Asian Journal of Medical Sciences 3(1): 8-13, 2011
ISSN: 2040-8773

Received: September 06, 2010 Accepted: October 19, 2010 Published: February 25, 2011

Effects of Vitamins A, C and E on Liver Function in Pregnancy

[1]O.I. Iribhogbe, [1]J.E. Emordi, [1]A. Aigbiremolen, [2]O.B. Idonije, [3]E.O. Nwoke and [3]U. Akpamu
[1]Department of Pharmacology and Therapeutics,
[2]Department of Chemical Pathology,
[3]Department of Physiology, College of Medicine,
Ambrose Alli University, Ekpoma Edo State Nigeria

Abstract: In a bid to investigate the effect of sub chronic administration of vitamin A, C and E on liver function in early pregnancy, eighty-five female Wister rats (255-300 g) were employed for the study. They were randomly assigned to three test groups (I, II and III) having 5 treatment subgroups with five rats each, and a control and vehicle group with five rats each. After allowing the female rats to mate with male rats for 6 days, pregnancy was confirmed. The control group was administered 1ml of distilled water, vehicle group 1ml of tween 80, while test groups I, II and III received varying doses of vitamin A, C and E respectively via the intragastric route for 11days. The results showed a significantly elevated albumin, total protein, AST and ALP levels ($p<0.05$), following vitamin C and vitamin A administration when compared with control. In the vitamin A treated group there was a dose dependent significant increase ($p<0.05$) in total protein, AST and ALP levels while vitamin E treated group presented a significantly reduced ALT ($p<0.05$) with no significant change in total protein levels when compared with control. Conclusively the therapeutic benefit of vitamin A, C and E supplementation in pregnancy is not without an impact on the functional and cellular integrity of the liver.

Key words: Cellular integrity, liver function, pregnancy, vitamin A, vitamin C, vitamin E

INTRODUCTION

Pregnancy is a physiological condition associated with many changes in the internal milieu of the body, in which more and more stress is being laid on the biochemical changes of the body (Kashinakunti *et al*., 2010). While a number of associations between hepatic dysfunction and pregnancy exist (Hunt and Sharara, 1999), reports show that the physiological changes in liver function during pregnancy are commonly transient and rarely permanent (Jamjute *et al*., 2009). Disorders arising in pregnancy, such as pre-eclampsia and eclampsia, acute fatty liver of pregnancy (AFLP), haemolysis, elevated liver enzyme and low platelets (HELLP) syndrome, cholestasis, hyperemesis gravidarum and isolated cases of raised liver enzymes can have serious implications (Jamjute *et al*., 2009).

The liver serves multiple functions viz a viz the biotransformation of insoluble compounds (e.g., drugs, toxins, bilirubin), the metabolism and excretion of cholesterol and bilirubin, the production of plasma proteins (e.g., albumin, coagulation factors, alpha- and beta-globulins, transferrin, haptoglobin), the metabolism of amino acids, carbohydrates and lipids (Hunt and Sharara, 1999) and the storage of blood, vitamins and iron (Guyton and Hall, 2006). The diagnostic work-up of abnormal Liver Function Test (LFT) is challenging, as the conditions peculiar to pregnancy have to be considered in addition to the causes affecting the non-pregnant population (Riely, 1994; Knox, 1998). The spectrum of disease is varied and abnormal LFT can be mild with no long-term consequences, or it can be severe, leading to both maternal and fetal mortality (Riely, 1999).

More recently, the observation that women with pre-eclampsia have decreased plasma and a placental concentration of antioxidants (Hubel *et al*., 1997; Wang and Walsh, 1996) has led to the proposal that placental under perfusion may mediate a state of oxidative stress (Rumbold *et al*., 2008). It has been suggested that deficiency in antioxidant vitamins would be associated with the development of pregnancy complications like pre-eclampsia, eclampsia (Hubel *et al*., 1997; Erkkola, 1997). These observations have given rise to increased interest in antioxidants (Kashinakunti *et al*., 2010). In this context, this study was undertaken to investigate the impact of antioxidant vitamins A, C and E on liver biomarkers in early pregnancy.

