**The anthelmintic effect of ginger *(Zingiber officinale*) powder on the biochemical and haematological parameters of pigs experimentally infected with some gastrointestinal nematodes.**

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**Abstract**

 The effective use of plant derivatives as anthelmintics is often overlooked due to their toxicity. Hence the objective of this work was to evaluate the toxicity of ginger powder used as an anthelmintic in pigs. A total of 24 pigs were randomly allocated to four treatment groups and infected with 2650L3 mixed nematode larvae. The negative control (T0-) was not treated. The positive control (T0+) was treated with Mebendazole. T1 and T2 groups were treated with 12.5g/kg and 25g/kg of ginger crude powder respectively. The animals were followed for 4 weeks during which fecal and blood samples were collected for coprological and haemato-biochemical analyses. T2 group had the highest faecal egg count reduction **(**FECR) of 92.3% followed by T1 and T0+ groups with a FECR of 86.4% and 58.5% respectively. T0- animals recorded the highest levels of creatinine, ALAT, ASAT, Total cholesterol and albumin compared to treated animals. T2 group recorded the lowest levels of urea, total protein, albumin, ALAT, ASAT, Total cholesterol and globulin and all the values were within the normal range. Similarly, T1 group obtained normal values of cholesterol, ASAT and ALAT. Blood parameters were not affected by treatments, apart from platelets (359.50 x103/ μL) which were significantly (p≤0.05) higher in T2 group. In conclusion, half (12.5g/kg) and full (25g/kg) dose of ginger powder were not toxic but rather improved the renal and hepatic functions. Therefore, treatment with 25g/kg of ginger powder can be recommended for effective and safe use in the treatment of gastrointestinal nematodes in pigs.

**Key words:** Blood parameters,Gastrointestinal nematodes, Ginger powder, Pigs.

**1. Introduction**

Parasitism in domestic pigs most especially due to gastrointestinal nematodes has been reported to be the most common and important disease in tropical and subtropical countries (Nganga *et al,* 2008). After African swine fever and erysipelas gastrointestinal parasites are responsible for the substantial losses in the productivity of swine and other livestock industry (Boes *et al.,* 2000*;* Joachim *et al.,* 2001), since the infections result in reduced weight gains, decreased litter sizes, poor growth rates, visceral organ condemnation at slaughter and deaths (Stewart and Hoyt, 2006). Parasitism is crucial in livestock production but often overlooked due to the fact that clinical signs are not often obvious or acute.

 For the past years chemotherapeutic drugs such as ivermectin, levamisole, albendazole or piperazine among others have been widely used in controlling parasites. However, the over use and mismanagement of these drugs has led to several limitations. Firstly, is the rapid development of anthelmintic resistance of some parasitic strains to most anthelmintics especially those belonging to the strongyloidae family (Cheng *et al.,* 2014*;* Brunet *et al.,* 2008*;* Hoste *et al.,* 2006) Secondly, there is an increased concern of consumers over drug residues in meat and milk products, and a potential risk for environmental contamination (Barrau *et al.,* 2005). Also, most of these anthelmintics are usually unaffordable and unavailable to most farmers especially in the remote areas. It is therefore necessary to seek alternative means against gastrointestinal parasites which are readily available, affordable and environmentally friendly. These include substances produced from plants, since plants have the advantage of sustainable supply and are ecologically friendly (Al-Shaibani et al., 2009; Bachaya et al., 2009; Deeba et al., 2009; Sindhu et al., 2010).

 One of the most commonly used plants which have been tested for their anthelmintic properties is ginger (*Zingiber officinale).* Some in vitro studies (Lin et al., 2010; Moazeni and Naser, 2011) as well as in vivo (Iqbal et al., 2006; Matthews et al., 2016; Mostafa et al., 2012; Kiambom et al., 2020) studies have been carried out to proof the anthelmintic effects of *Z*. *officinale* rhizome on various parasite species. These studies demonstrated the anthelmintic effect of this plant on gastrointestinal parasites but the toxicity of this plant has not yet been investigated. The toxicity can be evaluated by assessing the haematological parameters as well as the biochemical parameters relevant to kidney and hepatic functions. Indeed, any difference in the values of these parameters when compared with the normal values could be used to assess the toxicity of a plant, and thus to interpret the health status of treated animals (Omidi et al., 2018). Therefore, the main objective of this study was to evaluate the anthelmintic effect of *Zingiber officinale* powder on the haematological and biochemical parameters of infected pigs.

**2. MATERIAL AND METHODS**

**2.1. Study Area**

 The experiment was carried out at the teaching and research farm of the University of Dschang which is located between the latitudes 05°22’58” and 05°30’40” N and longitudes 9°58’55” and 10°7’23” E. Dschang has an average altitude of 1420m and experiences the rainy season from mid-march to mid-November and the dry season from mid-November to mid-march. Precipitations vary between 1500 and 2000 mm/year and the temperatures vary between 14 and 25°C. Dschang has an average relative humility of 76, 8% (Pamo et *al*., 2005).

