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SELECTED LIVER AND KIDNEY BIOCHEMICAL PROFILES OF HYBRID CATFISH EXPOSED TO *JATROPHA CURCAS* LEAF DUST

Kabir M. Adamu*, M. C. Isah, T. A. Baba, T. M. Idris

Department of Biological Sciences, Ibrahim Badamasi Babangida University, P.M.B. 11, Lapai, Nigeria, tel: +234(0)8035826075
* Corresponding Author, E-mail: kabrmoh@yahoo.com

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ABSTRACT

This study investigated the effect of sublethal concentrations (7.50, 5.00, 2.50 and 0.00 g\L (control)) of *Jatropha curcas* leaf dust on some liver and kidney biochemical profiles such as total protein, total bilirubin, total albumin and total globulin of hybrid catfish (*Clarias gariepinus* (♀) and *Heterobranchus bidorsalis* (♂)) after a 14-day experimental period. The result was significantly different in the mean value of determined liver total bilirubin, while total protein, albumin and globulin were insignificantly different in the exposed fish compared with the control. The kidney revealed varying levels of insignificant difference in its level of total protein, total albumin, total bilirubin and total globulin. Therefore, this investigation has revealed that sublethal concentration of *Jatropha curcas* has no effect on the basic function of the determined biochemical profiles of hybrid catfish and that the changes were directly proportional to *J. curcas* concentration.

INTRODUCTION

Plants are virtually inexhaustible source of structurally diverse and biologically active substance (Ujváry, 2000). *Jatropha curcas* (linn) or physic nut or Kashia Bukun in Nupe language is found growing on uncultivated land in central Nigeria. It is a tropical plant that can be grown in low to high rainfall areas, thus can be used to reclaim land as a hedge and as a commercial crop. J. curcas plant has a caustic milky sap and toxic seed. It is one amongst many in family Euphorbiaceae with wide range of medical importance in Nigeria (Fajas et al., 1986 and Igoli et al., 2005). The bark is used as fish poison and the oil from seed is used as bio-diesel (Achten et al., 2008). However, Makkar and Becker, (1998) reported some toxins and anti-nutritive compounds such as curcin, tannins, phytates, flavonoids, saponins, vitexine, cyanide and trypsin inhibitor in the plant.

Its seed are considered highly nutritious and could be exploited as a rich and economical protein supplement in animal feed (Makkar et al., 1998). *J. curcas,* which was found growing in semi arid, arid and tropical environments, contains various anti-nutrients which if properly processed could be used to replace most conventional feedstuffs (groundnut cake, soybean cake and cotton seed cake, etc.) (Belewu et al., 2010).

Hybrid catfish is the hybrid catfish *Clarias gariepinus* (♀) and *Heterobranchus bidorsalis* (♂). They are desirable food worldwide. The choice of the test catfish in this study was attributed to the fact that in order to extrapolate meaningful, relevant and ecologically significant results from aquatic toxicity test, not only appropriate test but also appropriate organism should be used. Hybrid cattish constitutes one of the main fish species of economic importance in Nigeria and elsewhere in the developing world, thus its use in this study.

Studies on the characterizations of J. curcas include Tint and Mya (2009), Demissie and Lele, (2010) and Abdurrahman and Oladele, (2011). The effect of J. curcas has been previously studied on different organisms (Adam 1974, Adam and Magzoub, 1975,

Liu et al., 1997, Makkar et al., 1998, Chivandi et al., 2006, Goel et al., 2007, Igbinosa et al., 2009, Awasthy et al., 2010, Dheeraj et. al., 2010, Arekemase et al., 2011, Abo et al., 2011 and Musa et al., 2011), but none have reported its effect on the liver and kidney biochemical profiles of hybrid catfish. Thus this research was poised to ascertain the effect of *J. curcas* leaf dust at sublethal concentrations on selected liver and kidney biochemical profiles such as total protein, total albumin, total globulin and total bilirubin of hybrid catfish in a static semi-renewable bioassay system after a 14-day exposure period with the objective to determine the involvement of these examined biochemical profiles in environmental monitoring and health condition of the experimental fish, and to provide information on the sublethal effects of the plant material on the examined biochemical profiles of the test fish.

