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Hepato-Protective Potentials of Formulated Purple Fleshed Potato-Soybean and Peels of Purple Fleshed Potato-Soybean *(Solanum Tuberosum)* Based Diets on Wistar Rats Induced with Trypanosomiasis

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Annotation: The study evaluates the protective potential of formulated purple fleshed potato-soybean diets on Trypanosomiasis induced hepatotoxicity in wistar rats. The inadequacy of chemotherapy in in curing Trypanosomiasis and other dysfunctions caused by synthetic drugs is enough reason to formulate and evaluate some diets hepa-protection effect. The coded diets PFP1SJC3G2, PFP1SJC3G2 and PFP3SJC3G2 were formulations for HAT while PPL4SJC3G2 for AAT. Forty-Two animals were randomly divided into seven (7) groups (non-infected fed with rat pellet, infected fed with rat pellet only, infected treated with formulated diet PFP1SJC3G2 only, infected treated with PFP2SJC3G2 diet, infected treated with PFP3SJC3G2 diet, infected treated with PPL4SJC3G2 diet and infected treated with drug) of six rats each. The diets were administered once daily and drugs once in the cause of the experiment. AST, ALP and ALT level were returned to normal and showing that the effects of the formulated diets to the liver function are dose dependent and higher dose administration seems to be safe than the lower

dose. The study demonstrated that the diets could ameliorate or reverse liver damage induced by Trypanosomiasis in wistai rats.

Keywords: Trypanosomiasis, Formulated Purple Fleshed Potato, Peels of Purple Fleshed Potato-soybean Based Diets and Cardiac function.

INTRODUCTION

Trypanosomiasis is a disease that affects both humans and animal causing mortality and morbidity in the poor rural sub-Saharan region of Africa and among the world's most neglected tropical diseases (NTDs) (Gao *et al*, 2020; Rume *et al.,* 2022; BUsche *et al*., 2017and Yusuf *et al*., 2023). Neglected tropical diseases (NTDs) are groups of diseases caused by infections with parasites, viruses and or bacteria (WHO, 2022).

Health conditions often throw trypanosomiasis patients into confusion as regards their medication, due to drugs resistance and lack of appropriate preventive measures. Drugs has one or more of these drawbacks: high cost, highly toxic, need parenteral administration and parasites increasing resistance, while some were developed more than 30 years ago and are old (Marin and Soto, 2020). This confusion comes as a result of ignorance of what to choose as remedy even around the locality.

Organ damages in trypanosomiasis disease and treatment has been linked to infections and some medication (Ghaffar *et al.*, 2022). Therefore, investigations into some enzyme levels and histopathology of certain tissues in case of Trypanosomiasis disease and treatments is expedient.

Liver as an organ in the body, plays a central role in all metabolic processes. The organ liver is the site for bile production, which is essential for the fat metabolism and absorption. The liver cells break down fats to yield energy. It aids in conversion and production of some other nutrients also, such as carbohydrates, protein and vitamin K also into forms that the body uses, store and supplies to cells when need arises. Liver ensures the stability of blood glucose level by removal of excess sugar through the portal vein and stores as glycogen and incase of low sugar the same liver breaks down glycogen and release sugar (Gray, 1969. In protein metabolism, liver cells change amino acids in foods to energy. The by-product of this process is called ammonia which is a toxic substance. Liver also traps this toxic substance and convert it to a less toxic substance called urea then released to the blood. Urea is then transport to the kidneys which filters it from the blood as urine. Also, with the help of vitamin K, the liver produces proteins which is important for blood clothing. Liver serves as store house for vitamins and minerals (coper and iron) Friedman, 2020).

Liver Enzymes are specialized biocatalyst or proteins that catalyze a wide array of vital metabolic processes. As a result of their remarkable properties, they are used extensively in medical diagnosis. Researchers in the last two decades have concentrated more on enzyme such as amino transferase, aspartate aminotransferase and alkaline phosphatase etc. for clinical applications (Obasi and Ogugua, 2021. Enzymes are the prepared markers in various disease states such as myocardial infarction, jaundice, pancreatitis, cancer, neurodegenerative disorders etc. They provide insight into the disease process by diagnosis, prognosis and assessment of response to therapy (Akpanabiatu, 2019). Enzymes are used clinically in three principal ways: As indicators of enzymes activities or enzymes concentration in the body fluids (e.g. serum and urine) in the diagnosis and prognosis of various diseases; analytical reagents in measuring reagent of effects of other proteins or non-enzymes substance (e.g. substrates, proteins and drugs) in the body and as therapeutic agents.

