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A study of the effect of curcumin on plasma free radicals in cirrhotic rats using Bile Duct Ligation (BDL)

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ABSTRACT

Cirrhosis of the liver is an important cause of mortality in the world and results in about ten thousand deaths each year. Cirrhosis of the liver, due to its high prevalence and its debilitating and sometimes deadly complications, is one of the main problems in the country's health care system. The method of measuring free radicals was based on 1, 2-diphenyl Pykrya Hydrazyl (DPPH). This combination is based on reviving color from pink to pale yellow in the 520nm absorbance readings. In this study, the mean and standard deviation in sham + curcumin (166.11±10.12), compared with BDL + curcumin (86.53±13.93), are higher and the difference between the two groups is statistically significant (P <0.05).

Key words: Free radicals, curcumin, cirrhotic, 1, 2-diphenyl Pykrya Hydrazyl

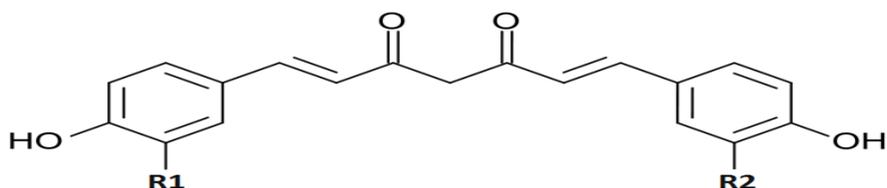
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1. INTRODUCTION

Liver cancer is the fifth most common cancer in cells and the third leading cause of cancer deaths in the world (1). The main risk factors for Hepatocellular carcinoma (HCC) include cirrhosis, liver cirrhosis and age which are common in 80% of patients (2). Cirrhosis of the liver is an important cause of morbidity and mortality throughout the world and leads to around ten thousand deaths each year. Cirrhosis of the liver, due to its high prevalence and its debilitating and sometimes deadly complications, is one of the main problems in the country's health care system. The final outcome of cirrhosis is irreversible cirrhosis and fibrosis. Excessive accumulation of extracellular matrix proteins such as collagen occurs in most patients with chronic liver diseases such as fibrosis. Advanced liver fibrosis results in cirrhosis, liver damage

and increased blood pressure in the portal vein (3). In the US, excessive alcohol consumption and chronic hepatitis C are the most common causes of cirrhosis. Factors leading to cirrhosis include alcoholic liver disease, chronic hepatitis C, non-alcoholic fatty liver disease, autoimmune hepatitis, biliary tract disorders, hereditary diseases, drugs, toxins and infections (4). In a healthy cell, there is a proper balance between prooxidants and antioxidants. Prooxidants increase or decrease antioxidants. Oxidative stress occurs when prolonged, serious harm occurs in cells (5). Liver fibrosis progression due to oxidative stress and free radicals in chronic inflammation is caused by the release of proinflammatory cytokine from Kupffer cells in the liver before the inflammation (6). Curcumin (figure 1) is the main color compound of color compounds which are all derivatives of dicinnamomoylmethane. All therapeutic applications are related to three materials.

- 1) 1,7-Bis-(4-hydroxy-3-methoxyphenyl)-hepta-1,5-dione(C₂₁H₂₀O₆:MW:368)
Diferuloylmetaneor Curcumin
- 2) 1-(4-Hydroxyphenyl)-7-(4-hydroxy-3-methoxyphenyl)-hepta-1,6-diene-3,5 dione(C₂₀H₁₈O₅:MW:338)
p-hydroxycinnamoylferuloylmethanor Demethoxycurcumin
- 3) 1,7-Bis-(4- hydroxyphenyl)-hepta-1,6-dione(C₁₉H₁₆O₄:MW:308)
p,p-dihydroxydicinnamoylmethanor Bis-demethoxycurcumin



- 1) $R_1=R_2=OCH_3$ 2) $R_1=OCH_3, R_2=H$ 3) $R_1=R_2=H$

Figure 1. The overall structure of the main components of turmeric and the place of curcumin in three different functional groups

Free radicals are atoms or molecules having unpaired electrons, and highly reactive in the body that can and damage many of the body's macromolecules such as proteins, lipids, carbohydrates and DNA (7). Increase in free radicals leads to oxidative stress. There are certain systems in the body known as antioxidant defense systems that deal with injuries resulted from free radicals (8). In a healthy person, there is a balance between free radical production and antioxidant defense system. Imbalance in the production of free radicals and antioxidant defense system is called oxidative stress (9).

2. MATERIALS AND METHODS

In this study, 16 Rats were randomly divided into 2 groups of 8 each, control with curcumin treatment (Sham + Cur), and cirrhosis by BDL with curcumin treatment (BDL + Cur). Curcumin was given for suspension in carboxymethyl cellulose 0.5% on the gavage 100 mg / kg / day food during 28 days under optimal conditions, temperature and moisture. After 28 days, rats were anesthetized with ether, liver tissue was extracted, washed, transferred into an amount of in formalin and was finally sent to the pathology lab to determine the extent of fibrosis

(figure 2(a & b)). Blood samples were taken from cardiac puncture at the same time. The method of measuring free radicals, considering the test, was based on the reaction of 1, 2-diphenyl Pykrya Hydrazyl (DPPH) free radicals (10). This parameter was performed by restoring composition 1, 2-diphenyl Pykrya Hydrazyl (DPPH) according to the method described by Janaszewska et al (11) . First, a total of 20 ml of patient's plasma was solved in 380 ml of phosphate buffer, pH=7.4 and then it was solved in 400 ml of DPPH and incubated for 30 minutes. Inhibition mechanism of polymer chain, R, by DPPH is shown in figure 3. At the end, the obtained absorption was placed in the following equation and the absorption percentage of DPPH was measured.

$$\text{Activity [\% of DPPH reduction]} = [(A - Ax) / A] \times 100\%$$

$$A = \text{DPPH} + \text{methanol}$$

$$Ax = \text{DPPH} + \text{sample}$$

This examination was measured using Eliza Model EPOCH-Bio Tek. Data were analyzed using SPSS software and T- test.