**2.2: Plant material**

 The plant material was made of the ginger rhizome (Z*ingiber officinale)* harvested from the Santa sub division in the North West region of Cameroon. The rhizomes of this plant were bought directly from a farmer and then were washed, cleaned and air-dried under a shade for at least 2 weeks. The dry product was then blended with an electric blender to obtain the crude powder.

**2.3 Animal material**

 Animals used for this experiment were 24 cross breed pigs, two months old and having an average weight of 20kg. These pigs were purchased from a single local farm. They were hosted in a raised floor piggery built with hard wood. The piggery had four different compartments of 6m² (2m×3m) each corresponding to the four different treatments. Plank feeders with a 50-litre capacity were constructed and placed in each compartment. Well-designed tire rings with a 30-litres capacity were placed in each compartment to serve as water through.

In addition to the proper hygiene and sanitation that was carried out before and during the experiment, vaccination against erysipelas was provided by officials of the Ministry of livestock and fisheries (MINEPIA) in Dschang. Also antiboitics such as combikel and penstrip as well as multivitamins such as stress-vita were given to prevent interference with other diseases.

**2.4. Culture of nematode parasitic larvae**

 Before the experiment proper was conducted, a pre-survey was carried out, whereby pig farms in the Dschang locality were visited and fresh faeces was collected and analysed using the simple flotation technique in order to identify and quantify the most prevalent association of parasites which were *Strongyloides ransomi* and *Strongyle* parasites. Then a faecal culture was performed to obtain infective larval stages as described by Soulsby (1982).

Briefly, the culture was done by placing 5grams of positive faecal samples of faeces in Petri dishes in layers of 2mm depth. The dishes had loose covers that did not prevent air circulation but deterred flies and reduced desiccation. There were two different batches of the same sample. The first batch was incubated at a temperature of about 27ºC for 48hrs to collect *Strongyloides ransomi* and *Trichostrongylus axei* nematodes and the second batch was incubated for 11 days more to obtain *Hyostrongylus rubidus* and *Globocephalus urosubulatus.*

After incubation, the larvae were collected using the Baermann technique and identified under the microscope with the help of keys (Soulsby, 1982: Van et al., 2013). The different L3 larvae were identified to determine the composition of the species involved in the mixed infection which were *Strongyloides ransomi, Hyostrongylus rubidus, Trichostrongylus axei* and *Globocephalus urosubulatus.*

**2.5. Experimental design**

 Before the experiment, all the pigs were treated against gastrointestinal parasitism using Mebendazole 5mg/kg. The pigs were randomly divided into four comparable groups of 6pigs each (3males, 3 females) and housed per group of treatment. Males and females of each treatment were housed separately. The pigs were fed commercially compounded dry feed supplied by one of the major animal feed manufacturer of the country (*Société des provenderies du Cameroun* -SPC) and given tap water to drink ad libitum. The feed was given to the pigs at equal quantities (3kgs per pen) and intervals (7am and 6pm). The pigs were given an adaptation period of one week before inducing the treatments.

Group 1 which is the negative control (T0-) was infected with 2650L3 larvae and was not treated. Group 2 which is the positive control (T0+) was infected with 2650L3 larvae and treated with Mebendazole. Group 3(T1) and Group 4(T2) were each infected with the same number of larvae (2650 L3) and treated respectively with 250g (12.5g/kg) and 500g(25g/kg) of ginger crude powder. Six weeks after the second inoculation, faecal samples were collected directly from the rectum of all the pigs to determine the presence of eggs and the faecal egg count. Faecal culture was performed again to verify whether the species involved in the infection were effectively those that were used to infest the pigs at the beginning. When all the pigs were confirmed of shedding at least 200 egg per gram(epg), then various treatments were administered. The ginger powder was mixed with pig feedstuff. After administration of various treatments, faecal samples were collected twice a week for 3 months to evaluate the evolution of egg shed.

**2.6 Faecal egg count reduction test (FECRT)**

 Plant efficacy was evaluated using the fecal egg count reduction test by the following formula:

FECR = [{EPG (pre-treatment) – EPG (14-day post-treatment)} /EPG (pre-treatment] X100 (Roepstorff and Nansen, 1998).

**2.7. Haematological parameters**

An amount of 5mls of blood was collected one week prior to treatment, 7days, 14 days and 21days post treatment from the marginal ear vein of pigs into ethylene diamine tetracetic acid (EDTA) tubes. The tubes were coded and gently rotated to ensure proper mixing of the blood with the anticoagulant without damaging the integrity of the cells and transported to the laboratory of the district hospital in Dschang whereby the haematological parameters were determined using an automatic blood analyser (KT-6180).