Alanine Amino Transferase (ALT) which catalyze amino transfer reactions between an amino acid and a carbonyl – containing compound. They transfer the amino group and convert the amino acid to a keto acid; the accepting substrate, a keto acid is transformed into an amino acid (Devlin, 2011). They are often referred to as transaminase. Transaminases are predominantly present in most tissues of the body. The activity of ALT is notably high in tissues especially liver, heart and muscles. Any damage to the cells of these tissues may result in the release of this enzyme along with other intracellular proteins/enzymes into circulation, leading to increased activities of these enzymes in the blood (Uhegbu *et al*., 2015). Determinations of activity of ALT in serum of patients with liver disease such as viral hepatitis and various other forms of liver disease with necrosis, give high values even before the appearance of clinical signs and symptoms like jaundice. Activity levels of 20 to 50-fold higher than normal are often seen in liver cells damage but it may reach as high as 100 times in severe damage to cells. Alanine aminotransferase is a liver specific enzyme. Increased ALT in serum is hardly seen in tissues other than those associated with liver cell damage (Sherlock, 1997). Alanine amino transferase activity is within normal range or slightly increase in uncomplicated myocardial infarction (Friedman, 2003).

Aspartate aminotransferase is found in the cytosol as well as the mitochondria and is abundant in the liver, heart and skeletal muscle. It catalyses the reaction expressed below: -
L-Aspartate + 2 – Oxoglutarate → Oxalocetate + L – glutamate

AST is very useful in evaluation of liver injury although it is less specific than ALT. Elevated levels of AST may also be seen in cardiac or skeletal muscle injury. It is therefore not uncommon for AST to be assayed for evaluation of non-hepatic conditions such as myocardial infarction. Generally, common causes of raised transaminases include: alcohol, medications, non-alcoholic steatohepatosis, chronic hepatitis B and C, autoimmune disease, congestive heart disease, glycogen storage disease etc. (Li, *et al*, 2023). A ratio of AST: ALT of 2:1 is an indication of alcohol induced liver injury. Several drugs have been known to induce liver injury (Park and Ngu, 2000). They include antibiotics, statins, antiepileptic drugs, etc. as well as herbal medicine (Linda and Hyde, 2003).

Alkaline Phosphatase (ALP) exists in almost all tissues of the body. They are zinc containing metalloenzymes and membrane bound. Alkaline phosphatases belong to isoenzymes family. Hydrolysis of diverse organic phosphate esters, relocating of phosphate groups from donor substrate to an acceptor encompassing a hydroxyl group are done by Alkaline phosphatase. The active center of the enzyme contains a serine residue. High levels of these isoenzymes are present in intestinal epithelium, kidney tubule, osteoblasts in the bone, bile canalicular and sinusoidal membrane of the liver, placenta and the lactating breast (John *et al.,* 2012).

Roles of enzymes in chemical diagnosis of organ and dysfunction cannot be over emphasized. High liver enzymes are sign of liver damage that can lead to serious, life threatening conditions like liver cirrhosis, that can be fatal if not arrested on time.

Histology is fundamental in diagnosing diseases. Histology provides valuable insights into the microscopic examination of tissues structure, cellular composition and function, contributing to understanding health, disease, and biological processes. The present study, investigates liver tissue.

Liver is located in the upper right quadrant of the abdominal cavity with numerous functions such as metabolism of macronutrients, detoxification by removing toxic substances and drugs from the body; bile production for fat metabolism; storage of glycogen and vitamins and protein synthesis like albumin and blood clothing factors. The multifaceted roles of the liver make it crucial for overall health and metabolic balance. Liver are made up of structure such as lobes, hepatocytes, portal triads and sinusoids (Katunguka-Rwakishava, 1997).

Chemotherapy which form the major means of treatment for Trypanosomiasis has numerous short comings such as limited efficacy and lethal consequence (Field *et al*, 2017). Food is an

important source of chemical substances with potential therapeutic effects and therefore, research into food with formulation for use as antitrypanosomal is a useful strategy in the search for new antitrypanosomal diets, trypanosomiasis being one of the causes of scourge in human and animal.

