Research Article

Impact of *Carica papaya* linn. Seed Extraction on Some Marker Enzymes in Male Albino Rats

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ABSTRACT

Administration of papaya seed extract (150mg/kg body wt/day for 15 days) to Wistar male rat through oral route affected the enzymes that involved in the protein metabolism. The protease activity was elevated in glandular system (testes, seminal vesicles and prostate gland) while in duct system, in epididymis it was reduced. The decreased protease activity in both liver and serum indicates that there was no risk of carcinoma by the administration of Carica papaya seed extract. Decrease in ATPase activity could be attributed to androgen dependent parameters, as there was suppression in the energy metabolism. The normal levels of serum Alanine aminotransferase and Aspartate aminotransferase suggest that ethanol extract of *C. papaya* seeds is not a hepatotoxin.

Keywords: Carica papaya, Testes, proteolytic enzymes, ATPase, hepatotoxin.

INTRODUCTION

Among plant products so far tested for male anti-fertility, seeds of *Carica papaya* have shown promise in male contraception. Crude extracts of *Carica papaya* seed have an anti-fertility effect on male rats, rabbits and monkeys[1]. Udoh et al., (2005)[2] also showed that oral administration of *C. papaya* seed extract could induce reversible male infertility. Therefore CPL seed extract could be used for the pharmaceutical development of a male contraceptive. This abortifacient property will only occur when high doses of the extract are given[3]. Chinoy *et al* (1985)[4] have also shown that intramuscular administration of aqueous Carica papaya seed extract in 0.5 and 5.0 mg/kg body weight/day for 7 days caused a selective androgen deprivation effect on reproductive organs resulting in infertility with complete reversibility on withdrawal of the treatment. These results indicate that reversible sterility could be induced in male rats by papaya seeds aqueous extract treatment without adverse effects on libido and toxicological profile.

Oral administration of crude aqueous Carica papaya seed extract in doses of 10 and 50 mg/animal/day orally for 30, 60 and 90 days and 0.1 and 1.0 mg/animal/day intramuscularly for 15 and 30 days in male albino rats caused a significant reduction in cauda epididymal sperm motility, count and fertility rate which returned to normal on cessation of the treatment[5]. It has been reported that protein level is directly correlated with the secretory activity of the testis and accessory glands, which in turn depends on the androgen levels[6&7]. The most pronounced general metabolic action of the androgen is the promotion of protein anabolism[8]. Hence the present study was undertaken in order to estimate different enzyme activity levels in protein metabolism. Thus, the present study was aimed to know the effect of Carica papaya seed extraction on proteolytic enzymes that are involved in metabolism.

MATERIALS AND METHODS

Healthy adult male Wistar strain albino rats (90days old, weight 180±10g) were administered with 150mg/kg body wt/day of ethanol extract of Carica papaya Linn. seed orally for 15days. The ethanol extract was prepared according to WHO protocol CG-04, 1983[9]. Seeds were shed-dried, powdered and extracted with 95%ethanol (v/v) at 55-60°C for 3h. The solvent was distilled off under reduced pressure; the resulting mass was dried under vacuum and kept at 24°C until use. The control animals were given normal saline or sterile distilled water. Both control and experimental rats were maintained in standard air conditioned animal house at a temperature of 25±2°C, photoperiod of 12 hours light and 12 hours dark cycle, with a relative humidity of 50 ± 5% and fed on standard rat feed obtained from Hindustan Lever Ltd., Mumbai, India. The usage of animals was approved by the Institutional Animal Ethics resolution number 18/2012-2013/ (i)/a/CPCSEA/IAEC/SVU/CC-SHB Dt: 01-07-2012.

Twenty four hours after the last dose, the animals were autopsied. The tissues like testes, epididymis, seminal vesicle, prostate gland, liver and blood were isolated; serum was collected immediately from blood and used for biochemical analysis. The protease activity[10], ATPase activity[11], Alanine amino transferase and Aspartate amino transferase activities[12] were estimated in control and experimental rat tissues.

RESULTS AND DISCUSSION

The data represented in table 1 shows the levels of protease activity, ATPase activity, AlAT activity and AAT activity in reproductive tissues of control and treated rats. The data represented in table- 2 indicates the levels of protease activity, ATPase activity, AlAT activity and AAT activity in liver and serum of control and treated rats.

A protease (also termed peptidase or proteinase) is any enzyme that conducts proteolysis, that is, begins protein

catabolism by hydrolysis of the peptide bonds that link amino acids together in the polypeptide chain forming the protein. Proteases determine the lifetime of other proteins playing important physiological role like hormones, antibodies, or other enzymes this is one of the fastest "switching on" and "switching off" regulatory mechanisms in the physiology of an organism.

The protease activity was elevated in glandular system (testes, seminal vesicles and prostate gland) while in duct system, in epididymis it was reduced. This may be due to the reduced proteins in glandular system and elevated proteins in the duct system[13]. Thus the protein biosynthesis machinery was inhibited in glandular system[14]. The decreased protease activity was observed in both liver and serum but the extent of decrement is more in serum. It was found that the patients with pancreatic carcinoma have increased concentration of protease activity in serum[15]. But in the present study the serum protease activity was reduced. Hence, there was no risk of carcinoma by the administration of Carica papaya seed extract.