**2.8 Biochemical parameters**

In other to assess the effect of the plant treatment on the biochemical parameters linked to toxicity, the same amount of blood was collected in similar conditions as mentioned above. The tubes were coded and stored at -20°C before analysis. The samples were analysed at the Laboratory of Animal Health and Physiology at the University of Dschang. The total serum proteins, Serum albumin, serum globulin, Alanine aminotransferase (ALAT), Aspartate aminotransferase (ASAT), urea, creatinine and total cholesterol was analysed and recorded using the Chronolab kit (2013-03) from Barcelona Spain.

**2.9 Statistical analysis**

 The data collected was entered into Microsoft Excel and exported to SPSS version 20.0 for statistical analysis. All data collected for the different haematological and biochemical parameters were analysed using one-way ANOVA with significance level of 5%. Results were expressed as mean±standard deviation. Significant differences between the means of the different treatments were separated using the Duncan test.

**3. RESULTS**

**3.1 Plant efficacy.**

The anthelmintic efficacy of *Zingiber officinale* on gastrointestinal nematodes is shown in table 1. Irrespective of sex, treatment T2 showed the highest FECR of 92.3% followed by treatments T1 and T0+ with a FECR of 86.4% and 58.5% respectively.

**Table 1:** Faecal egg count reduction (FECR) in pigs infected with gastrointestinal nematodes.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Treatment | Pig sex | *Strongyloides ransomi*Mean EPG±SD | Strongylid eggsMean EPG±SD | TOTALMean EPG±SD |
|  |  | **pre- treatment** | **Post- treatment** | **FECR**  | **pre-treatment** | **post -treatment** | **FECR** | **pre-treatment** |  **post -treatment** | **FECR** |
| T0+ | ♂ | 2378 ± 236.76 | 1025 ± 1097.6 | 56.9% | 4785 ± 202.97  | 2058 ± 2760.18 | 57% | 3582±219.87 | 1542±1928.89 | 56.95% |
| ♀ | 2128 ± 243.94  | 235 ± 27.83  | 89% | 4245 ± 594.30  | 2301 ± 11.54  | 45.8% | 3187±419.12 | 1268±19.69 | 67.4% |
| ♂♀ | 2253 ± 254.90 | 630 ± 818.18  | 72% | 4515 ± 495.21  | 2180 ± 1993.30  | 51.7% | 3384±375.06 | 1405±15.74 | 58.5% |
|  T1 | ♂ | 2390 ± 126.78  | 268 ± 85.44  | 88% | 4963 ± 137.50  | 420 ± 95 .01 | 91.5% | 3677±131.90 | 344±90.23 | 89.75% |
| ♀ | 2522 ± 120.86  | 545 ± 149.77  | 78.4% | 4815 ± 786.19  |  621 ±100.66 | 87.1% | 3669±453.53 | 623±125.22 | 82.75% |
| ♂♀ | 2456 ± 132.18  | 407 ± 186.69  | 83.4% | 4889 ± 511.27  | 521 ± 140.94  | 89.4% | 3673±321.73 | 464±163.82 | 86.4% |
| T2 | ♂ | 2433 ± 72.85  | 207 ± 36.17  | 91.5% | 5113 ± 105.03  | 475 ± 115 .00 | 90.7% | 3783±88.94 | 341±75.59 | 91.1% |
| ♀ | 2312 ± 269.45  | 143 ± 67.14  | 93.8% | 5037 ± 308.59  | 333 ± 50.08  | 93.4% | 3675±289.02 | 238±52.08 | 93.6% |
| ♂♀ | 2373 ± 188.69  | 175 ± 59.41  | 92.6% | 5075 ± 210.40  | 404 ± 110.96 | 92.0% | 3724±199.55 | 290±85.19 | 92.3% |

**T0+:** positive control (animals treated with Mebendazole),**T1:** animals treated with a dose of 12.5g/kg of ginger powder, **T2:** animals treated with a dose 25g/kg of ginger powder.

**3.2. Effects of ginger powder on the biochemical parameters of infected and treated pigs*.***

**3.2.1 Effects on renal function.**

The effects of ginger powder on the biochemical markers of the renal function in pigs infected with gastrointestinal nematodes, and submitted to different anthelmintic treatment are shown in Table 2.

