EXTRACTION OF TOMATO LYCOPENE AND ITS EFFECTS ON ELECTROLYTE BALANCE, KIDNEY AND LIVER FUNCTION OF RABBIT BUCKS

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ABSTRACT
Lycopene is an antioxidant carotenoid compound occurring in plants and is responsible for the red coloured pigment of tomato, watermelon and other fruits mostly red coloured. In this study lycopene was extracted from tomato using acetone and dichloromethane as extraction solvents. Identification of lycopene was carried out using UV-Visible spectrophotometer. It was further fed to 15 mongrel rabbit Bucks in five graded levels at 0.00, 0.25, 0.50, 0.75 and 1 gram using a completely randomized design for a period of 8 weeks (56days) to determine its effects on electrolyte balance (sodium, potassium, chloride and bicarbonate), kidney function (uric acid, urea nitrogen and creatinine) and liverfunction (total protein, albumin, total bilirubin, alkaline phosphatase, aspartate aminotransferase and alanine aminotransferase). The results showed that the peaks of lycopene in UV-Visible spectrophotometer were found at 475nm and 470.5nm in chloroform and hexane respectively. Also, all parameters tested for electrolyte balance, kidney and liver function were within the normal range. From the results of this study it was concluded that lycopene did not have any adverse effects on electrolyte balance, kidney and liver function but it was well tolerated by these delicate organs. This study therefore recommends that lycopene can be used an antioxidant in rabbit Bucks to reduce oxidative stress.
Keywords: Lycopene, Tomato, Antioxidant, Electrolyte, Kidney, Liver

INTRODUCTION
Rabbits have an important role in the supply of animal protein for humans. Rabbit occupies a vital midway position between ruminants and non-ruminant animals and can effectively utilise feed in rations containing less than 20% grain (Saleh *et al.*, 2010). Male rabbits are undoubtedly the basis of reproductive success, given that farm profiability depends not only on the female's fertility but also upon the welfare of the buck which is subject to stress factors such as oxidative stress. Reactive oxygen species (ROS) are the major cause of oxidative stress and are continuously produced during metabolism.Indeed, when produced in excess and not tightly kept under control by the antioxidant "detoxifying" systems, ROS can potentially damage all components of the cell (Jean, 2012). Lycopene is a carotenoid that has antioxidant properties and imparts the red-colored pigment in both fruits and vegetables, it is a fat soluble substance. Epidemiological research studies have demonstrated positive benefits in consuming lycopene. Fruits and vegetables that are high in lycopene include tomatoes, watermelon, pink guava and red pepper. The lycopene in tomato is considered to possess the highest antioxidant activity. The antioxidant property of lycopene is highlighted by its capacity to sequester and its ability to trap reactive oxygen species (ROS) (Amany *et al.*, 2009). This study was therefore designed to determine the safety of lycopene administered to rabbit Bucks by evaluating its effects on electrolytes balance, kidney and liver function.

MATERIALS AND METHODS
This study was carried out at Abubakar Tafawa Balewa University Teaching and Research Farm, Bauchi, Bauchi state, Nigeria. It is located in the north eastern part of the country and has a tropical climate. Lycopene used for this experiment was obtained by extraction from tomato which was procured from an established market in Bauchi metropolis. Fifteen mature mongrel rabbit Buckswere randomly assigned to different treatments in a complete randomized design. The experiment lasted for a period of 12 weeks comprising 4 weeks of stabilization and acclimatization and 8 weeks for oral administration of graded levels of tomato lycopene.

Extraction and packaging of Lycopene from Tomato
Lycopene was extracted using Acetone and Methylene chloride (Dichloromethane) following the method of Gordon *et al* (2007) with some slight modifications.
The following steps were followed:-

- Tomato was washed in a basket with running water only
- It was milled into paste with the aid of a petrol powered 5.5 horse power grinding mill, it was heated and allowed to simmer for five minutes.