MATERIALS AND METHODS

Experimental animals: Eighty- five adult female Wister albino rats weighing (225 - 300 g) were obtained from the

Corresponding Author: O.I. Iribhogbe, Department of Pharmacology and Therapeutics, College of Medicine, Ambrose Alli University Ekpoma, Edo State Nigeria. Tel: +2348065794437

Table 1: Effect of vitamin A, C and E in pregnancy on liver functionality

	Albumin			Protein		
Group	Vit. A	Vit. C	Vit. E	Vit. A	Vit. C	Vit. E
Control	2.92±0.16	2.92±0.16	2.92±0.16	7.18±0.12	7.18±0.12	7.18±0.12
Tween 80	3.08±0.17	-	3.08±0.17	7.00±0.10	-	7.00±0.10
T1	3.00±0.10	4.06±0.15*	2.96±0.15	8.28±0.37*	7.20±0.14	7.36±0.12
T2	3.14±0.11	6.00±0.23*	3.80±0.22*	8.34±0.48*	7.58±0.33	7.00±0.13
T3	3.80±0.43*	6.70±0.15*	4.22±0.13*	8.76±0.21*	8.50±0.16*	6.80±0.17
T4	4.24±0.36*	7.26±0.16*	5.16±0.12*	9.18±0.40*	8.80±0.26*	6.72±0.17
T5	4.60±0.46*	7.78±0.17*	6.48±0.17*	9.64±0.70*	9.64±0.34*	6.80±0.29

Values are mean±SEM (g/dL); Vit. = Vitamin; T = Treatment; *: $p<0.05$ with control

Animal House, College of Medicine, Ambrose Alli University, Ekpoma between August and October 2009. They were housed in a stainless steel cage with plastic bottom grid and a wire screen top in physiology Lab II in the Department of Physiology, Ambrose Alli University, Ekpoma, Edo State, Nigeria. They were assigned into five groups; a control group (n = 5), vehicle group (n = 5) and three test groups (I, II and III) made up of five sub-groups (n = 5). They were fed *ad libitum* with tap water and pellated feeds purchased from Bendel feeds and flour meal Ewu, Nigeria Limited and allowed to acclimatize for 2 weeks. After which two male Wister albino rats were introduced into each group to allow for mating. The animals were allowed to mate for 6 days after which the male animals were removed from the cage. Pregnancy was confirmed using the palpation method as described by Agematsu *et al*. (1983) and vaginal smear microscopy method (Long and Evans, 1922; Daly and Kramer, 1998). From the 7th day, administration of the different Vitamins began using orogastric tubes and syringes to minimize the loss of test substance (Ejebe *et al*., 2009). This lasted for a period of 11 days. The administrations were conducted between the hours of 08.00 am and 10.00 am daily.

Vitamins preparation: Vitamin A, C and E were purchased from Clarion Medical Pharmaceuticals Nigeria Limited. Tween 80 vehicle was purchased from Sigma Pharmaceuticals Limited. 200 mg of the powdered form of vitamin C was dissolved in 10mls of distilled water and the appropriate dose per kg were prepared for administration. Vitamin A (25,000 IU equivalent to 6 mg retinol and vitamin E, 100 mg) was dissolved in 0.2 mL of tween 80 and water in a ratio of 0.2:0.2:9.6.

Vitamin administration: In addition to normal feed, group II received Vitamin C as follows; 200 mg/kg, 250, 300, 350 and 400 mg/kg for treatment sub- groups 1, 2, 3, 4 and 5, respectively. In addition to normal feed, group I and III received Vitamin A and E dissolved in tween 80 respectively as follows; 0.6, 0.7, 0.8, 0.9 and 1.0 mg/kg of vitamin A and 16.4, 18.4, 19.4, 20.4 and 22.4 mg/kg of vitamin E administered to treatment sub- group 1, 2, 3, 4 and 5, respectively.

Sample collection: Twenty-four hours after the last administration of vitamins was carried out, the animals were sacrificed after inhalation of chloroform. Cardiac and jugular vein puncture were used to collect blood samples into tubes containing EDTA as anticoagulant and centrifuged plasma preparation was assayed for biomarkers of liver function using standard laboratory procedures.