**Table 2: Effects of ginger powder used as anthelmintics on the biochemical markers of the renal function in infected pigs**

|  |  |  |  |
| --- | --- | --- | --- |
| **Biochemical markers** | **Treatment duration (weeks)** | **Treatments** | **p- value** |
| **T0**-**(n = 6)** | **T0**+**(n = 6)** | **T1****(n = 6)** | **T2****(n = 6)** |
| **Creatinin (mg/dl)** | wk1 pre | 1.22±0.14 | 1.18±0.38 | 1.16±0.12 | 1.18±0.26 | 0.167 |
| Wk1 | 1.25±0.33 | 1.21±0.27 | 1.17±0.29 | 1.19±0.15 | 0.443 |
| Wk2 | 1.36±0.30 | 1.24±0.21 | 1.20±0.24 | 1.26±0.20 | 0.336 |
| Wk3 | 1.66±0.33a | 1.30±0.18ab | 1.27±0.25b | 1.36±0.20ab | 0.049\* |
| **Urea (mg/dl)** | wk1 pre | 32.49±2.52ab | 30.00±2.54b | 36.04±3.64a | 29.28±2.35b | 0.009\* |
| Wk1 | 25.58±2.02 | 27.70±2.47 | 26.71±2.36 | 25.60±3.28 | 0.519 |
| Wk2 | 27.96±2.48ab | 30.19±2.12a | 28.20±2.48ab | 26.45±3.15b | 0.048\* |
| Wk3 | 35.86±3.48 | 36.57±3.40 | 36.46±4.31 | 35.72±3.90 | 0.785 |
| **Total protein (g/dl)** | wk1 pre | 5.35±0.88b | 6.04±0.48ab | 6.75±1.08a | 5.26±0.32b | 0.033\* |
| Wk1 | 5.24±0.29 | 5.65±0.76 | 5.55±0.30 | 5.23±0.33 | 0.419 |
| Wk2 | 5.05±0.67 | 5.20±0.94 | 5.66±0.49 | 5.03±0.82 | 0.214 |
| Wk3 | 5.21±0.50 | 5.67±0.38 | 5.47±0.54 | 5.09±0.52 | 0.191 |
| **Albumin (g/dl)** | wk1 pre | 3.42±0.53 | 3.40±0.26 | 3.40±0.63 | 3.31±0.79 | 0.910 |
| Wk1 | 3.83±0.35 | 3.55±0.28 | 3.49±0.56 | 3.33±0.66 | 0.559 |
| Wk2 | 3.58±0.54 | 3.33±0.96 | 3.35±0.30 | 3.32±0.44 | 0.430 |
| Wk3 | 3.83±0.25 | 3.61±0.28 | 3.67±0.63 | 3.30±0.39 | 0.470 |
| **Globulin (g/dl)** | wk1 pre | 1.93±0.34b | 2.64±0.52ab | 3.35±0.98a | 1.92±0.76b | 0.008\* |
| Wk1 | 1.41±0.52 | 2.09±0.60 | 2.06±0.66 | 1.13±0.27 | 0.295 |
| Wk2 | 1.72±0.42 | 1.87±0.58 | 2.31±0.37 | 2.18±0.38 | 0.228 |
| Wk3 | 2.18±0.60 | 2.06±0.17 | 1.80±0.52 | 1.79±0.15 | 0.573 |

 TO- (negative control; infected but no treatment), TO+(positive control ; infected and treated with mebendazole), T1 (infected and treated with 250g(12.5g/kg) of ginger powder), T2 (infected and treated with 500g(25g/kg)of ginger powder), Wk1 pre: week before treatment.. n: number of samples; a, b, c on the same line, values affected with the same letter do not differ significantly (p>0.05); \* significant difference (p<0.05) mg/dl; milligram per decilitre, g/dl; gram per decilitre.

At week 3 post treatment, there was a significant difference (p≤0.05) recorded amongst treatments on the level of creatinine with the negative control (T0-) recording the highest level (1.66mg/dl) of creatinine compared to T1 with the lowest level (1.27mg/dl). All treatments progressively recorded increasing levels of creatinine as time progressed. At week 2 post treatment, the level of urea with the treatment with 25g/kg of ginger (T2) was significantly lower (26.45mg/dl) compared to the positive control (T0+) recording the highest value (30.19mg/dl). Meanwhile no significant difference was seen amongst treatment on the level of albumin for all periods. The negative control (T0-) recorded the highest levels of creatinine and albumin as time evolved compared to treated animals. Animals who were infected and treated with 25g/kg of ginger (T2) recorded the smallest levels of urea, total protein, albumin and globulin throughout the study.

**3.2.2 Effects on hepatic function**

Table 3 displays the data on the effects of ginger powder on the biochemical markers of the hepatic function in pigs infected with gastrointestinal nematodes, and submitted to different anthelmintic treatment.