MATERIALS AND METHODS

Hybrid catfish was chosen because of the position it occupied within the food chain; it is amenable to laboratory conditions/testing, shows genetic stability thus uniform population can be tested; it is produced artificially and commands high preference by consumers. Juvenile of the fish, mixed sex and the same brood stock of mean weight (9.701.17 g) were obtained from Makolo Fish Farm in Chanchaga, Local Government Area of Niger State, Nigeria. They were transported to the Department of Biological Sciences Laboratory, Ibrahim Badamasi Babangida University, Lapai in the morning hours (08:30 – 09:00) in a large plastic container. The fish were held in the laboratory in large glass aquaria of 80 L capacity with clean borehole water. They were then acclimatized for 14 days during which they were fed to satiation with commercial fish feed pellets (Copens 2.0 mm) twice daily. Leftover feed and faces were siphoned off promptly and dead fish were promptly removed to avoid contamination. The percentage of death fish recorded during acclimatization was less than 1%, as such the fishes were accepted as being adapted to the laboratory conditions. They were then transferred to the experimental plastic aquaria (ten (10) fish/40 L aquaria) in two replicates.

The leaves of *J. curcas* were obtained from Patigi in Patigi local Government Area of Kwara State, Nigeria. The collected leaf samples were air-dried for 21 days, grounded into powder with the use of laboratory mortar and pestle therefore sieved and stored in a sealed plastic container until required. The concentrations used for the experiment were 7.50, 5.00, 2.50 and 0.00 g/L which were obtained after preliminary investigation. These concentrations were introduced in four (4) sets of aquaria with two replications.

Each of the ten (10) juvenile fish were placed in the 40 L glass aquarium. Bore-hole water was used during the acclimatization and exposure period. Feeding regime (08:00 and 18:00 hours) during exposure period was the same as that of the acclimatization period. In order to monitor the toxicant strength, level of dissolved oxygen, the effects of evaporation, Ammonia concentration and reduced stress during experimentation, the test media were replaced by 50% prepared concentrations of the same quality, after removing its equivalent along with undigested food and defecation every 48 hours to maintain the requisite level and potency of the concentration. The exposure period lasted for fourteen (14) days. At the start (0 hour) of the experiment, the sum total of ten fish were sacrificed and analyzed for the biochemical profiles. The water quality parameters of the experimental water used were 25.20±0.24^{o}C, pH 7.15±0.12, dissolved oxygen 9.21±0.32 mgl^{-1} and total alkalinity 27.89±0.41. At the end of the 14-day experimental period, fifteen fishes were sacrificed from each concentration where liver and kidneys were removed from the fish and pulverized in a laboratory mortar and pestle separately while extractions were prepared by adding 2 ml of 10% sucrose solution before been centrifuged (Mahobia, 1987) and stored in another test-tube in the refrigerator until analysis.

Total protein was determined in accordance to the principle of Tietz (1995), total albumin by colorimetric method of Doumas et al. (1971) and total bilirubin by colorimetric method of Sherlock (1951), thereof their absorbance (A) was read against the blank using a spectrophotometer (20D PEC Medicals, U.S.A).

All data were presented as means ± standard error, the data from the 14-day *J. curcas* leaf dust exposure were first analyzed using a one-way (concentration) analysis of variance, after which individual means were compared using Bonferoni multi-sample correction/test. Control values obtained at the beginning and the end of the 14-day exposure period were not significantly different ($p>0.05$) and were therefore combined as one control. In all cases, differences were considered statistically significant at either $p<0.01$ or $p<0.05$.

RESULTS

The changes in biochemical profiles (total protein, albumin, bilirubin and globulin) recorded in the test fish following exposure to the various sublethal concentrations of *J. curcas* leaf dust after the 14-day exposure period are as presented herein.