Enzymatic assays: Determination of plasma albumin (ALB) and total protein (TP) for liver functionality, aspartate amino transferase (AST) and alanine amino transferase (ALT) for cellular integrity of the liver and alkaline phosphatase (ALP) for condition linked to the billary tract were analysed using standard methods as described below;

Albumin and total protein determination: In the plasma samples, the levels of albumin (ALB) and total protein (TP) were determined using a Technicon RA-XT autoanalyzer (Karakilcik *et al*., 2005).

Alanine and aspartate aminotranferase determination: Plasma assays for tests on the function of liver viz-a-viz serum aspartate amino transferase (AST) and alanine amino transferase (ALT) activities were estimated with the Randox reagent kit using 2,4-dinitrophenylhydrazine as substrate according to the method described by Reitman and Frankel (1957).

Alkaline phosphatase determination: Alkaline phosphatase (ALP) activity was determined with the Randox reagent kit using the p-nitrophenylphosphate as substrate according to the method described by Bassey *et al*. (1946).

Data analysis: The mean±standard error of mean (X±SEM) and one-way ANOVA (LSD) statistical test was performed using SPSS version 17 soft ware. The significance level was set at $p<0.05$.

RESULTS

Liver functionality: There was a significant elevation of albumin levels following supplementation with vitamin A, C and E ($p<0.05$). This was however sustained in the vitamin C treated group. In the vitamin A and E treated group, significant elevation occurred after the 2nd and the 1st treatment respectively (Table 1). Total protein was significantly elevated in the vitamin A and C treated

Table 2: Effect of vitamin A, C and E in pregnancy on liver cellular integrity

	Aspartate amino transferase			Alanine amino transferase		
Group	Vit. A	Vit. C	Vit. E	Vit. A	Vit. C	Vit. E
Control	266.00±1.64	266.00±1.64	2.66.00±1.64	88.00±0.89	88.00±0.89	88.00±0.89
Tween 80	264.60±3.56	-	264.60±3.56	89.60±0.68	-	89.60±0.68
T1	259.80±6.58	265.6±3.71	244.00±2.07*	106.60±1.94*	101.80±1.24*	104.00±1.92*
T2	261.6±2.79	257.80±3.28	253.80±2.35*	115.20±5.27*	107.80±4.60*	108.00±4.74*
T3	264.00±1.45	244.00±2.77*	258.20±2.56*	122.00±2.49*	137.00±7.41*	111.80±4.32*
T4	276.40±6.34	232.20±6.73*	263.40±1.89	121.40±2.93*	152.00±2.95*	116.0±4.42*
T5	303.40±2.64*	212.00±4.21*	270.20±1.85	128.60±3.65*	169.80±3.65*	126.20±11.30*

Values are mean±SEM (IU/L); Vit. = Vitamin; T = Treatment; *: p<0.05 with control

Table 3: Effect of vitamin A, C and E in pregnancy on biliary tract integrity

	Alkaline phosphatase		
Group	Vit. A	Vit. C	Vit. E
Control	88.00±1.00	88.00±1.00	88.00±1.00
Tween 80	87.80±2.65	-	87.80±2.65
T1	130.00±1.64*	135.40±3.43*	159.60±5.73*
T2	145.00±1.52*	139.60±1.83*	137.40±3.88*
T3	149.00±1.00*	146.00±2.72*	127.00±7.99*
T4	149.00±1.92*	147.20±3.14*	104.00±1.97*
T5	171.2±3.81*	157.8±3.23*	91.40±2.99*

Values are mean±SEM (IU/L); Vit. = Vitamin; T = Treatment; *: p<0.05 with control

group, however, this occurred after the third treatment in the vitamin C treated group (8.50±g/dL) and was sustained through out the duration of treatment in the vitamin A treated group (p<0.05), when compared with control. There was no significant different (p>0.05) compared with the control in the vitamin E treated group.

Cellular integrity of the liver: Table 2, revealed a significant reduction in alanine transaminase activity in the vitamin C and E treated groups (p<0.05) compared with the control. This occurred after the second treatment in vitamin C group and was sustained in the vitamin E group. This was not the case in the vitamin A group, which appeared unchanged through out treatment except for the significant elevation of ALT (303.4± IU/L) after the fifth treatment (p<0.05) when compared with control. However, AST was markedly elevated in all the groups that received vitamin supplementation (p<0.05).