**Table 3: Effects of ginger powder used as anthelmintics on the biochemical markers of the hepatic function in infected pigs**

|  |  |  |  |
| --- | --- | --- | --- |
| Biochemical markers | Treatment duration (weeks) | Treatments | p-value |
| **T0**-**(n = 6)** | **T0**+**(n = 6)** | **T1****(n = 6)** | **T2****(n = 6)** |
| ALAT (U/L) | wk1 pre | 78.25±13.00 | 73.50±13.42 | 71.23±12.75 | 70.66±13.27 | 0.429 |
| Wk1 | 103.47±13.69a | 66.50±17.43b | 58.98±18.62b | 58.19±10.71b | 0.003\* |
| Wk2 | 59.50±11.33 | 44.10±11.27 | 42.00±12.33 | 40.31±10.81 | 0.172 |
| Wk3 | 81.53±13.65 | 77.34±21.32 | 77.22±18.91 | 72.72±12.11 | 0.558 |
| ASAT (U/L) | wk1 pre | 142.33±27.28 | 136.66±33.41 | 107.10±33.68 | 94.85±17.57 | 0.182 |
| Wk1 | 110.78±32.54 | 105.22±20.31 | 88.81±18.17 | 80.15±12.86 | 0.177 |
| Wk2 | 86.41±18.81 | 77.00±16.95 | 78.44±18.76 | 76.22±19.53 | 0.570 |
| Wk3 | 98.25±12.40 | 92.75±17.98 | 95.16±18.77 | 80.50±26.66 | 0.611 |
| Total cholesterol (mg/dl) | wk1 pre | 129.67±14.09 | 126.94±17.84 | 117.19±15.96 | 116.16±13.07 | 0.227 |
| Wk1 | 124.55±15.73a | 107.11±12.14b | 97.45±8.54b | 96.84±11.03b | 0.014\* |
| Wk2 | 101.65±16.60 | 95.21±17.77 | 92.23±13.13 | 83.97±6.73 | 0.299 |
| Wk3 | 107.44±17.29 | 98.35±16.17 | 98.64±14.31 | 95.04±6.89 | 0.618 |

TO- (negative control; infected but no treatment), TO+(positive control; infected and treated with mebendazole), T1 (infected and treated with 250g(12.5g/kg) of ginger powder), T2 (infected and treated with 500g(25g/kg) of ginger powder), Wk1 pre: week before treatment. n: number of samples; a, b, c on the same line, values affected with the same letter do not differ significantly (p>0.05); \* significant difference (p<0.05), U/L; unit per litre, mg/dl; milligram per decilitre.

At week 1 post treatment, the level of Alanine aminotransferase (ALAT) and Total cholesterol of the negative control group (T0-) was significantly (p≤0.05) lower compared to the treated groups with T2 recording the smallest value for both parameters. Animals who were infected and received no treatment (T0-) recorded the highest levels of ALAT, Aspartate aminotransferase (ASAT) and Total cholesterol at all periods compared to animals that were treated. Animals treated with 25g/kg of ginger (T2) recorded the smallest levels of ALAT, ASAT and Total cholesterol at all periods.

**3.3. Effects of ginger powder on the haematological parameters of infected and treated pigs.**

**3.3.1 Effects on the white blood cells**

The effects of ginger powder on the white blood cells of pigs infected with gastro intestinal nematodes, and submitted to different anthelmintic treatments are presented in Table 4.

**Table 4: Effects of ginger powder used as anthelmintics on the white blood cells in infected pigs**

|  |  |  |  |
| --- | --- | --- | --- |
| **Blood parameters** | **Treatment duration (weeks)** | **Treatments** | **p- value** |
| **T0**-**(n = 6)** | **T0**+**(n = 6)** | **T1****(n = 6)** | **T2****(n = 6)** |
| **Total white blood cells (x103/ μl)** | wk1 pre | 19.23±2.39 | 16.55±1.63 | 14.25±2.37 | 16.68±2.58 | 0.251 |
| Wk1 | 16.35±1.54 | 14.74±3.31 | 16.23±1.47 | 16.30±4.07 | 0.561 |
| Wk2 | 16.28±4.66 | 15.47±2.88 | 16.18±2.26 | 16.17±3.32 | 0.750 |
| Wk3 | 15.48±2.20 | 14.24±2.09 | 15.23±1.87 | 15.26±2.60 | 0.653 |
| **Lymphocytes (x103/ μl)** | wk1 pre | 16.74±2.43 | 14.33±1.64 | 11.27±1.96 | 12.23±1.92 | 0.234 |
| Wk1 | 12.83±2.78 | 12.73±2.87 | 11.88±0.96 | 11.90±1.88 | 0.753 |
| Wk2 | 12.98±3.91 | 12.40±2.24 | 11.25±2.07 | 12.27±0.86 | 0.846 |
| Wk3 | 11.98±2.33 | 11.94±1.53 | 11.43±1.40 | 11.74±2.26 | 0.969 |
| **Monocytes (x103/ μl)** | wk1 pre | 1.62±0.20 | 1.77±0.28 | 1.56±0.53 | 1.89±0.79 | 0.713 |
| Wk1 | 1.66±0.55ab | 1.22±0.23b | 2.12±0.47a | 2.12±0.60a | 0.022\* |
| Wk2 | 1.14±0.57 | 1.18±0.46 | 1.63±0.43 | 1.47±0.60 | 0.342 |
| Wk3 | 1.44±0.50 | 1.30±0.22 | 1.53±0.27 | 1.28±0.30 | 0.569 |
| **Granulocytes (x103/μl)** | wk1 pre | 0.87±0.30 | 0.46±0.09 | 1.42±0.57 | 2.56±1.09 | 0.523 |
| Wk1 | 2.66±0.93a | 0.79±0.22b | 2.23±0.67a | 2.08±0.63a | 0.002\* |
| Wk2 | 2.06±0.87 | 1.88±0.51 | 3.30±0.98 | 3.43±0.79 | 0.276 |
| Wk3 | 2.14±0.72a | 1.00±0.28b | 2.77±0.73a | 2.74±0.40a | 0.001\* |