- After simmering for five minutes it was poured into a cheese cloth which was suspended above ground level and allowed to drain until 10 percent of the initial weight was attained.
- Tomato paste was processed in batches of 1kg at a time in 1.4 litre of acetone (This was done to dehydrate the tomato paste)
- It was stirred until no longer gummy and then Filtered using a cheese cloth
- The resultant solid material was Placed in 1 litre of Methylene Chloride and then
- Filtered with the aid of 110mmØ whatman paper
- The filtrate was allowed to evaporate at room temperature leaving behind lycopene at the base of the beaker.
- It was then subjected to light spectrophotometric authentication in selected solvents such as Hexane and chloroform
- It was further graded into 0.25gram, 0.50gram, 0.75gram and 1gram
- Wheat flour was used as excipient which was mixed thoroughly with lycopene and finally packaged in capsules.

Blood Collection

Blood was collected from each buck through the marginal ear vein (ear veinipuncture) every fortnight (two weeks) to examine major electrolytes, liver and kidney function.

RESULTS AND DISCUSSION

Figure1 and 2 below shows that the maximum absorbance of lycopene in chloroform and hexane was at 475nm and 470.5nm respectively, this very closely coincides with the three peaks characteristics of Lycopene at λ = 446nm, 472nm and 505nm (Monica, 2011).

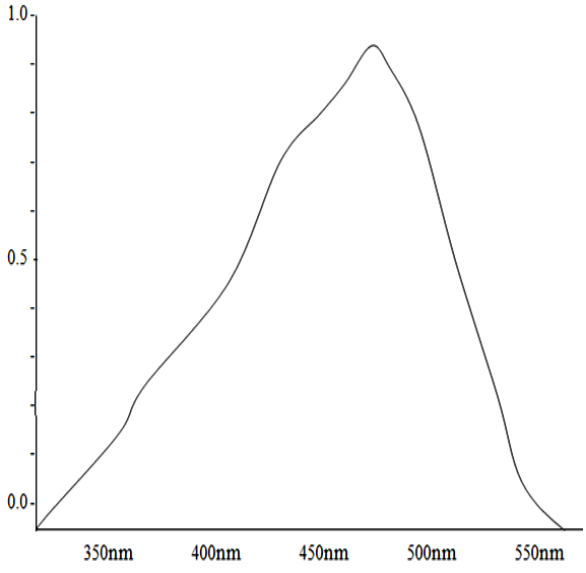

Fig1: Absorbance Spectrum of Lycopene in Chloroform

nm (nanometre)

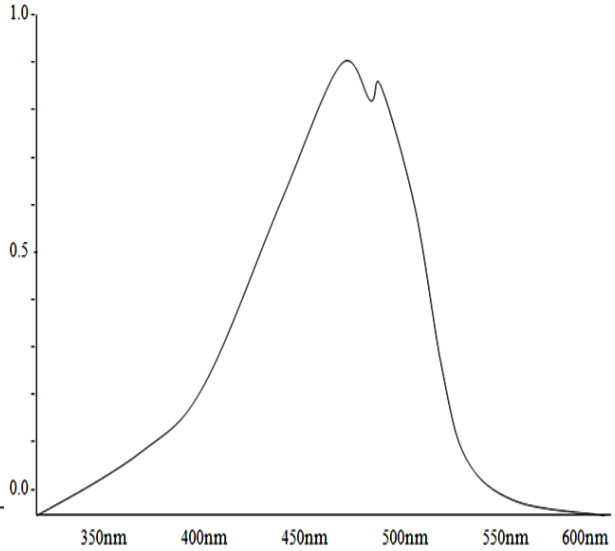

Fig2: Absorbance Spectrum of Lycopene in Hexane

nm (nanometre)

Electrolytes Balance

The role of balanced electrolytes in male animals cannot be overemphasised. In this study sodium, potassium, chloride and bicarbonate were tested. The results in table 1 shows that all the electrolytes were within the normal range except for bicarbonate in treatment one (T1) which is the control group with no lycopene administered. This indicates that lycopene helped to maintain electrolytes balance.