MATERIAL AND METHODS

Materials

The materials for this study were purple-flesh potato (*Solanum tuberosum* L), Soybean (*Glycine max*), striped shore crab (*Pachygrapsus crassipes*), Crayfish **(***Cambarus spp***),** *Justicia seccunda vahl,* clove, ginger, garlic, *Trypanosoma Brucei, brucei*, albendazole, diminazene aceturate (Veriben). Others include: Camry kitchen weighing balance, hot air oven (Gallenkamp BS oven 250, Model No. 320), hammer mill, A Kenwood mixer, kitchen knife, sodium chloride (Dangote), sieve (5.3 mesh), water, Basins, Cups, plates, spoons and cellophane, spectrophotometer, EDTA, plain bottles and rags, 5ml syringes, thermometer, beaker, foil paper, cotton wool, normal saline, assay kits and reagents from Randox Laboratories Ltd, UK.

Sources of Raw Materials

Purple-flesh potato (*Solanum tuberosum* L) were purchased from a private garden in Elile Town, Eastern Obolo LGA, Akwa Ibom State, Nigeria. Blood leaves *(Justicia seccunda vahl)* were harvested (plucked) at convenience from three locations (Elile, Okorombuko and Kampa) all in Eastern Obolo LGA. Crayfish and crabs were purchased at Edowi and Lagos fishing ports respectively in Eastern Obolo LGA, Akwa Ibom State, Nigeria. Soybean, ginger, garlic, clove were purchased from Urua Akpan Andem Market in Uyo Metropolis, Akwa Ibom State, Nigeria.

Preparation of Purple Flesh Potato (*Solanum tuberosum L*) Flour

Purple flesh potato was sorted manually, washed with tap water, weighed with Kitchen scale, peeled manually with kitchen knife and sliced into 1mm thick slices. The slices were washed with clean tap water and blanched in sodium chloride solution (1 %, w/v) at 8^{o}C for 5min. They were oven dried at low temperature (50^{O} – 60^{O}) to constant weight, milled and sieved through 5.3 mm mesh sieve. The flour was packed in high density polyethylene bags and labelled as Purple flesh potato flour**.** The flow chart for the preparation of Purple flesh potato flour is presented in Figure 3.1.

Purple Fleshed Potato (*Solanum tuberosum* L*)*

↓

Sorting

↓

Washing

↓

Weighing

↓

Peeling

↓

Slicing the flesh to chips (1mm thick slice) for easy drying

↓

Blanching (in sodium chloride solution (1 %, w/v) 8^{o} C for 5 min.) to reduce Anti-nutrient content

↓

Drying (to constant weight)

↓

Milling: (Milled to fine flour, avoiding excessive handling to reduce the risk of contamination)

↓

Sieving (5.3mm screen size)

↓

Store: (package as Purple flesh potato flour in sealed containers at room temperature)

Preparation of Soybean (*Glycine max*) Flour

Two kilograms of soybean were sorted by hand picking, cleaning and soaked in tap water (H_2O) in the ratio of 1:3 (w/v) for 6 hours. The grains were oven dried at low temperature of 40 -50^{O} C for 10 minutes in a hot air oven (Gallenkamp BS oven 250, dehulled manually and sun dried to constant weight. The dried sample were then milled to fine flour, sieved through 70 mm mesh screen. The flow chart for the preparation of soybean flour is presented in Figure 3.3.

Soybean (*Glycine max*)

↓

Sorting (Assessment of physical appearance, size and shape, Moisture content determination, insect infestation assessment)

↓

Cleaning (3 steps washing process of pre-rinse, scrubbing and final rinse)

↓

Soaking (for six hours)

↓

Drying (Oven dried at 50-60^{o} for 10 minutes)

↓

Dehulled (manually)

↓

Sun dry (to constant weight)

↓

Milling: Milled to fine flour (avoid excessive handling to reduce risk of contamination)

↓

Sieving (7.0 mm screen size)

↓

Soybean flour (storage in sealed container at room temperature)

Preparation of Crayfish (*Cambarus sp)*

5 Kg of dry crayfish was sorted, heated up in the oven, and filtered using a colander, milled into fine flour and stored at room temperature.

Preparation of Crabs (Stripe shore crab (*pachygrapsus crassipes*)

5 Kg of crabs were washed, boiled, and filtered using a colander, cooled, shell separated from the eatables, oven dry at low temperature of 40-50^{O} to constant weight, milled into fine flour, sieved through 70 mm mesh screen and stored at room temperature.

