Bioremediation of cadmium induced renal toxicity in *Rattus norvegicus* by medicinal plant *Catharanthus roseus*

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**Article Info**

Cadmium is the second most hazardous metals with bio-concentration factor (BCF) > 100. Although WHO permitted cadmium concentration in drinking water is 0.005 mg/L, yet the reality is far above to this limit because of industrial utility of this metal. Oral exposure of cadmium to human results in dreadful symptoms of metabolic disorders especially in liver and kidneys. Endogenous protection could be supported by some exogenous herbal supplement (viz., *Catharanthus roseus* in this case) to overcome the toxic effects. Present Study has been designed to find out the functional renal changes under the effect of cadmium and *Catharanthus roseus* in the model organism albino rats. Cadmium significantly (p > 0.01) increases the level of nitrogenous waste (Urea, BUN, Uric Acid and Creatinin), while decreases the serum protein profile in acute and sub-acute sets. Urea concentration of control ranged from 16.56 to 17.72 mg/dl while that of Group-B and D were 19.84 to 20.87 mg/dl and 17.56 to 17.59 mg/dl respectively. Similarly uric acid concentration ranged in control form 6.98 to 8.01 mg/dl in group-B from 7.58 to 10.25 mg/dl, in Group-D 8.02 to 8.59 mg/dl respectively. Creatinin concentration ranged in control 0.57 to 0.65 mg/dl, in group-B 0.97 to 1.02 mg/dl, in group-D – 0.95 to 0.98 mg/dl respectively.

These results might be due to altered filtration rate of kidney because of protein disruption. The studies conclude the efficient nephro-protection offered by *Catharanthus roseus* extract against Cadmium toxicity.

**1. Introduction**

Heavy metals are the most consistent pollutants of present environment due to their non-biodegradable nature and high bioaccumulation potential (Mohanta et al., 2017). Cadmium is a widespread industrial and environmental pollutant that may cause adverse effects on human beings and animals. Major anthropogenic sources of cadmium exposure to human are mining, electrodes of rechargeable batteries, alloys production, tobacco smoking, electroplating, plastic stabilizers, vapor lamps, engraving, fertilizers and old galvanized PVC water supply pipes (Duan et al., 2016; Stachowiak et al., 2015). In Present Scenario discarded electronic materials and their components have arisen as the biggest source of Cadmium exposure.

Cadmium from various sources percolates through the ground and approaches the drinking water, thus directly enters to food chain. Oral exposure leads cadmium to liver and kidneys for further metabolism and excretion. Thus kidneys are the primary organ inside the body to encounter with cadmium load. Long term chronic exposure of cadmium many result in severe kidney damage to renal failure (Hao et al., 2015). Renal damage primarily alters the filtration rate, thus impaired renal functions could be measured by serum level of nitrogenous waste and proteins. Although endogenous protection mechanism of body overcomes these damages to a vary extent, but it might not be sufficient enough for overflown limits of cadmium exposure. In this study we have tried to establish *Catharanthus roseus* extract to provide exogenous supplement against nephrotoxicity of cadmium.

**Keywords:**

Cadmium  
*Catharanthus roseus*  
Nephrotoxicity  
*Rattus norvegicus*
Catharanthus roseus, the Madagascar periwinkle or common Sadabahar is a perennial herb of family Apocynaceae. Besides its ornamental value, this plant is known to have a number of pharmacological actions of clinical importance like anti-inflammatory, antioxidant and anti-carcinogenic properties (Chen et al., 2017; Van der Heijden et al., 2004). Chemically, Catharanthus roseus pertains over hundred different terpenoid indole alkaloids (TIAs) (Mustafa and Verpoorte, 2007) among which vinblastine and vincristine are well known clinically used anti-carcinogenic alkaloids (Leveque and Jehl, 2007).

2. Material and methods

Eighty adult male albino rats (Rattus norvegicus) of wistar strain were selected from inbred colony. All the rats were of almost same age and weight (100 ± 10 gm). They were acclimatized at room temperature with 12 h dark/light cycle, fed on standard diet and water ad-libitum. Further sub lethal doses were calculated by standard method.

Experimental plant, Catharanthus roseus was grown in garden associated to our laboratory and methanolic extract was prepared from all aerial parts. The extract was analyzed for its chemical ingredient through GC/MS with the help of ThermoFischer ISQ 7000 instrument. Further safety trail was performed to determine the dose of extract and set it to be 500 mg/kg body weight.

For experimentation all rats were grouped in to four sets one acute (1d) and there sub-acute (7, 14, 21 ds). Each set contain four groups one control (group-A) and three treated (group B, C, and D). Details of doses to various group has been shown in Table 1.

At the end of designated days, rats of various groups were sacrificed and blood samples were collected from the ventricle of heart. Serum was separated for determination of Urea and BUN by Berthelot Method (Newman and Price, 1999), Serum uric acid by Uricase Trinder Method (Newman and Price, 1999), Serum creatinine by Alkaline Picrate Method (Tietz et al., 1994) and protein profile by modified Biuret assay method (Dumas et al., 1975). The results were validated by histological study of kidneys.