The mean values of total protein (g/dL) in the liver and kidney of hybrid catfish exposed to sublethal concentrations of *J. curcas* leaf dust after the 14-day exposure period were presented in Fig. 1a and b respectively. There was an insignificant ($p>0.05$) increase and decrease in the liver and kidney total protein values respectively when compared with the control group.

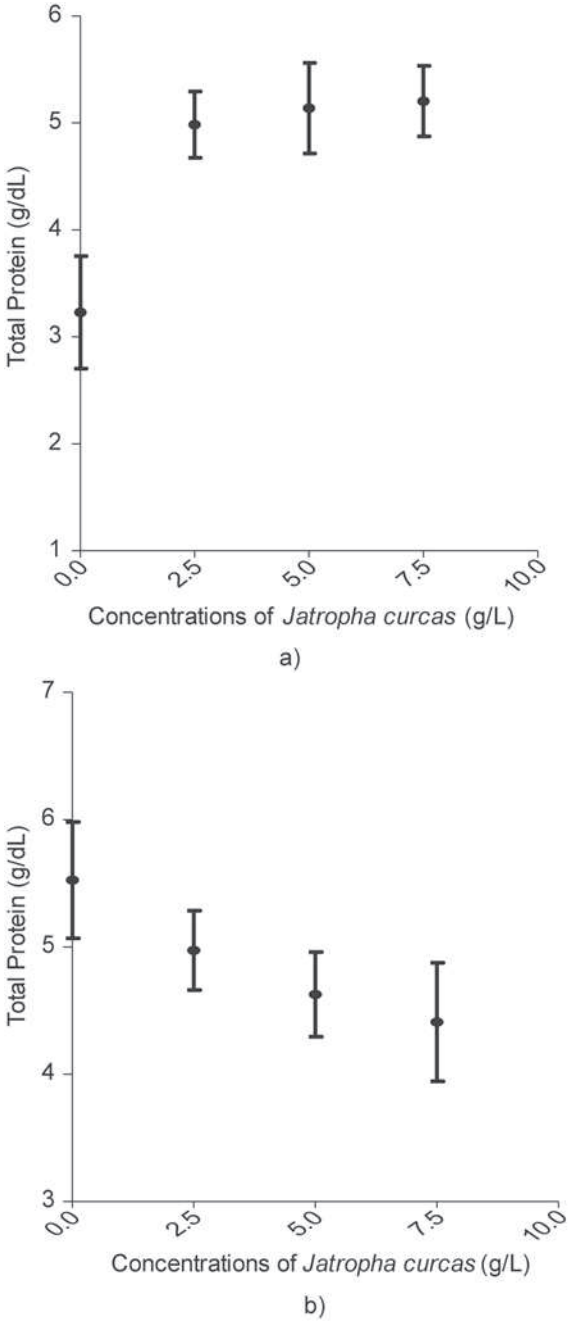


Fig 1: The mean total protein (g/dL) value of hybrid catfish after 14-day exposure period to sublethal concentrations of *J. curcas* leaf dust. Each point represents the mean and vertical bars indicate the standard error of the mean. a=liver, b=kidney

The mean values of total albumin (g/dl) in the liver and kidney of hybrid catfish exposed to sublethal concentrations of *J. curcas* leaf dust after the 14-day exposure period were presented in Fig. 2a and b respectively. There was an insignificant ($p>0.05$) decrease in the level of liver and kidney total albumin values with 5.0 g/L showing insignificant ($p>0.05$) increased values.