Processing of Blood Leave (*Justicia seccunda vahl)*

One kilogram of *Justicia secunda vahl* were sorted, washed with cleaned tap water and blanched in sodium chloride solution (1 %, w/v) at 8^{o}C for 2 min, dried to constant weight at room

temperature, grind, sieved through 7.0 mm meshed and stored as *Justicia secunda vahl* powder in an airtight container.

Formulation of Purple Fleshed Potato-Soybeans Based Diets Therapy

The anti-trypanosomal diets therapy were formulated based on the procedure of Pearson square method. Parasite infected patient are recommended to have increase protein, low carbohydrate, high in phytonutrient, high fibre per unit of food volume by percentages (WHO, 2022; CDC, 2022 and NIH, 2022) and modified for this work as presented in Table 3.1

The PFPSJC3G2 flour: Purple (P), flesh (F), Potato (P), soybean (S), *Justicia secunda vahl* (J), Crayfish (C), crab (C), Clove (C), Ginger and garlic for Potato man diets. While, PPL4SJC3G2: Purple fleshed Potato (P), Peels (PL) soybean peels (S), *Justicia secunda vahl* (J), crayfish fur (C), Crab shells, Clove, Ginger and garlic for animals' diet were mixed as shown in Table 3.1 below. A Kenwood mixer were used for mixing the samples for 2 minutes to achieve uniform mixing.

Table 3.1: Percentage Composition of the Purple-Fleshed Potato-Soybeans based Diets

Group	Diets	Potato (%)	Soybean (%)	*Justicia secunda vahl* (%)	Crayfish (%)	Crab (%)	Clove (%)	Ginger (%)	Garlic (%)
A	PFP1SJC3G2	74	-	7.4	7.4	7.4	0.8	1.5	1.5
B	PFP2SJC3G2	59.2	14.8	7.4	7.4	7.4	0.8	1.5	1.5
C	PFP3SJC3G2	37	37	7.4	7.4	7.4	0.8	1.5	1.5
D	PPL4SJC3G2	42	32	7.4	7.4	7.4	0.8	1.5	1.5

Estimation of serum Liver Enzymes

Estimation of Alanin Amino Transferase (ALT) in serum of *T. Brucei, brucei* Infected and Treated Wistar Rats.

Procedure:

100 µl of distilled water and sample were pipette into sterile plain tubes labeled blank and sample respectively. The sample tubes were arranged serially according to the number of samples. 500 µl of reagent R I was added to all the tubes mixed and incubated for 30 minutes at 37^{o} c. Thereafter, 500 µl of reagent R2 was added to all tubes, mixed and allowed to stand for exactly 20 minutes at room temperature. 5 ml of 0.1N NaOH was added to all tubes and the Absorbance of ALT in the sera was read immediately at 546 nm against reagent blank.

Calculation:

ALT activity in the serum was obtained proportionally from the concentration given in the kit's manual.

Activity of ALT in serum (µ/L)

$$\frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \quad \text{X conc. of standard X Activity of Standard ALT}$$

Estimation of serum Aspartate aminotransferase (AST) Activity of *T. Brucei, brucei* Infected and Treated Wistar Rats

100 µl of distilled water and sample were taken into sterile plain tubes labeled blank and sample respectively. The sample tubes were arranged serially according to the number of samples. 500 µl of R1 was added across board, mixed and incubated at 37^{o} c for 30 minutes. Reagent R2 was added to all the tubes after incubation and allowed to stand at room temperature for 20 minutes. Exactly 5 ml of 0.1N NaOH was also added to all tubes. Absorbance of AST in the sera was read immediately at 546 nm against reagent blank.

Calculation:

AST activity in the serum was obtained proportionally from the concentration of standard AST given in table contained in the kit's manual.

AST activity in serum (1µ/L)

$$\frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} = \text{concentration of standard AST}$$

Estimation of Alkaline Phosphate (ALP) in serum of *T. Brucei, brucei* Infected and Treated Wistar Rats.

Substrate R1b was reconstituted with 10 ml of buffer R1a, 500 µl of this solution was added to 10 µl of sample, mixed and initial absorbance read immediately while starting a timer simultaneously. Absorbance was read at 1, 2, and 3 minutes intervals all at 405 nm. This analysis was carried out at room temperature.