The ATPase activity levels were decreased in all tissues indicates the suppression in the energy metabolism[16]. ATP is the source of energy for sperm motility. ATP is hydrolyzed by ATPase activity, Extracellular adenosine 5'-triphosphate alters motility and improves the fertilizing capability of mouse sperm[17&18]. Motility of spermatozoa is due to the flagellar beat, which is dependent on the micro tubular apparatus of the flagellum and adenosine triphosphate (ATP)[19].

Table 1: Enzymatic profiles in reproductive tissues of Control and Papaya seed extract treated rats.

S. No	Parameter	Control, seed extract, % change and significance			
		Testis	Epididymis	Seminal vesicles	Prostate gland
1.	Protease activity (µ moles of tyrosine /mg protein/hr)	0.589±0.011 0.719±0.056 +22.07*	0.827±0.075 0.659±0.048 -20.31*	0.913±0.06 1.185±0.05 +29.79*	1.038±0.09 1.315±0.08 +26.68*
2.	ATPase activity (µ moles of Pi formed/mg protein/hr)	16.41±1.13 11.57±0.96 -29.49*	14.77±1.09 11.45±1.08 -22.47*	12.78±1.14 9.15±0.85 -28.40*	15.24±1.18 10.62±0.91 -30.31*
3.	AlAT activity (µ moles of sodium Pyruvate formed/mg protein/hr)	0.950±0.05 0.623±0.03 -34.42*	0.982±0.06 0.589±0.02 -40.02*	0.761±0.03 0.612±0.02 -19.57*	0.837±0.06 0.638±0.04 -23.77*
4.	AAT activity (µ moles of sodium Pyruvate formed/mg protein/hr)	0.762±0.05 0.531±0.03 -30.31*	0.945±0.07 0.754±0.04 -20.21*	0.634±0.03 0.418±0.02 -34.06*	0.733±0.05 0.542±0.02 -26.05*

Mean± SD of six individual observations; + and – indicates percent increase and decrease respectively over control; *P<0.001 the level of significance.

Table 2: Enzymatic profiles in Liver and Serum of Control and Papaya seed extract treated rats.

S. No	Parameter	Control, seed extract, % change and significance	
		Liver	Serum
1.	Protease activity (µ moles of tyrosine /mg protein/hr)	1.171±0.05 1.085±0.08 -7.34***	1.153±0.09 0.921±0.07 -20.12*
2.	ATPase activity (µ moles of Pi formed/mg protein/hr)	6.73±0.54 4.14±0.32 -38.48*	66.73±2.18 41.94±1.15 -37.14*
3.	AlAT activity (µ moles of sodium Pyruvate formed/mg protein/hr)	0.932±0.07 0.811±0.06 -12.98*	107.02±8.16 115.32±8.54 +7.75NS
4.	AAT activity (µ moles of sodium Pyruvate formed/mg protein/hr)	0.896±0.06 0.676±0.03 -24.55*	76.53±5.28 75.02±5.26 -1.97NS

Mean±SD of six individual observations; + and – indicates percent increase and decrease respectively over control; *P<0.001, *** P<0.05 the level of significance & NS- non significant changes.

Reduced protein and glycogen content could be correlated with low sperm density[20]. Low levels of fructose could inhibit the sperm motility by deficient generation of ATP[21]. Hence, the reduced ATPase activity levels results the reduction in the energy metabolism which leads to alterations in sperm motility and reduces the fertilizing capacity of the sperm[15].

The declined ATPase activity was noticed in liver and serum also. Liver mitochondria are the major sources of ATP. ATP can be used for active transport within all cells (such as sodium-potassium pumps) for which energy must be used in order to exchange materials. (This is extremely important in the liver since it filters blood and waste materials).

AAT and AlAT are the important aminotransferases and are widely distributed in mitochondrion, which are associated with the integrality of spermatozoa acrosome and cells stress[22].

The levels of Alanine amino transferase and Aspartate amino transferase activity was reduced in all tissues with no significant changes in serum over control[23]. Alanine aminotransferase (AlAT) and aspartate amino transferase (AAT) play an important role in the mobilization of amino acids into gluconeogenesis[23]. Alanine release depends on protein concentration. The observed reduced protein may be the reason for reduced Alanine aminotransferase (AlAT). It is also possible that changes in hormone levels affect the release of Alanine[24] .

Alanine aminotransferase (AlAT) and aspartate amino transferase (AAT) play an important role in the mobilization of amino acids into gluconeogenesis in the liver and kidney[23]. The plasma AAT and AlAT activities are markers of hepatocellular damage[25].

The serum levels of Alanine aminotransferase (AlAT) and Aspartate aminotransferases (AAT) are usually elevated in conditions associated with injuries or diseases affecting the liver which leads to the release of these hepatocellular enzymes into the bloodstream[26]. The reduced levels of these enzymes and hematological indices in the treated Wistar rats implies that the ethanol extract of *C. papaya* seed is generally free of anti-metabolic properties which is consistent with the findings of other researchers.[27, 28] Hence, the normal levels of serum Alanine aminotransferase and Aspartate aminotransferase suggest that ethanol extract of *C. papaya* seeds is not a hepatotoxin.

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