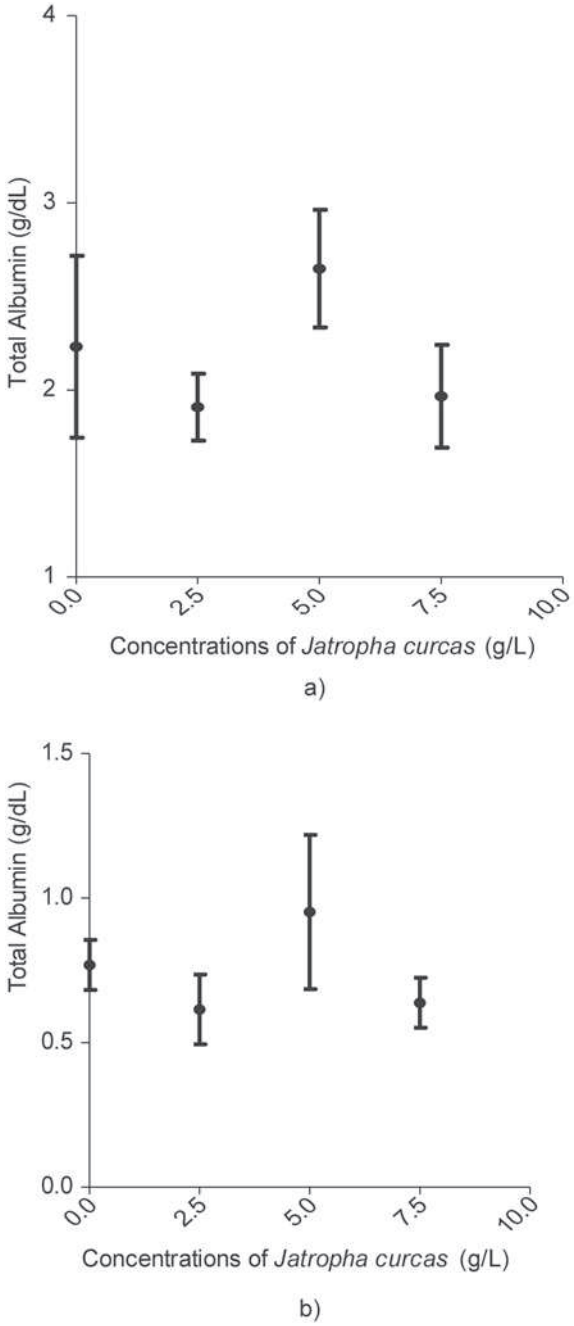
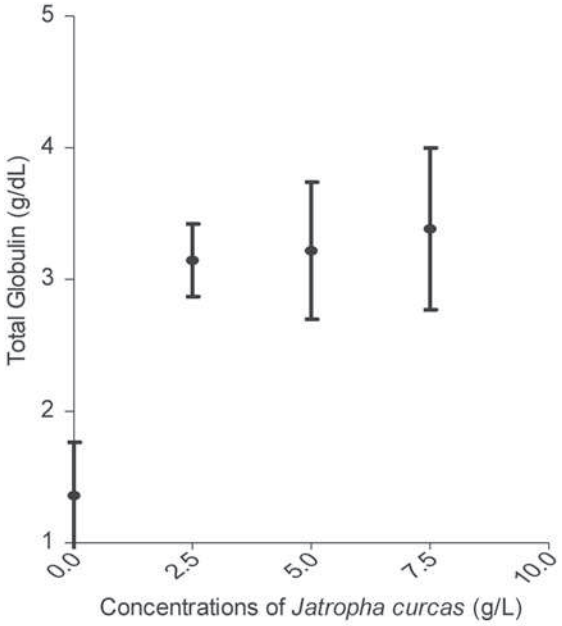


Fig 2: The mean total Albumin (g/dL) value of hybrid catfish after 14-day exposure period to sublethal concentrations of *J. curcas* leaf dust. Each point represents the mean and vertical bars indicate the standard error of the mean. a=liver, b=kidney

The mean value of total globulin (g/dL) in the liver and kidney of hybrid catfish exposed to sublethal concentrations of *J. curcas* leaf powder after the 14-day exposure period was presented in Fig. 3 a and b respectively. There was an insignificant ($p>0.05$) increase and decrease in the liver and kidney total globulin values respectively.