Calculation:

ALP activity in the serum was calculated using this formula

ALP activity (1µ/L) = 2760 x DA 405 nm/minutes

Where

ΔΔ = change in absorbance

Histopathological Analysis of *T. Brucei, brucei* Infected and Treated Wistar Rats

Wistar rats were euthanized at the end of the experiment, liver was collected in formalin and processed for histopathological examination using standard histological techniques (Luna, 1968). About 1m thick tissue sections from the liver was collected and fixed in 10 % phosphate buffered formalin for a minimum of 48 hours before tissue preparation. The tissue sections were prepared using Bancroft and Layton, 2013 technique. The tissue was subsequently trimmed, dehydrated in four ascending grades of alcohol (70 %, 80 %, 90 %, and 100 %), cleared in 3 grades of xylene, and embedded in molten wax. After embedding, the tissue containing wax blocks were cut into 5µm thick sections with a rotary microtome, floated in water bath at 60°C, placed on clean grease free glass slides, and placed on a slide warmer set at 60°C overnight. The Sum thick sectioned tissues on glass slides were subsequently cleared in 3 grades of xylene and rehydrated in 3 descending grades of alcohol (90 %, 80 %, and 70 %). The sections were then stained with Mayer's hematoxylin for 5 minutes. Bluing was done with ammonium chloride. Differentiation was done with 1% acid alcohol before counter staining with Eosin. Permanent mounts were made on degreased glass slides using a mountant; DPX. The slides were examined using a Motic™ compound light microscope and then microphotographs were taken using 5 megapixels C-mount™ microscope camera. The pathological effects on the morphology of liver of the *Trypanosoma Brucei, brucei* infected albino rats was examined for organs alteration and organ failure.

RESULTS

Table 4.1 shows therapeutic effect of formulated purple fleshed potato-soybeans based diets on liver function of *T. Brucei, brucei* infected wistar rats. The means therapeutic effect of formulated purple fleshed potato-soybeans based diets on the *T. Brucei, brucei* infected liver function are presented in Table 4.1.

The mean ALT concentration of *T. Brucei, brucei* infected rats treated with control sample A (RPSANF) was 22.00 µl, sample B was 14.50 µl, sample C was 14.00 µl, sample D was 14.40 µl, and sample E was 15.20 µl. The mean concentration of ALT of infected rats treated with control sample A (RPSANF) was significantly higher ($p < 0.05$) than that of sample B, C, D and E. Among the formulated purple fleshed potato-soybeans based diets, sample E was significantly

higher in ALT than samples B, C and D. By implication the formulated purple fleshed potato-soybeans based diets had lower ALT when compared with control sample A (RPSANF) (pellet commercial animal feed). Infected rats treated with sample E (PPL4SJC3G2) had the highest concentration of ALT among the formulated purple fleshed potato-soybeans based diets.

The mean of AST concentration in *T. Brucei, brucei* infected rats fed with control sample A (RPSANF) was 20.80 µl, sample B (PFP1SJC3G2) was14.00 µl, sample C (PFP2SJC3G2) was 15.60 µl, sample D (PFP3SJC3G2) was 14.80 µl, and sample E (PPL4SJC3G2) was 15.40 µl. The mean concentration of AST of *T. Brucei, brucei* infected rats treated with control sample A, was significantly increase ($p < 0.05$) than that of samples B, sample C and D but not different from that of sample E. Among the formulated purple fleshed potato-soybeans based diets, sample E was significantly higher ($p < 0.05$) in AST than samples B, C and D. By implication the formulated purple fleshed potato-soybeans based diets (E) had lower AST compared with control sample A RPSANF (commercial animal feed).

The mean of ALP concentration in *T. Brucei, brucei* infected rats treated with control sample A (RPSANF) was 113.40 µl, sample B (PFP1SJC3G2) was 68.25 µl, sample C (PFP2SJC3G2) was 73.00 µl, sample D ((PFP3SJC3G2) was 77.00 µl, and sample E (PPL4SJC3G2) was 80.40 µl. The mean concentration of ALP of infected rats treated with control sample A (RPSANF) was significantly higher ($p < 0.05$) than that of samples B, C, D and E. Among the formulated purple fleshed potato-soybeans based diets, sample B was significantly lower in ALT than samples C, D and E. By implication the formulated purple fleshed potato-soybeans based diets (C, D and E) had lower ALP compared with control but not different from pellet (commercial animal feed). ALP levels were significantly ($p < 0.05$) reduced in the treatment groups when compared to the control group.