TO- (negative control; infected but no treatment), TO+(positive control ; infected and treated with mebendazole), T1 (infected and treated with 250g(12.5g/kg) of ginger powder), T2 (infected and treated with 500g(25g/kg) of ginger powder), Wk1 pre: week before treatment.. n: number of samples; a, b, c on the same line, values affected with the same letter do not differ significantly (p>0.05); \* significant difference (p<0.05), μL; microliter

One week after treatment, T2 group significantly recorded the highest count (2.12(103/μL)) of monocytes compared to the positive control (T0+) recording the lowest count (1.22(103/μL)). The granulocytes count was significantly (p≤0.05) higher in the negative control (T0-) (2.66(103/ μL)) than in the positive control (T0+) (0.79(103/ μL)). At week 3 post treatment, T1 significantly (p≤0.05) recorded higher count (2.77(103/ μL) of granulocytes compared to the positive control
(T0+) recording the lowest count (1.00(103/ μL)) of all the groups.

**3.3.2 Effects of ginger powder on the red blood cells, haemoglobin and platelets in infected and treated pigs.**

These effects are displayed in Table 5.

**Table 5: Effects of ginger powder used as anthelmintics on the red blood cells, haemoglobin and platelets in infected pigs**

|  |  |  |  |
| --- | --- | --- | --- |
| **Blood parameters** | **Treatment duration (weeks)** | **Treatments** | **p- value** |
| **T0**-**(n = 6)** | **T0**+**(n = 6)** | **T1****(n = 6)** | **T2****(n = 6)** |
| **Red blood cells(x106/μl)** | wk1 pre | 9.07±0.85 | 8.58±0.58 | 8.10±0.72 | 8.09±0.45 | 0.148 |
| Wk1 | 8.30±0.37 | 8.22±0.55 | 7.62±0.75 | 7.62±0.62 | 0.198 |
| Wk2 | 8.23±0.51 | 8.13±0.32 | 7.59±0.74 | 7.53±0.57 | 0.229 |
| Wk3 | 7.49±0.61 | 8.30±0.53 | 7.72±0.76 | 7.68±0.68 | 0.806 |
| **Haematocrit (%)** | wk1 pre | 48.68±4.52 | 48.67±4.43 | 46.73±5.04 | 42.03±2.74 | 0.144 |
| Wk1 | 45.58±2.25 | 45.04±3.02 | 42.95±2.88 | 41.58±2.67 | 0.194 |
| Wk2 | 43.56±3.45 | 43.55±2.22 | 42.10±1.08 | 41.37±1.93 | 0.324 |
| Wk3 | 49.06±3.48 | 44.22±1.48 | 41.68±0.83 | 40.26±2.94 | 0.607 |
| **Haemoglobin(g/dl)** | wk1 pre | 14.45±1.36 | 14.18±0.44 | 14.00±0.93 | 13.40±0.54 | 0.239 |
| Wk1 | 13.75±0.82 | 13.96±0.99 | 13.15±0.93 | 12.68±0.95 | 0.142 |
| Wk2 | 13.64±1.00 | 13.83±0.75 | 13.18±0.31 | 13.00±0.63 | 0.180 |
| Wk3 | 12.96±0.25 | 14.22±0.31 | 13.30±0.37 | 12.86±0.76 | 0.598 |
| **Mean cell volume (FL)** | wk1 pre | 53.60±1.33 | 56.75±3.96 | 57.93±6.08 | 51.98±2.40 | 0.400 |
| Wk1 | 53.68±1.06 | 54.84±2.88 | 54.65±3.89 | 54.25±2.26 | 0.374 |
| Wk2 | 55.66±1.33 | 53.62±3.14 | 53.85±4.67 | 52.37±2.30 | 0.341 |
| Wk3 | 55.14±0.80 | 53.86±2.83 | 53.07±4.59 | 52.04±2.51 | 0.416 |
| **Platelets (x103/ μL)** | wk1 pre | 259.25±79.07 | 305.83±52.46 | 246.50±83.54 | 315.00±75.61 | 0.578 |
| Wk1 | 261.75±90.77ab | 203.00±83.49b | 204.00±82.33b | 359.50±76.59a | 0.044 |
| Wk2 | 292.60±86.11 | 289.00±94.54 | 235.83±76.04 | 385.50±72.81 | 0.804 |
| Wk3 | 319.80±88.41 | 272.00±69.47 | 243.50±44.49 | 344.60±49.72 | 0.561 |