Biliary tract integrity: ALP was markedly elevated in all the vitamin treated groups, this was significant when compared with control (p<0.05), however the elevation of ALP was only sustained in the vitamin A and C treated groups. In the vitamin E treated group (Table 3) there was an initial marked elevation of ALP (200.4±IU/L), which was not sustained, though subsequent values remained significantly elevated when compared with control (p<0.05).

DISCUSSION

While antioxidant vitamins have been reported to play an important role in the regulation and eventual outcome of human pregnancy (Dakshinamuti and Dakshinamuti, 2001), scarce studies have link antioxidant vitamins to abnormal liver function in pregnancy. Liver enzymes are marker enzymes for liver function and integrity (Jens and Hanne, 2002; Adaramoye *et al*., 2008). These enzymes are usually raised in acute heapto-toxicity or mild hepato-cellular injury, but tend to decrease with prolonged intoxication due to damage to the liver (Cornelius, 1979; Jens and Hanne, 2002).

In this study, administration of vitamin A produces increase in plasma Alb, TP, ALT, APT and ALP activities. Comparatively, with the control, this increase was statistically significant in the entire treatment in ALP, APT and TP, in 3rd to 5th treatments in Alb, and conversely non significant different in plasma ALT activity except for the 5th treatment. Thus, this present available data suggest that vitamin A supplementation in pregnancy exerts possible hepatotoxic effect as the significant increase in Alb, ALP, APT and TP suggest liver damage. Vitamin A is a known hepatotoxin and multiple cases of hepatotoxicity have been documented with the ingestion of vitamin A in high dosages, usually more than 100,000 IU per day, and rare cases have occurred with dosages of approximately 25,000 IU per day (Riley and Bhatti, 2001). Furthermore, Riley and Bhatti (2001) reported the degree of liver injury associated with vitamin A to depends on the dose. This is true in pregnancy supplementation, as justified by the plasma activity of ALP, APT, TP and Alb in this study. Alcohol potentates the hepatotoxicity of this vitamin as vitamin A can cause steatosis, perisinusoidal fibrosis, chronic hepatitis and cirrhosis (Bashir and Lewis, 1995).

From the result of this study, except for ALT which reduces as dose increases, vitamin C supplementation in pregnancy significantly increases plasma ALB, TP, APT and ALP. This effect was more potent with increase dose of vitamin C; however, at a very low dose, vitamin C (as ALT in 1st treatment and TP in the 1st and 2nd treatments) was not significantly different with the control. Of interest are the findings which reported, vitamin C may partially prevent certain types of hepatic cellular damage (McDowell, 1989; Parola *et al*., 1992; Sies *et al*., 1992; Burtis and Ashwood, 1994; Netke *et al*., 1997).

Report on the effect of vitamin E on liver function in pregnancy is lacking. However, an article by *Sarah Ince in e*How (2010) reported Vitamin E to helps improve liver function by protecting Vitamin A. Furthermore, the report

stated that 'when the two Vitamins (Vitamin A and E) are combined, a potent liver tonic is achieved and that both vitamins work together to strengthen and rejuvenate a tired liver. This present study presented a non significant change ($p>0.05$) in protein and a reducing ALP (which is non significant at the 5th treatment) with vitamin E when compared with control. While albumin is favoured at a very low dose (1st treatment) with vitamin E, AST is favoured at high dose (4th and 5th treatment).

Abnormal liver tests occur in 3-5% of pregnancies, with many potential causes, including coincidental liver disease (most commonly viral hepatitis or gallstones) and underlying chronic liver disease (Hay, 2008). Furthermore, there have been numerous publications on the specific causes of abnormal liver function test in pregnancy (Knox and Olans, 1996; Castro *et al*., 1999; Davidson, 1998; Hunt and Sharara, 1999; Sabai *et al*., 1993). According to Hay (2008), most liver dysfunction in pregnancy is pregnancy-related and caused by 1 of the 5 liver diseases unique to the pregnant state: these fall into 2 main categories depending on their association with or without preeclampsia. An increased risk of fetal loss has been noted in pregnant patients with chronic liver disease (Lee, 1992). While pregnancy are accompany with elevated liver enzymes, however the cause need to be examined as Smoleniec and James (1993) reported viral hepatitis to be the most common cause of jaundice in pregnancy. The course of most viral hepatitis infections (e.g., hepatitis A, B, C and D) is unaltered by pregnancy (Mishra and Seeff, 1992; Snydman, 1985). However, a more severe course of viral hepatitis in pregnancy has been noted in patients with hepatitis E (a waterborne virus spread through fecal-oral transmission) and disseminated Herpes Simplex Virus (HSV) infections (Riely, 1994; Mishra and Seeff, 1992; Tsega *et al*., 1992; Bile *et al*., 1994; Stagno and Whitley, 1985; Glorioso *et al*., 1996).