The observations were analyzed statistically using analysis of variance (ANOVA) followed by Student’s Newmann Keul's (SNK) test. The obtained values were compared to control and signified at the level of p < 0.01.

3. Result

Results of the study have been summarized in Tables 2 and 3, showing that cadmium intoxication significantly enhances the serum level of nitrogenous wastes (urea, BUN, Uric Acid and Creatinin) while decreases the serum proteins in acute and Sub-acute sets both. This altered level of nitrogenous wastes and serum proteins are brought back to normalcy by pretreatment with Catharanthus roseus extract, observed as non-significant result of group-D compared to control. Whereas C. roseus itself does not produce any harmful effect, again observed as non-significant results of Group-C as compared to control. For validation of amelioration histology was performed for all group of sub-acute sets (21 days), which is shown as Fig. 1A–D.

Table 1

| Serum nitrogenous waste | Treatment days | Control | Cadmium treated | Catharanthus treated | Cadaranthus + Cadmium treated |
|-------------------------|----------------|---------|-----------------|----------------------|-------------------------------|
| Urea (mg/dL)            | Acute (1 day)  | 17.42 ± 0.12 | 19.84 ± 0.62 | 17.19 ± 0.18 ns  | 17.56 ± 0.07                |
|                         | Sub-acute (7 days) | 16.56 ± 0.15 | 20.83 ± 0.34 | 17.26 ± 0.57 ns  | 17.58 ± 0.12 ns             |
|                         | Sub-acute (14 days) | 17.72 ± 0.11 | 20.56 ± 0.35 | 17.61 ± 0.03 ns  | 17.57 ± 0.98 ns             |
|                         | Sub-acute (21 days) | 17.37 ± 0.27 | 20.87 ± 0.41 | 17.28 ± 0.47 ns  | 17.39 ± 0.29                |
| BUN (mg/dL)             | Acute (1 day)  | 6.98 ± 0.08  | 9.58 ± 0.16  | 7.64 ± 0.08 ns  | 8.02 ± 0.16                 |
|                         | Sub-acute (7 days) | 7.94 ± 0.04  | 10.01 ± 0.13 | 7.98 ± 0.33 ns  | 8.09 ± 0.93 **              |
|                         | Sub-acute (14 days) | 8.01 ± 0.08  | 9.89 ± 0.16  | 7.37 ± 0.12 ns  | 8.36 ± 0.47 ns              |
|                         | Sub-acute (21 days) | 7.64 ± 0.04  | 10.25 ± 0.10 | 7.98 ± 0.02 ns  | 8.59 ± 0.64 ns              |
| Uric acid (mg/dL)       | Acute (1 day)  | 2.12 ± 0.09  | 2.27 ± 0.05  | 2.13 ± 0.02 ns  | 2.09 ± 0.02                 |
|                         | Sub-acute (7 days) | 2.09 ± 0.13  | 2.28 ± 0.05  | 2.19 ± 0.01 ns  | 2.09 ± 0.01 ns              |
|                         | Sub-acute (14 days) | 2.08 ± 0.08  | 2.29 ± 0.06  | 2.17 ± 0.03 ns  | 2.07 ± 0.04                 |
|                         | Sub-acute (21 days) | 2.06 ± 0.08  | 2.34 ± 0.06  | 2.13 ± 0.01 ns  | 2.13 ± 0.02 ns              |
| Creatinin (mg/dL)       | Acute (1 day)  | 0.57 ± 0.17  | 1.02 ± 0.08  | 0.89 ± 0.03 ns  | 0.96 ± 0.02 ns              |
|                         | Sub-acute (7 days) | 0.63 ± 0.09  | 0.99 ± 0.08  | 0.88 ± 0.04 ns  | 0.98 ± 0.02 ns              |
|                         | Sub-acute (14 days) | 0.62 ± 0.08  | 0.97 ± 0.05  | 0.93 ± 0.07 ns  | 0.95 ± 0.01 ns              |
|                         | Sub-acute (21 days) | 0.65 ± 0.03  | 1.01 ± 0.03  | 0.98 ± 0.06 ns  | 0.98 ± 0.04                 |

* p < 0.01 significant Vs control.
** p > 0.01 non-significant Vs control.
4. Discussion

The results of the study are in agreement with Ahmad and Misra (2014). Since liver and kidneys are the first organ in the body to encounter with cadmium thus most studies of cadmium are centered on detection of early sign of renal dysfunction. Once absorbed from GIT, cadmium forms durable combination with thionein protein in the liver, now called as metallothionien, which play an important in further metabolism of this metal. This is eliminated via kidney (Ruttkay-Nedecky et al., 2013; Yang and Shu, 2015).