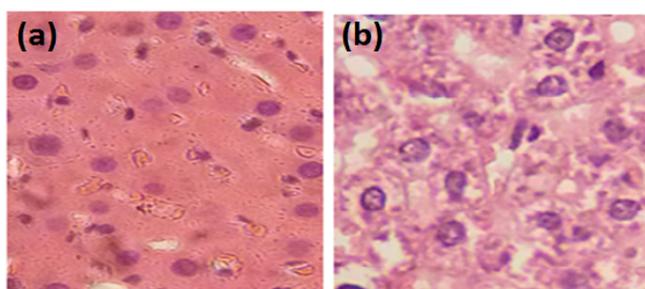


Figure 2. Extracted liver tissue; a) sham+Cur, b) BDL+Cur

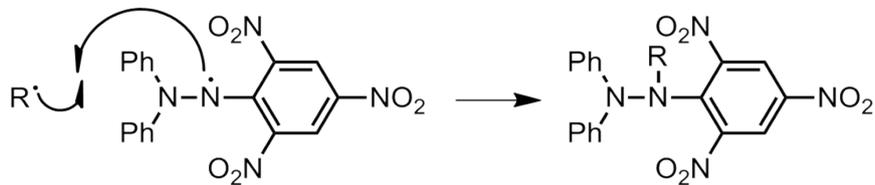


Figure 3. Inhibition of polymer chain, R, by DPPH (12)

3. RESULTS AND DISCUSSION

In this study, the mean and standard deviation in sham + cure (166.11±10.12), compared with BDL + cure (86.53±13.93), were higher and the result is statistically significant (P <0.05) (Diagram 1). Recent studies have shown that apoptotic cells have inhibitory effects on hepatic inflammatory response. According to the findings curcumin material, by regulating the expression of Bcl-2 and Bcl-xL, are effective against necrosis and fibrosis (11, 13). If the damage to the liver tissue is severe, injuries and persistent infection disrupt the normal remodeling process, leading to the accumulation of collagen and fibrosis (13). This study and similar studies have shown that the substance curcumin is able to prevent liver fibrosis in

infected rats. According to previous studies this state of the effect of curcumin on the expression of genes can be created. Curcumin induced apoptosis, by increasing the expression of the P53 gene in the damaged liver cells, reduces the anti-apoptotic gene expression (14). Apoptosis progress not only reduces necrotic cells during chronic liver injury but also inhibits the activation of the inflammatory response and the proliferation of liver cells containing damaged DNA (15). Based on this information, the anti-fibrotic effect of curcumin in the study is due to the effect of this agent in the induction of apoptosis in damaged cells and liver damage responses in the formation of fibrosis in the liver tissue.

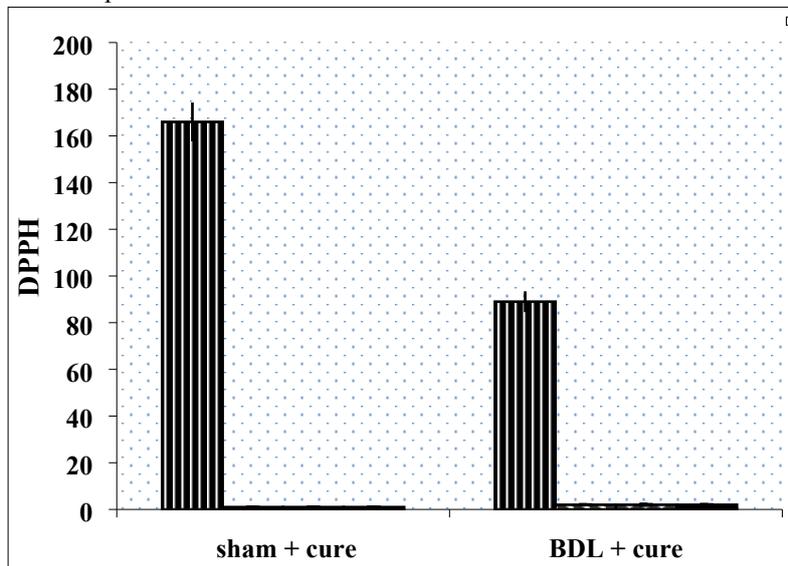


Diagram 1. DPPH level in sham+cure with BDL+cure. * P<0.05. The value is mean±SD

4. CONCLUSION

Curcumin can be used for prophylaxis of liver cirrhosis because it both increases Paraoxonase-1 (PON1) and reduces free radicals. On the other hand, decrease of hepatic parameters (Alkaline Phosphatase, Alanine aminotransferase, Aspartate aminotransferase and direct bilirubin) is increased in cirrhosis. Malondialdehyde (MDA) and total capacity too reduce free radicals in serum and liver tissue in rats.

AUTHORS CONTRIBUTION

This work was carried out in collaboration among all authors.

CONFLICT OF INTEREST

The authors declared no potential conflicts of interests with respect to the authorship and/or publication of this article.

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