Table 4.1: Therapeutic Effect of Formulated Purple Fleshed Potato-Soybeans and peels Based Diets on Liver Function of *T. Brucei, brucei* Infected Wistar Rats

SAMPLES	TREATMENTS	AST	ALT	ALP
A	NON-INF RPSANF	13.40 ± 1.67^{a}	13.80 ± 1.30^{a}	87.80 ± 14.94^{a}
B	INF RP5ANF	20.80 ± 2.39^{b}	22.00 ± 1.00^{b}	113.40 ± 15.39^{c}
C	PFP1SJC3G2	14.00 ± 1.63^{a}	14.50 ± 1.29^{a}	68.25 ± 4.43^{a}
D	PFP2SJC3G2	15.60 ± 2.07^{a}	14.00 ± 1.58^{a}	73.00 ± 3.39^{ab}
E	PFP3SJC3G2	14.80 ± 1.30^{a}	14.40 ± 2.07^{a}	77.00 ± 4.69^{ab}
F	PPL4SJC3G2	15.40 ± 2.39^{a}	15.20 ± 1.09^{a}	80.40 ± 2.70^{b}
G	INF RPSANF + DRUG	20.84 ± 2.39^{b}	21.00 ± 1.58^{b}	98.80 ± 2.28^{b}

Note: *Each value is a mean of triplicate determinations (n = 5). Mean value with the same alphabet as superscript on the same column is not significantly different from one another ($p < 0.05$). (Mean ± Standard Error of Mean).*

A = $p < 0.05$ (all treatment group in comparison with formulation PFP1SJC3G2 100 control)

B = $p < 0.05$ (all treatment group in comparison with formulation PFP2SJC3G2 70: 30)

C = $p < 0.05$ (all treatment group in comparison with formulation PFP3SJC3G2 50: 50)

D = $p < 0.05$ (all treatment group in comparison with PPL4SJC3G2 55:45 peels of Potato-soybean animal feed)

Source: Field work (2025).

Plate 1 – 7 shows the therapeutic effect of formulated purple fleshed potato-soybeans Based diets on histology of liver of *Trypanosome Brucei, brucei infected* wistar rats Histology of Liver of *Trypanosome Brucei, brucei infected* wistar rats (Liver treated with diets Only)

Plate 1 shows the photomicrograph of a transverse section of a *T. Brucei, brucei* non-infected liver tissue fed with RPSANF, demonstrating a mild alteration in the hepato-architecture with

populated hyperchromatic cells within the hepatic lobules. (H&E x100). Inference: Mildly affected.

Histologic section of the *T. Brucei, brucei* infected liver treated with control (RPSANF only) Plate 4.2 revealed severe alterations in the hepato-architecture with degenerating hepatocytes (black arrow), eroding portal area, and hyperplasic ductal epithelium (yellow arrow) within the hepatic lobules. (H&E x100). Inference: Severely affected when compared with formulated purple fleshed potato-soybeans based diets.

Plate 4.3 shows the photomicrograph of a transverse section of a *Trypanosome Brucei, brucei* infected liver tissue treated with PFP1SJC3G2, demonstrating a moderately altered hepato-architecture with degenerating hepatocytes (black arrow) and vacuolated hepatocytes (red arrow) within the hepatic lobules. (H&E x100). Inference: Moderately affected.

Plate 4.4 shows the photomicrograph of a transverse section of a *Trypanosome Brucei, brucei* infected liver tissue treated with PFP3SJC3G2 demonstrating a mild alteration in the hepato-architecture with hyperplasic ductal epithelium (yellow arrow) and proliferating Kupfer cells within the sinusoids of the hepatic lobules. (H&E x100). Inference: Mildly affected.

Plate 4.5 shows the photomicrograph of a transverse section of a *Trypanosome Brucei, brucei* infected liver tissue treated with PFP3SJC3G2 demonstrating a mild alteration in the hepato-architecture with hyperplasic ductal epithelium (yellow arrow) and proliferating Kupfer cells within the sinusoids of the hepatic lobules. (H&E x100). Inference: Mildly affected.

Plate 4.6 shows the photomicrograph of a transverse section of a *Trypanosome Brucei, brucei* infected liver tissue treated with PPL4SJC3G2 demonstrating a normal hepato-architecture with well-presented portal area, protected hepatocytes and proliferating Kupfer cells within the sinusoids of the hepatic lobules. (H&E x100). Inference: Not affected.