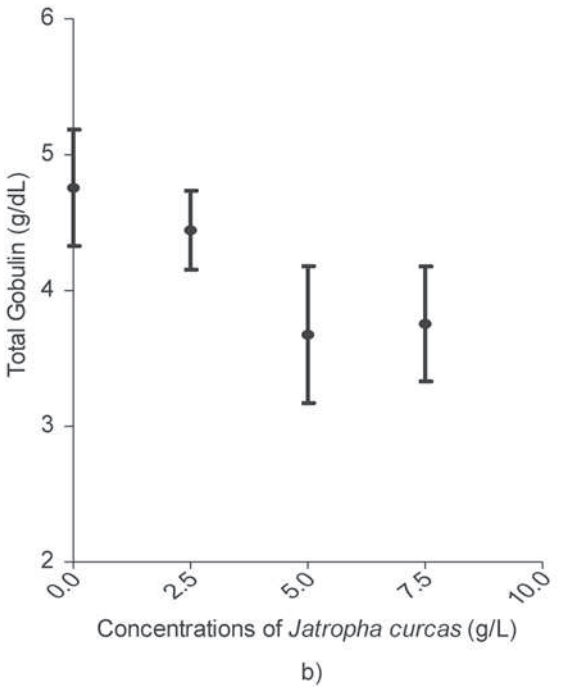
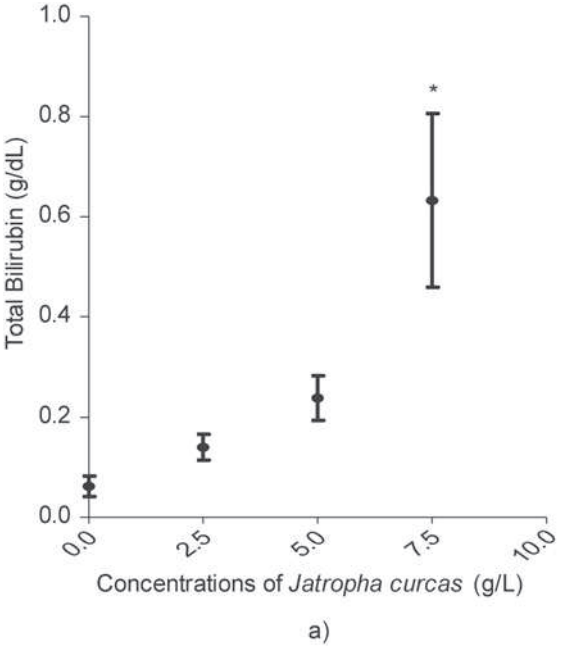


Fig 3: The mean total globulin (g/dL) value of hybrid catfish after 14-day exposure period to sublethal concentrations of *J. curcas* leaf dust. Each point represents the mean and vertical bars indicate the standard error of the mean. a=liver, b=kidney

The mean values of total bilirubin (g/dL) in the liver and kidney of hybrid catfish exposed to sublethal concentrations of *J. curcas* leaf dust after the 14-day exposure period was presented in Fig. 4 a and b respectively. There was an insignificant ($p>0.05$) and significant ($p<0.05$) increase in the kidney and liver total bilirubin values respectively.



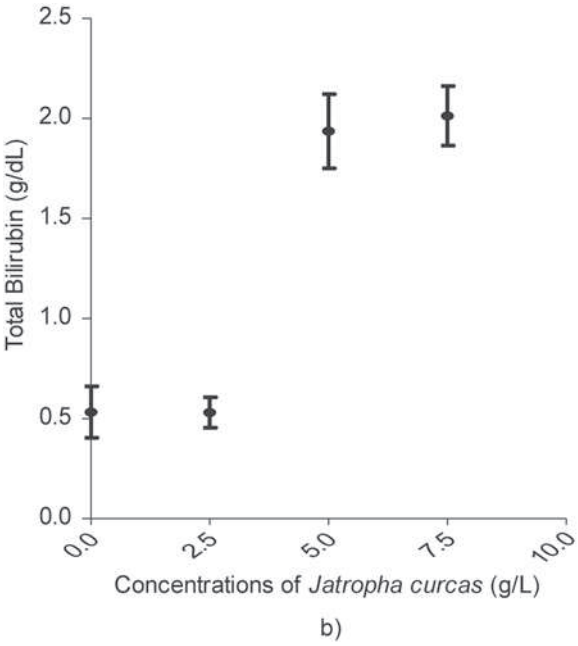


Fig 4: The mean total bilirubin (g/dL) value of hybrid catfish after 14-day exposure period to sublethal concentrations of *J. curcas* leaf dust. Each point represents the mean and vertical bars indicate the standard error of the mean and the asterisk represents a significant difference between the control and experimental group at 0.05 level a=liver, b=kidney