TO- (negative control; infected but no treatment), TO+(positive control ; infected and treated with mebendazole), T1 (infected and treated with 250g(12.5g/kg) of ginger powder), T2 (infected and treated with 500g(25g/kg) of ginger powder), Wk1 pre: week before treatment.. n: number of samples; a, b, c on the same line, values affected with the same letter do not differ significantly (p>0.05); \* significant difference (p<0.05), μL; microliter, g/dl; gram per decilitre, FL; femtoliters

One-week post treatment, T2 group significantly (p≤0.05) recorded the highest count of platelets (359.50 (103/ul) compared to the other treated groups with the positive control (T0+) recording the lowest count (203.00(103/ul).

**4. DISCUSSION**

The anthelmintic efficacy of *Zingiber officinale* on gastrointestinal nematodes showed that irrespective of sex, treatment T2 had the highest FECR of 92.3% followed by treatments T1 and T0+ with a FECR of 86.4% and 58.5% respectively. The World Association for the Advancement of Veterinary Parasitology guideline (Coles et al., 1992), states that a FECR >90% is considered effective while a FECR between 80%-90% is considered equivocal and needs to be repeated for confirmation and a FECR <80% means that the worms are resistant to the tested anthelmintic. Therefore, treatment with 25g/kg of ginger (T2) was effective in the treatment of the studied nematodes meanwhile these same parasites were resistant to treatment with mebendazole (FECR= 58.5%). This tendency is in conformity with a previous study on *Strongyloides ransomi* and strongyles (Kiambom et al., 2020) showing that ginger powder treatment at a dose 25g/kg was more effective than half dose (12.5g/kg), followed by mebendazole treatment. The intermediary value (86.4%) of FECR for the dose of 12.5g/kg (T1) showed that this dose is not appropriate for the treatment of gastrointestinal nematodes in pigs. Thus the treatment with 25g/kg of ginger (T2) is as efficient on a wide range of gastrointestinal nematodes as it is on individual groups of gastrointestinal nematodes. However, the findings by Iqbal et al., (2006) differs as they reported ginger efficacy (FECR=66.6%) to be less effective than the positive control levamisole (FECR=99.2). Also Aurora et al., (2006) contrasts the results of this study as he reported ginger to be less effective (FECR=87%) than mebendazole (FECR=100%) against *Ascaris suum*. These discrepancies in results could be attributed to the differences in parasite species involved as some parasites could be more susceptible than others. Also the differences in the treatment doses and ginger form (extract vs powder) could be the reason of the contrast in results. In fact, Aurora et al., (2006) and Iqbal et al., (2006) used aqueous ginger extract for treatment which differs from our study where ginger crude powder was used.

In the present study, serum creatinine, albumin, and urea were analysed to assess the effect of ginger powder on the renal glomerular function. At week 3 post treatment, there was a significant difference (p≤0.05) recorded amongst treatments on the level of creatinine with the negative control (T0-) recording the highest level of creatinine compared to T1 with the lowest level. At week 2 post treatment, the urea level with the treatment with 25g/kg of ginger (T2) was significantly lower compared to the positive control (T0+) recording the highest level. This shows that administration of 25g/kg (T2) and 12.5g/kg (T1) of ginger crude powder significantly improved the renal function by decreasing the levels of urea and creatinine respectively. This decrease in the levels of urea and creatinine is due to the presence of polyphenols and flavonoids such as gingerols, shogoals, gingerdiol, gingerdione and some phenolic ketone derivatives which possess antioxidant and nephroprotective properties that exert a positive effect on the levels of renal function markers (Hamed et al., 2012) thus proving the safe use of this plant on kidneys. The high values of creatinine in the untreated (T0-) animals indicated that there was a deficiency in the glomerular filtration rate by the kidneys hence a decrease in the kidney function. This data matched those of Ajith et al., (2007) who reported that the presence of polyphenols and flavonoids in ginger extract might be responsible for the antioxidant nephroprotective activities and the reduction of serum urea and creatinine levels. Creatinine is an organic base formed during muscle protein metabolism as a degradation product of creatine phosphate. Like many other organic bases, creatinine is filtered at the glomerulus and eliminated from plasma by the kidney. Therefore, ginger improves the glomerular filtration rate. However, this result contrasts that of Dias et al. (2006) who reported that serum creatinine levels were not modified by 1% ginger extract treatment. The discrepancies in these results may be due to differences in treatment doses used, form of plant (powder, extract, essential oils).

