25.35±0.31 ^a	15.50±0.27 ^a	15.07±0.17 ^b	15.49±0.33 ^b	18.52±0.22 ^b
75 mg/mL	13.24±0.59 ^b	22.36±0.02 ^b	13.92±0.25 ^b	20.44±0.15 ^a	21.91±0.95 ^a	20.93±0.28 ^a
T+	5.88±0.13 ^e	17.07±0.79 ^d	11.90±0.17 ^d	9.34±0.03 ^d	14.59±0.08 ^c	16.88±0.40 ^d

*Means with the same letter in the same column are not significantly different according to Duncan's $P \leq 0.05$. T- = negative control (without any supplement) and T+ = positive control (addition of Monchamp).

accessions with *Calotropis procera* extracts, at all concentrations, showed a higher number of leaves than the negative control seedlings. The number of leaves varied between 9 and 11.67 leaves.

The different plant extracts improved seedling vigour (Table 6). Seedlings from the Koumra and Ngaoundéré accessions treated with *Azadirachta indica* extract at a concentration of 75 mg/mL showed greater vigour than those from the two controls. These vigour levels were 20.19 for seedlings from Koumra and 18.29 for those from Ngaoundéré. The same observation was made for seedlings from the Moundou, Sarh and Garoua accessions when the seeds were treated with *A. indica* extracts at a concentration of 25 mg/mL. These vigours were 23.88; 21.41 and 19.23. With *Calotropis procera* and *Cassia occidentalis* extracts, seedlings from Koumra, Moundou and Sarh accessions showed significantly higher vigour than others when seeds were treated with extracts of these two plants at 25 mg/mL. These vigours ranged from 15.5 to 25.35.

With the Yagoua accession, the negative control seedlings showed a vigour of 18.77. This vigour was significantly higher than that of the seedlings treated with *A. indica* and *C. procera* extracts according to

Duncan's test at $P \leq 0.05$. For seedlings from seeds treated with *Cassia occidentalis* extracts, seeds from Cameroon at the concentration of 75 mg/ml showed the highest vigour (these vigours were 20.44, 21.91 and 20.93 respectively for Garoua, Ngaoundéré and Yagoua).

4. Discussion

4.1 Evaluation of the efficacy of the plant extracts on cashew seed germination and seedling infection percentage

The results of the present study showed that the extracts of the three plants had effects in improving the percentage of seed germination and in reducing fungal infections of seedlings. All plant extracts resulted in higher germination percentages than the negative controls with seeds of all accessions. These results could be explained by the fact that the different accessions would contain compounds within them that would promote germination. Similar results were reported by [33], who showed that aqueous extracts of *Cymbopogon citratus* improved the germination percentage of maize seeds; as well as those of [34], who revealed that extracts of *Carica papaya* resulted in high germination percentages with common bean and cowpea seeds. *Azadirachta indica* extracts allowed 100 % germination with seeds from accessions in Garoua, Koumra, Moundou, Ngaoundéré, Sarh and

Yagoua. These results corroborate with those of [28], who showed that aqueous extracts of *A. indica* would improve the germination percentage of cotton seeds. Furthermore, the different plant extracts induced a significant decrease in fungal infection of cashew seedlings. The reduction in fungal infection of the seedlings is thought to be due to the fact that when the seeds were treated with the plant extracts, the compounds with antifungal properties present in the plant extracts inhibited or destroyed the fungi associated with the seeds that could be transmitted to the seedlings. These results are in agreement with several studies that have shown that plant extracts inhibit the development of seed-associated fungi and reduce infections in seedlings. This is the case with the work of [35] where aqueous extracts of *Carica papaya* enhance seedling emergence from African yam bean seeds and reduce mycelial growth of fungi such as *Aspergillus niger*, *Aspergillus flavus*, *Botryodiplodia theobromae* and *Fusarium moniliforme* associated with African yam bean seeds (*Sphenostylis stenocarpa*). Similarly, the work of [36, 37] showed that extracts of several plants inhibited the development of fungi associated with *Sorghum bicolor* and *Arachis hypogaea* seeds.

4.2 Evaluation of the efficacy of plant extracts on cashew seedling growth parameters

The different aqueous extracts of the three plants improved the growth parameters of the seedlings such as crown diameter, height, number of leaves and vigour. The extracts of *Azadirachta indica* and *Calotropis procera* resulted in high leaf number, large crown diameters and heights; and good vigour in the seedlings. These results are believed to be due to the fact that extracts of these two plants contain substances with growth and development stimulating properties. These results are in agreement with the work of [38], who showed that aqueous extracts of *Ageratum conizoides*, *Chromolaena odorata* and *Pteridium aquilinum* improved the number of leaves, crown diameter, height and vigour of common bean seedlings. Treatment of Assamela seeds with aqueous extracts of *Ageratum conizoides*, *Citrus sinensis* and *Carica papaya* have a positive effect on seedling emergence, vigour and height [39]. Similarly, those of [28], showed that treatment of cotton seeds with aqueous extracts of *Azadirachta indica* leaves and *Boswellia dalzielii* bark prior to sowing, improved the

hootability and vigour index of cotton seedlings compared to those treated with distilled water. Those of showed that, seed treatment of a common bean variety (Faba bean) with aqueous extracts of *Asclepias sinaica* improved seedling vigour index.

5. Conclusions

This study revealed that seed treatment of *Anacardium occidentale* L. with aqueous extracts of *Azadirachta indica*, *Calotropis procera* and *Cassia occidentalis* resulted in high germination percentages, less infected seedlings with significant leaf number, collar diameter and seedling vigour. The extracts of *A. indica* and *C. procera* at tested concentrations resulted in 100 % germination of cashew seeds from Koumra and Garoua accessions; as well as uninfected seedlings. This suggest the use of these plant extracts to reduce cashew seed infections, therefore improving seed germination and seedling vigour. However, further studies on field trials with these extracts are still going before recommendation.

Authors' contributions

This work was carried out in collaboration among all authors. Prof. Tsopmbeng designed the study, wrote the protocol and reviewed all drafts of the manuscript. Dr. Keuete ran the data analysis, wrote the first draft of the manuscript. Dr Sali managed the literature searches and improved the first draft. Mr Bemadjita managed the experimental set up. All authors read and approved the final manuscript.

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Availability of data and materials

All data will be made available on request according to the journal policy

Conflicts of interest

Authors have declared that there is no potential conflict of interests exist.

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