Serum nitrogenous waste concentration is determined largely by efficiency of renal clearance say glomerular filtration rate [GFR] (Dioka et al. 2004; Mobin, 2013). The metal accumulated in renal cortex may result in reduced renal clearance (Marya, 2003). Accumulation also causes cellular depolarization thus imbalanced flux of sodium and potassium ion resulting in to loss of tubular function. Cadmium may also bind to proteins of filtration membrane causing tubular proteinuria. Cadmium increases sodium reabsorption, decreases cell membrane resistance and stimulates potassium channels (Jungwirth et al., 1990; Newman and Price, 1999). Cadmium interferes with glomerular filtration rate and tubular re-absorption from any of these possible ways leading to increased serum concentration of nitrogenous wastes and decreased protein profile. The same is shown by comparison of Fig. 1(A & B). Serum proteinuria might be the possible reason for significant reduction in serum proteins. Proteinuria is the major outcome of oxidative stress produced by cadmium (Alba Bhutkar and Bhise, 2011). This may overcome significantly glomerular endothelial and tubular injury and provide an extra support to endogenous protection mechanism. This pretreatment with Catharanthus roseus extract causes significant increase in serum nitrogenous wastes and protein profile to normalcy. This ameliorative potential of Catharanthus roseus extract is further validated by comparative study of Fig. 1(A & D).

Table 3

| Protein | Treatment day | Control | Cadmium treated | Catharanthus treated | Cadmium + Catharanthus treated |
|---------|---------------|---------|-----------------|----------------------|-------------------------------|
| Total Proteins (g/dL) | Acute (1 day) | 7.01 ± 0.02 | 6.43 ± 0.02 | 7.83 ± 0.01* | 6.78 ± 0.17* |
|  | Sub-acute (7 days) | 7.04 ± 0.01 | 5.89 ± 0.07 | 8.29 ± 0.07* | 6.98 ± 0.48* |
|  | Sub-acute (14 days) | 6.98 ± 0.17 | 5.16 ± 0.09 | 7.89 ± 0.23* | 7.67 ± 0.24* |
|  | Sub- acute (14 days) | 6.94 ± 0.12 | 4.51 ± 0.08 | 7.87 ± 0.09* | 7.51 ± 0.31* |
| Albumins (g/dL) | Acute (1 day) | 4.27 ± 0.19 | 4.17 ± 0.09 | 4.68 ± 0.23* | 4.17 ± 0.09* |
|  | Sub-acute (7 days) | 4.68 ± 0.17 | 3.89 ± 0.17 | 4.47 ± 0.19 | 4.29 ± 0.06 |
|  | Sub-acute (14 days) | 4.89 ± 0.41 | 3.78 ± 0.08 | 4.87 ± 0.17* | 4.67 ± 0.07* |
|  | Sub-acute (14 days) | 4.87 ± 0.39 | 3.69 ± 0.07 | 4.89 ± 0.21* | 4.69 ± 0.08* |
| Globulin (g/dL) | Acute (1 day) | 4.09 ± 0.09 | 3.98 ± 0.01 | 4.17 ± 0.09* | 4.49 ± 0.09* |
|  | Sub-Acute (7 days) | 3.89 ± 0.09 | 3.19 ± 0.03 | 4.08 ± 0.008* | 4.27 ± 0.009* |
|  | Sub-acute (14 days) | 4.17 ± 0.08 | 2.81 ± 0.03 | 4.27 ± 0.07* | 4.28 ± 0.09* |
|  | Sub-acute (14 days) | 4.08 ± 0.05 | 2.87 ± 0.02 | 4.29 ± 0.05* | 4.63 ± 0.03* |
| A/G ratio | Acute (1 day) | 2.34 ± 0.02 | 2.09 ± 0.01 | 2.21 ± 0.04* | 2.13 ± 0.02* |
|  | Sub-acute (7 days) | 2.41 ± 0.04 | 2.64 ± 0.03 | 2.38 ± 0.03* | 2.42 ± 0.09* |
|  | Sub-acute (14 days) | 2.09 ± 0.03 | 2.24 ± 0.18 | 2.09 ± 0.03* | 2.12 ± 0.05* |
|  | Sub-acute (14 days) | 2.11 ± 0.04 | 2.63 ± 0.04 | 2.18 ± 0.01* | 2.09 ± 0.03* |

* p < 0.01 significant Vs control.
ns p > 0.01 non-significant Vs control.

Fig. 1. (A–D): Histomicrograph (100 x) of kidney of Rattus norvegicus. A: Control; B: Cadmium treated; C: Catharanthus treated; D: Catharanthus + Cadmium treated. Where Gl (Glomerulus); Tu (Tubules); IntTs (Inter-Tubular spaces).
5. Conclusions

*Catharanthus roseus* extract significantly brought back the changes in renal functions made by cadmium to normalcy. Further separation of extract ingredients and their individual effect studies are proposed. The study was performed in lab animals; further clinical trial is required to establish its utility to human being.

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