Animals that were infected and received no treatment (T0-) recorded the highest levels of creatinine, ALAT, ASAT, Total cholesterol and albumin at all periods compared to animals that were treated. On one hand, animals treated with 25g/kg of ginger (T2), recorded the lowest levels of urea, total protein, albumin, ALAT, ASAT, Total cholesterol and globulin at all the study weeks and all these values were within the normal range (ASAT: 0-110U/l, ALAT: 0-103U/I, Urea: 17-45mg/dl, Total protein: 4.5-6.8g/dl, Albumin: 2.8-4.1g/dl, creatinine: 0.8-1.4mg/dl,) as described by Klem TB et al., (2010). These findings indicate that the dose of 25g/kg of ginger powder is not toxic but rather promotes the normal liver activities of the pigs. These results are consistent with that of Rubinsky et al., (2010) who analysed the effect of *Z. officinale* on *Toxocara canis* and *Toxocara cati*, and proofed that animals treated with *Z. officinale* separately or in combination with albendazole showed a significant decrease in the levels of ALAT and Aspartate aminotransferase ASAT. It also concurs with the works of Shewita and Taha (2018) who concluded that ginger powder also significantly reduced the level of total cholesterol. Also it is in accordance with Mohamed et al., (2012) who proofed that cholesterol level was significantly lowered by ginger. However, the results of this study was in total disconformity with that of other works (George et al., 2015 and Al-azazi et al., 2018) which showed that ginger treatments had no significant effect on biochemical parameters. These discrepancies in results are probably due to differences in treatment doses used. In the same light treatment T1 was not also toxic as it recorded normal values of cholesterol, ASAT and ALAT at all periods.

On the other hand, biochemical markers of toxicity, organ damage and dysfunction such as ALAT, ASAT, Albumin and creatinine were increased in pigs that were infected but not treated (T0-). In fact, these pigs recorded 103.47 and 110.78 for ALAT and ASAT respectively at week1 post treatment. These values are far above the normal range (Klem TB et al., 2010). This increase in the activities of the transaminases in the serum of untreated pigs shows that gastrointestinal nematodes caused significant damage to the liver where these enzymes are abundant hence the hepatic cytolysis.

Blood parameters were not affected by treatments, apart from platelets which number was significantly (p≤0.05) higher in T2 group. Over all the experiment period, all blood parameters were still in the normal range (red blood cells: 6.4-8.4, haematocrit: 34-50%, haemoglobin: 10.5-14.5g/dl, mcv: 49-59fl, platelets: 211-887) (Klem TB et al., 2010). This indicates that treatment with 25g/kg of ginger powder increased the production of platelet cells thus promoting blood clotting which prevents excessive bleeding. The results of this study is consistent with previous works (George et al., 2015; Mohamed et al., 2012; Al-azazi et al., 2018) stating that ginger treatments have no significant effect on the haematological parameters of pigs. However, it is in disagreement with that of Shewita and Taha (2018) who reported that ginger powder instead significantly increased the white blood cells count. Though treated pigs showed lower red blood cells, haematocrit, haemoglobin and mean corpuscular volume (mcv) and the highest number of platelets for all periods, these values were still within the normal range and therefore cannot be said to be significantly associated to treatment.

In conclusion this study showed that inclusion of 25g/kg of ginger powder in pig feed is more effective in reducing the total parasitic load ofgastrointestinal nematodeswhen compared to Mebendazole.

Ginger crude powder treatment improved the renal and hepatic health functions as it lowers cholesterol, creatinine, ALAT and ASAT levels thus proving that ginger is not toxic. Half the dose (12.5g/kg) and full dose (25g/kg) were not toxic. Therefore, the treatment with 25g/kg of ginger powder can be recommended for effective and safe use in the treatment of gastrointestinal nematodes in pigs.

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**Conflicts of interest**

The authors have no conflicts of interest to declare

**Ethical standards**

The authors assert that all procedures contributing to this work comply with the ethical standards of the relevant national and institutional committees on human experimentation and with the Helsinki Declaration of 1975, as revised in 2008. The authors assert that all procedures contributing to this work comply with the ethical standards of the relevant national and institutional guides on the care and use of laboratory animals.

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