Photomicrograph of a transverse section of a *trypanosome Brucei, brucei* infected liver tissue treated with RPSANF+ drug demonstrating a normal hepato-architecture with well-presented portal area, protected hepatocytes and proliferating Kupfer cells within the sinusoids of the hepatic lobules. (H&E x100). Inference: Not affected Plate.4.7

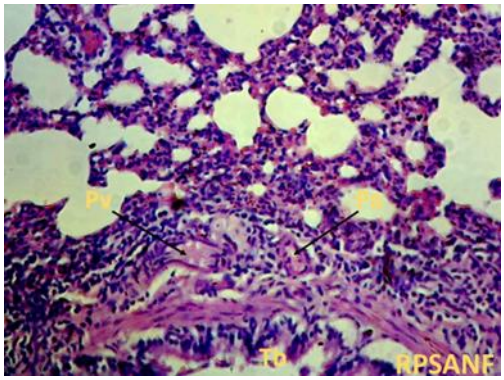

Plate 4.1

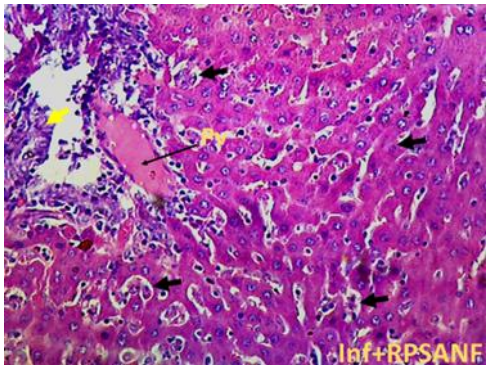

Plate 4.2

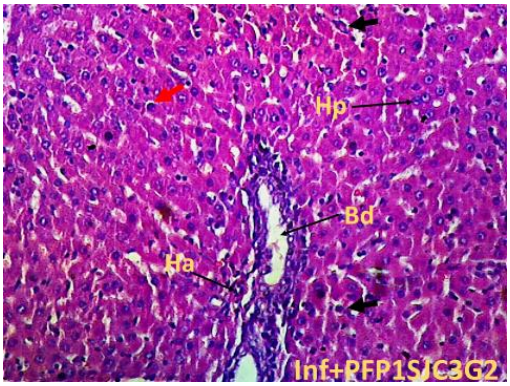

Plate 4.3

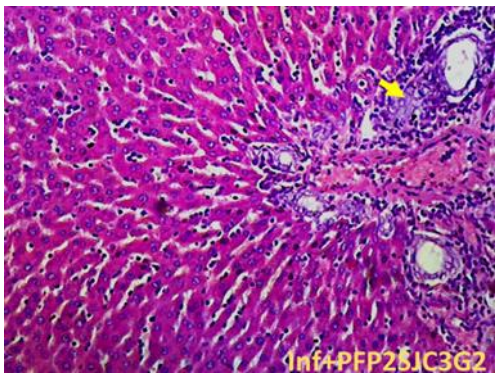

Plate 4.4.

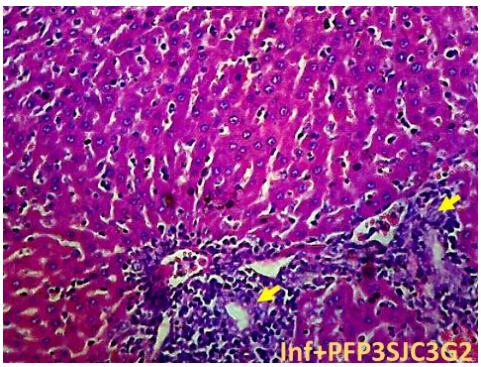

Plate 4.5

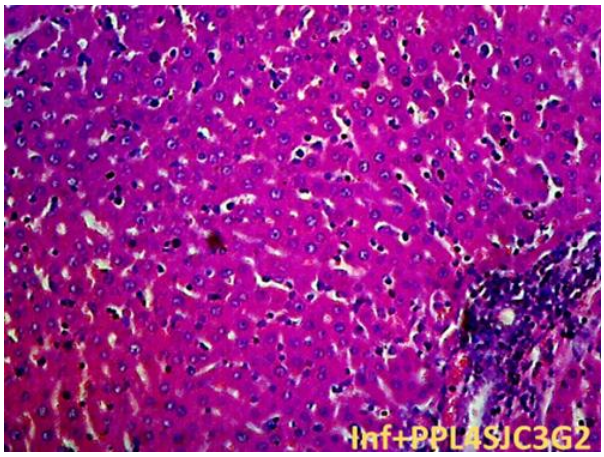

Plate 4.6.