DISCUSSION

Biochemical profiles such as total protein, albumin, globulin and bilirubin could be used as Biomarker (Barnhoorn and Van Vuren, 2004). Total protein is a vital constituent of all cells and tissues. It plays a vital role in the determination of the physiological phases of a cell. All biological activities are regulated by enzymes and hormones, which are also proteins. The insignificant increase in liver total protein may be due to a high demand of protein to metabolize the plant content or due to hemo-concentration arising from fluid loss (Awasthy et al., 2010). Thereof the quantity of total protein is dependent on the rate of protein synthesis or on the rate of its degradation; resulting in the decrease of kidney total protein thus consequent impairment in protein synthesis machinery (Adamu et al., 2008). This might also be a result of a higher demand from the body to counter stress, which triggers protein metabolism.

Albumin functions as transport proteins for several steroid hormones and for fatty acids from adipose tissue to muscle. It is responsible for about 80% of the total osmotic regulation.

The increased value of liver albumin at concentration of 5.0 g/l could be a result of accumulation of protein in the liver impeding other function of the liver, and the decrease in the value of liver albumin at 7.5 g/l may be due to fluctuation or instability in the protease which impede its function of transportation. The corresponding decrease in the value of kidney total albumin may be a result of *J. curcas* impeding its function of transportation (Adamu and Kori-Siakpere, 2011) or a result of inhibitory effect of the leaf powder on protein hydrolytic activity due to elevation of protease activity that corresponds with the values of total protein (Fafioye et al., 2005).

The increased value of liver globulin showed the effect of *J. curcas* leaf powder as an inhibitory agent on hydrolytic activity due to the elevation of protease activity in the liver (Adamu and Kori-Siakpere, 2011), thus decreased kidney total globulin values.

Bilirubin is a metabolic waste product which is important in the evaluation of liver function. The insignificance increase in bilirubin may be correlated to the destruction of red blood cells by the plant contents as reported by Adamu and Audu (2008).

Sažetak

ODABRANI JETRENI I BUBREŽNI BIOKEMIJSKI PROFILI HIBRIDNOG SOMA IZLOŽENOG LISNOM PRAHU *JATROPHA CURCAS*

Ova studija istražuje učinak subletalnih koncentracija (7,50, 5,00, 2,50 i 0,00 g\L (kontrola)) lisnog praha *Jatropha curcas* na neke jetrene i bubrežne biokemijske profile, kao što su ukupni protein, ukupni bilirubin, ukupni albumin i ukupni globulin hibridnog soma (*Clarias gariepinus* (♀) i *Heterobranchus bidorsalis* (♂)), nakon četrnaestodnevnog eksperimentalnog razdoblja. Rezultat se značajno razlikovao u srednjoj vrijednosti određenog jetrenog ukupnog bilirubina, dok se ukupni protein, albumin i globulin nije značajnije razlikovao u promatranoj ribi u usporedbi s kontrolnom. Bubreg je pokazao promjenjivi raspon malih razlika ($p>0,05$) u razini ukupnog proteina, ukupnog albumina, ukupnog bilirubina i ukupnog globulina. Prema tome, ovim se istraživanjem utvrdilo da subletalne koncentracije *J. curcas* nemaju učinka na osnovne funkcije određenih biokemijskih profila hibridnog soma te da su promjene izravno proporcionalne koncentracijama *J. curcas*.

Ključne riječi: *Jatropha curcas*, hibridni som, jetra, bubreg, biokemijski profil

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