Several procedures have been used to protect the liver from damage by administrations of antioxidants such as β-carotene (Olmez and Karakilcik 2004), vitamin C (Mitra *et al*., 1991; Netke *et al*., 1997), vitamin E (Parola *et al*., 1992; Harvey *et al*., 1994; Durak *et al*., 1996; Naziroglu, 1999) and selenium-vitamin E combination (Brucato *et al*., 1986; Sies *et al*., 1992; Naziroglu, 1999). However, this was not in a pregnant condition and thus, the significant of these vitamins in pregnancy on liver biomarker is not guarantee according to this present study.

Conclusively, rather than ingesting antioxidant vitamin in hyper- hepatic enzymes of pregnancy, the underlying cause need to be examined as this study presented significant increases in liver biomarker even in antioxidant vitamins (A, C, E) supplementation in early pregnancy. However, further research in this respect is required.

ACKNOWLEDGEMENT

The authors sincerely appreciate the technical assistance of Dr. Nwaopara, A.O., Head of Department of Anatomy, Ambrose Alli University, Ekpoma, Edo State, Nigeria. and Mrs. Mrs Oruware the Head of Animal Farm, Department of Physiology, Ambrose Alli University, Ekpoma, Edo State, Nigeria., for her assistance in rat procurement and detection of pregnancy. We are also grateful to our individual effort toward the success of this research.

REFERENCES

Adaramoye, O.A., D.O. Osaimoje, M.A. Akinsanya, C.M. Nneji, M.A. Fafunso and O.G. Ademowo, 2008. Changes in antioxidant status and biochemical indices after acute administration of artemether, artemether-lumefantrine and halofantrine in rats. Basic Clin. Pharmacol. Toxicol., 102: 412-418.

Agematsu, Y., H. Ikadai and H. Amao, 1983. Early detection of pregnancy of the rat. Jikken Dobutsu, 32(4): 209-212.

Bashir, R.M. and J.H. Lewis, 1995. Hepatotoxicity of drugs used in the treatment of gastrointestinal disorders. Gastroenterol. Clin. North Am., 24: 937-967.

Bassey, O.A., O.H. Lowry and M.J. Brock, 1946. A method for the rapid determination of alkaline phosphatase with five cubic millimetres of serum. J. Biol. Chem., 164: 321-325.

Bile, K., A. Isse, O. Mohamud, P. Allebeck, L. Nilsson, H. Norder, I.K. Mushahwar and L.O. Magnius, 1994. Contrasting roles of rivers and wells as sources of drinking water on attack and fatality rates in a hepatitis E epidemic in Somalia. Am. J. Trop. Med. Hyg., 51: 466-474.

Brucato, M., S.F. Sundlof, J.U. Bell and G.T. Edds, 1986. Aflatoxin B1 toxicosis in dairy calves pretreated with selenium-vitamin E. Am. J. Vet. Res., 47: 179-183.

Burtis, C.A. and E.R. Ashwood, 1994. Tietz Textbook of Clinical Chemistry. 2nd Edn., WB Saunders Co., Philedelphia, U.S.A., pp: 1275-1512.

Castro, M.A., M.T. Fassett, T.B. Reynolds, K.J. Shaw and T.M. Goodwin, 1999. Reversible peripartum liver failure. A new persective on the diagnosis, treatment and cause of acute liver of pregnancy, based on 28 consecutive cases. Am. J. Obstet. Gynecol., 181: 389-395.

Cornelius, C.E., 1979. Biochemical evaluation of hepatic function in dogs. J. Am. Anim. Hosp. Assoc., 15: 25-29.