Table 1: Electrolyte balance of rabbit Bucks administered graded levels of lycopene

Parameters	T1(0.00)	T2(0.25)	T3(0.50)	T4(0.75)	T5(1.00)
	Lycopene				
Sodium (140-160 mmol/L)	153.91±0.88	152.67±3.06	155.58±5.78	154.92±3.79	152.17±2.31
Potassium (4.3-5.8 mmol/L)	4.88±0.06	5.15±0.1	4.9±0.53	4.87±0.32	4.91±0.68
Chloride (90-110 mmol/L)	105±0.47	101.42±1.73	97.67±8.50	98.83±6.24	105.67±4.73
Bicarbonate (16-32 mmol/L)	33.66±3.95#	24.67±5.03	22.33±5.29	27.83±4.04	20.00±2.08

Means that it is outside the normal range

(mmol/L) Millimoles Per Litre

Liver Function

A loss of liver function will result in retaining waste metabolites in the body, there by affecting performance, in view of this, total protein, albumin, total bilirubin, alkaline phosphatase (ALP), aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were examined to determine the effect of lycopene on the functionality of the liver. The result in table 2 showed that lycopene did not affect liver function as all the parameters tested were within the normal range. This is also an indication that lycopene was well tolerated and metabolised by the liver without affecting its functionality.

Kidney Function

The kidneys are responsible for maintenance of homeostasis which directly impacts performance of rabbit Bucks, in this study parameters such as uric acid, urea nitrogen and creatinine were examined. Table 3 shows that all parameters tested were within the normal range at all lycopene levels. From the results therefore, it can be deduced that lycopene did not adversely affect the kidneys but rather maintained the functionality of the kidneys.

Table 2: Liver function of rabbit Bucks administered graded levels of lycopene

Parameters	T1(0.00)	T2(0.25)	T3(0.50)	T4(0.75)	T5(1.00)
	lycopene				
Total Protein (5.4-7.3g/dl)	6.12±0.75	5.77±0.26	6.34±0.29	6.14±0.15	6.40±0.60
Albumin (2.4-4.5g/dl)	3.98±0.21	3.71±0.29	3.52±0.50	3.52±0.38	3.48±0.70
Total Bilirubin (up to1.0 mg/dl)	0.48±0.10	0.61±0.17	0.56±0.06	0.53±0.23	0.59±0.17
ALP (4-20 U/L)	13.42±1.00	11.83±6.66	14.00±0.58	12.25±6.24	13.67±4.04
AST (10-120 U/L)	60.50±28.43	55.17±14.84	49.50±13.28	66.25±9.45	90.08±26.16
ALT (10-45 U/L)	25.50±2.89	24.03±5.51	22.67±16.00	32.42±12.70	22.75±11.06

(mg/dl) milligrams per deciliter (ALP) Alkaline phosphatase
(g/dl) grams per deciliter (AST) Aspartate aminotransferase
(ALT) Alanine aminotransferase

Table 3: Kidney function of rabbit Bucks administered graded levels of lycopene

	Lycopene				
Parameters	T1(0.00)	T2(0.25)	T3(0.50)	T4(0.75)	T5(1.00)
Uric Acid (1.0-4.3 mg/dl)	2.50±0.52	3.55±0.31	3.68±0.76	2.98±0.66	3.13±0.84
Urea Nitrogen (10-33 mg/dl)	22.33±6.66	22.00±6.24	25.08±7.94	21.42±10.82	21.92±6.00
Creatinine (0.5-2.2 mg/dl)	1.24±0.56	1.63±0.15	1.73±0.26	1.51±0.61	1.35±0.31

(mg/dl) milligrams per decilitre

CONCLUSION AND RECOMMENDATIONS

This study has shown that lycopene does not have any adverse effect on the electrolyte balance, kidney and liver function of rabbit Bucks. This is an indication that lycopene up to 1gram is safe to use as an antioxidant to combat oxidative stress and improve overall performance of rabbit Bucks.

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