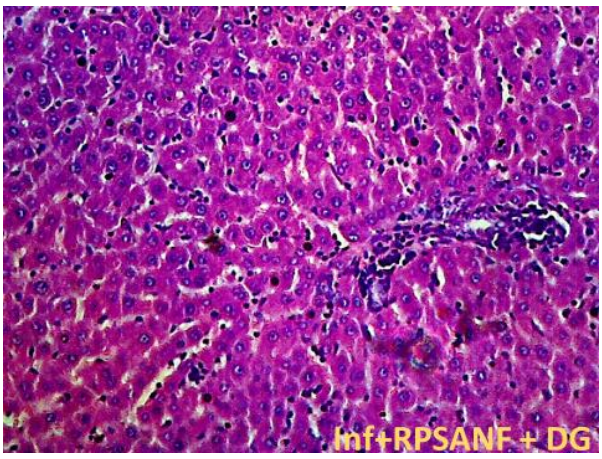

Plate 4.7.

DISCUSION

Therapeutic Effect of Purple fleshed Potato on Liver Functions of *T. Brucei, brucei* Infected Rats

Some of the considered liver biomarkers evaluated in our study are AST, ALT, ALP. An increase in these enzymes are indication of extra-hepatic damage or sign of hepatoxicity. A significant ($p<0.05$) increase in the AST and ALT in the blood of the infected and untreated rats (control group) clearly denotes an alteration in the liver function which was obviously seen as compared to the treated group which is an indication that there was a greater damage resulted from the parasite infection than the formulated purple fleshed potato-soybeans based diets. However, it was observed that there was no significant ($p>0.05$) difference in AST and ALT among the groups fed with formulated purple fleshed potato-soybeans based diets. Whereas, there was significant ($p>0.05$) in ALP as seen in the control group when compared with the formulated purple fleshed potato-soybeans based diets. Suggesting a deleterious effect attributed to *T. Brucei, brucei* parasites. There were no significant ($p>0.05$) in ALP among the formulated purple fleshed potato-soybeans based diets, except in PPL4SJC3G2. Hepatotoxicity may result from an infection. The present finding is not different from the findings of Alasrag *et al*. (2021) who reported a lowered level of AST and ALT following an oral administration of ethanolic extract of rosemary leaf. Sulaiman *et al*. (2022) reported a non-observable increase in ALT and AST indices in the ethanolic non- Trypanosoma challenged treated rats compared with the normal control. A similar study by Barghasi (2021) also reported an increase in AST, ALT, and urea following Salvia essential oil administration (Intraperitoneal) to the infected rat. However, rats in the groups treated with the PFP1SJC3G2, PFP2SJC3G2, PFP3SJC3G2 and PPL4SJC3G2 respectively) in our study followed similar trend where AST, ALP and ALT level were returned to normal showing that the effects of the formulated diets to the liver function are dose dependent and higher dose administration seems to be safe than the lower dose.

Histopathological Effects of Formulated Purple Fleshed Potato-Soybeans Based Diets on Trypanosomiasis in Experimental Animals

Histopathological indices are used to determine the level of organ and tissue damage resulted from the presence of either the antigen or parasite in the body of the humans and animals. Although some diets may not be able to protect the experimental animal from parasitemia, but has visible protection against the organs. Generally, the parasite (Trypanosoma species) releases toxins that disrupt organs and cause cell injury.

Liver: In this study liver of *Trypanosoma Brucei, brucei* infected rats treated with the control (RPSANF) sample show enlargements and severe alteration in the hepato-architecture with degenerating hepatocytes, eroding portal area, and hyperplasic ductal epithelium within the hepatic lobules when compared with the groups treated with formulated purple fleshed potato-soybean based diets (PFP1SJC3G2, PFP2SJC3G2, PFP3SJC3G2) which shows moderately, mildly and mildly respectively. while group treated with the formulated peels of purple fleshed potato- soybean based diet (PPL4SJC3G2) shows no alteration in of the liver tissues. This work is in agreement with Ghaffar *et al*. (2016) who reported that liver is another important organ that could be affected by Trypanosoma infection. Studies conducted by Ghaffar *et al*. (2016) reported that in Trypanosoma infected group, there was a mild-severe degenerative change in the liver and the hepatocytes lost their original shape, swollen and rounded with vacuolar spaces in the cytoplasm. The changes were attributed to anaemia caused by starvation of cells and anoxia which resulted to hypo glycaemia of the liver.

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