Dakshinamuti, K. and S. Dakshinamuti, 2001. Blood pressure regulation and micronutrients. Nutr. Res. Rev., 14: 3-43.

Daly, T.J.M. and B. Kramer, 1998. Alterations in rat vaginal histology by exogenous gonadotrophins. J. Anat., 193: 469-472.

Davidson, K.M., 1998. Intrahepatic cholestasis of pregnancy. Semin Perinatal, 22: 104-111.

Durak, I., T. Guven, M. Birey, H.S. Oztürk, O. Kurtipek and M. Yel, 1996. Halothane hepatotoxicity and hepatic free radical metabolism in guiena pigs: The effects of vitamin E. Can. J. Anaesth., 43: 741-748.

Ejebe, D.E., I.M. Siminialayi, J.O.T. Emudainohwo, S.I. Ovuakporaye, A.E. Ojieh, R. Akonoghrere, I.E. Odokuma and G.C. Ahatty, 2009. An improved technique for oral administration of solutions of test substances to experimental rats using Mediflon/Medicut intravenous cannula. Afr. J. Biotechnol., 8(6): 960-964.

Erkkola, R., 1997. Can pre-eclampsia be predicted and prevented? Acta Obstet Gynecol. Scand, 76: 98-100.

Glorioso, D.V., P.J. Molloy, D.H. Van Thiel and R.J. Kania, 1996. Successful empiric treatment of HSV hepatitis in pregnancy. Case report and review of the literature. Dig. Dis. Sci., 41: 1273-1275.

Guyton, A.C. and J.E. Hall, 2006. Textbook of Medical Physiology, 11th Edn., Philadelphia, Elsevier and Saunders, pp: 859-862.

Harvey, R.B., L.F. Kubena and M. Elissalde, 1994. Influence of vitamin E on aflatoxicosis in growing swine. Am. J. Vet. Res., 55: 572-577.

Hay, J.E., 2008. Liver disease in pregnancy. Hepatology, 47(3): 1067-1076.

Hubel, C.A., A.V. Kozlov, V.E. Kagan, R.W. Evans, S.T. Davidge, M.K. McLaughlin and J.M. Roberts, 1997. Decreased transferrin and increased transferrin saturation in sera of women with preeclamsia. Implications for oxidative stress. Am. J. Obstet. Gynecol., 175: 692-700.

Hunt, C.M. and A.I. Sharara, 1999. Liver disease in pregnancy. Am. Fam. Phys., 59: 829-836.

Jamjute, P., A. Ahmad, T. Ghosh and P. Banfield, 2009. Liver function test and pregnancy. J. Matern. Fetal. Neonatal. Med., 22(3): 274-283.

Jens, J.J. and H. Hanne, 2002. A Review on Liver Function Test. The Danish Hepatitis C. Retrieved from: http://home3.inet.tele.dk/omni/hemochromatosis_iron.htm.

Kashinakunti, S.V., H. Sunitha, K. Gurupadappa, D.S. Shankarprasad, G. Suryaprakash and J.B. Ingin, 2010. Lipid Peroxidation and Antioxidant Status in Preeclampsia. Al-Ameen J. Med. Sci., 3(1): 38-41.

Karakilcik, A.Z., A. Hayat, N. Aydilek, M. Zerin and M. Cay, 2005. Effects of vitamin C on liver enzymes and biochemical parameters in rats anesthetized with halothane. Gen. Physiol. Biophys., 24: 47-55.

Knox, T.A., 1998. Evaluation of abnormal liver function in pregnancy. Semin Perinatol., 22: 98-103.

Knox, T.A. and L.B. Olans, 1996. Liver disease in pregnancy. N. Engl. J. Med., 335: 569-576.

Lee, W.M., 1992. Pregnancy in patients with chronic liver disease. Gastroenterol. Clin. North Am., 21: 889-903.

Long, J.A. and H. Evans, 1922. The oestrous cycle in the rat and its associated phenomena. In: Lipman, F. and R. Hedrick (Eds.), Memoirs of the University of California. Berkeley, University of California Press, CA, 6: 1-148.

McDowell, L.R., 1989. Vitamins in animal nutrition-comparative aspects to human nutrition: Vitamin C. Acad. Press, London, pp: 10-52, 93-131.

Mishra, L. and L.B. Seeff, 1992. Viral hepatitis, A through E, complicating pregnancy. Gastroenterol. Clin. North Am., 21: 873-887.

Mitra, A., A.P. Kulkarni, V.C. Ravikumar and D.R. Bourcier, 1991. Effect of ascorbic acid esters on hepatic glutathione levels in mice treated with a hepatotoxic dose of acetaminophen. J. Biochem. Toxicol., 6: 93-100.

Naziroglu, M., 1999. Protective role of intraperitoneally administered vitamin E and selenium in rats anesthetized with enflurane. Biol. Trace Elem. Res., 69: 199-209.

Netke, S.P., M.V. Roomi, C. Tsao and A. Niedzwiecki, 1997. Ascorbic acid protects guinea pigs from acute aflatoxin toxicity. Toxicol. Appl. Pharmacol., 143: 429-435.

Olmez, D. and A.Z. Karakilcik, 2004. Protective role of β-carotene dietary supplementation on activities of functional liver enzymes and some biochemical parameters in experimental hepatotoxicity of rats. J. Nutr. Biochem., (In press).

Parola, M., G. Leonarduzzi, F. Biasi, E. Albano, M.E., Biocca, G. Poli and M.U. Dianzani, 1992. Vitamin E dietary supplementation protects against carbon tetrachlorideinduced chronic liver damage and cirrhosis. Hepatology, 16: 1014-1021.

Reitman, S. and S. Frankel, 1957. A colorimetric method for the determination of glutamic oxaloacetic and glutamic pyruvic transaminases. Am. J. Clin. Pathol., 28: 56-66.

Riely, C.A., 1994. Hepatic disease in pregnancy. Am. J. Med., 96(1A): 18S-22S.

Riely, C.A., 1999. Liver disease in the pregnant patient. Am. J. Gastroenterol., 94: 1728-1732.

Riley, R.T. and A. Bhatti, 2001. Preventive strategies in chronic liver disease: Part I. alcohol, vaccines, toxic medications and supplements, diet and exercise. Am. Fam. Phys., 64(9): 1555-1561.

Rumbold, A., L. Duley, C.A. Crowther and R.R. Haslam, 2008. Antioxidants for preventing pre-eclampsia. Cochrane Database Sys. Rev., Issue 1. Art. No.: CD004227. DOI: 10.1002/14651858. CD00 4227. pub3.

Sabai, B.M., M.K. Ramadan, I. Usta, M. Salamca, B.M. Mercer and S.A. Friedman, 1993. Maternal morbidity and motality in 442 pregnancies with haemolysis, elevated liver enzymes and low platelets (HELLP syndrome). Am. J. Obstet. Gynecol., 169: 1000-1006.

Sies, H., W. Stahl and A.R. Sundquist, 1992. Antioxidant functions of vitamins. Vitamin E and C, β-carotene, and other carotenoids. Ann. N. Y. Acad. Sci., 669: 7-20.

Smoleniec, J.S. and D.K. James, 1993. Gastro-intestinal crises during pregnancy. Dig. Dis., 11: 313-324.

Sarah Ince, 2010. What Vitamins Help the Liver? eHow, Inc. Retrieved from: www.ehow.com/ds_sarahin 8897.html (Accessed date: June 23, 2010).

Snydman, D.R., 1985. Hepatitis in pregnancy. N. Engl. J. Med., 313: 1398-1401.

Stagno, S. and R.J. Whitley, 1985. Herpesvirus infections of pregnancy. Part II: Herpes simplex virus and varicella-zoster virus infections. N. Engl. J. Med., 313: 1327-1330.

Tsega, E., B.G. Hansson, K. Krawczynski and E. Nordenfelt, 1992. Acute sporadic viral hepatitis in Ethiopia: causes, risk factors, and effects on pregnancy. Clin. Infect. Dis., 14: 961-965.

Wang, Y. and S.W. Walsh, 1996. Antioxidant activities and mRNA expression of superoxide dismutase, catalase, and glutathione peroxidase in normal and preeclamptic pregnancies. J. Soc. Gynecol. Invest., 3: 179-184.