

Article

EFFECT OF CROCIN ON PKHD1 AND KLLN GENES EXPRESSION IN KIDNEY TISSUE OF MALE RATS TREATED WITH CADMIUM

Efecto de la crocina sobre la expresión de los genes PKHD1 y KLLN en tejido renal de ratas macho tratadas con cadmio

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SUMMARY

According to data from studies, antioxidant herbal compounds are, likely to have a useful role in reducing the harmful effects of environmental pollutants and toxic chemicals that most people are exposed to. Cadmium is one of the toxic elements that accumulate in many organs, especially in kidneys. The aim of this study was to investigate the effect of crocin on the expression of PKHD1 and KLLN genes in cadmium-treated rats.

In this experimental study, 40 adults male Wistar rats (200-250 g) were randomly divided into the following groups: control group received normal saline, cadmium group (15mg/kg), crocin group (20mg/kg) and cadmium group daily fed with crocin at a dose of 20 mg/kg.

After eight weeks of treatment, rats were dissected, and kidney tissues were removed for evaluation of PKHD1 and KLLN gene expression by real time method. The data were analyzed using one-way ANOVA and significant difference between groups was $P < 0.05$.

Our results showed an increase in PKHD1 gene expression and a decrease in KLLN gene expression in kidney tissue in the cadmium group compared to the control group ($P < 0.001$).

Also, a significant decrease in PKHD1 gene expression ($P < 0.001$) and an increase in KLLN gene expression ($P < 0.05$) were observed in the tissues of all cadmium-treated rats compared to cadmium.

Crocine consumption can have a protective effect against the impaired expression of PKHD1 and KLLN cadmium-induced apoptotic pathway.

Keywords: Kidney Cancer; Cadmium; Crocin; PKHD1; KLLN.

1. Introduction

Nowadays, cancer is one of the diseases that affect populations all over the world and cause many problems. Among the types of cancers, a high percentage of people may get kidney cancer (Siegel et al., 2016). Genetic and environmental factors associated with kidney cancer one of the environmental factors is exposure to a variety of environmental toxins, heavy metals and environmental contaminants that may be risk factors for kidney cancer (Chow et al., 2010).

Cadmium (Cd) is one of the heavy elements that are abundant in the environment, soil, and many industries and humans are exposed to it (Chen et al., 2016). Cadmium has shown its side effects on various organs and causes kidney failure, lung cancer, brain diseases, anemia, skeletal changes, fetal abnormalities and reproductive disorders (Chen et al., 2016).

Previous studies have identified the kidney as the most common organ involved with this element (Sá et al., 2016). Effect on message transmission (Thévenod., 2009), peroxidation of unsaturated fatty acids, inhibition of DNA repair system, inhibition of DNA methylation (Hartwig & Schwerdtle, 2002), depletion of antioxidants, dysfunction of E-cadherin protein, interaction with different ions, oxidative stress induced by free radicals overproduction (ROS) (Liu et al., 2011), inflammation and subsequent DNA oxidation, decreased levels of antioxidants, apoptosis disorder and increased activity of some proteases are potential mechanisms of toxic effects of cadmium on tissue damage (Kim et al., 2017; Mahdavi et al., 2011). Therefore, cadmium has been proposed as a candidate for alteration in the expression of effective genes resulting from kidney cancer (Chen et al., 2016). As mentioned above, one of the suggested mechanisms for the effect of cadmium on apoptotic kidney tissue is that this pathway is mediated by different genes and impaired expression of each of these genes can have damaging consequences (Mahdavi et al., 2011). Two of the major genes in the renal tissue apoptosis pathway are polycystic kidney and hepatic disease 1 (PKHD1) and killin or p53-regulated DNA replication inhibitor (KLLN).

The PKHD1 gene is an important factor in inducing apoptosis and invasion of cancer cells. Therefore, downregulation of the PKHD1 gene may be of great help in promoting apoptosis and in reducing adhesion and metastatic properties of cancer cells (Ward et al., 2011), and the KLLN gene encodes a protein called Killin, which controls the expression of the Killin protein by the P53 protein (Thompson, et al. 2012). Changes in the expression of the KLLN gene can lead to severe cell damage as well as death. Therefore, the expression of killin protein prevents the cancer cells by controlling and inducing cell apoptosis (Nizialek et al., 2013).

Combined with the P53 is one of the KLLN mechanisms for inducing apoptosis. It has been demonstrated that after P53 induction during the S phase, KLLN binds highly to single-stranded and double-stranded DNA, causing stopping S phase and ultimately inducing apoptosis (Wang et al., 2015).

Since cancer is a malignant disease and its therapeutic drugs have high cost and side effects, it is logical to search for treatment and support strategies. Therefore, the attempt to introduce preventive compounds, therapeutic methods and reduce the effect of risk factors on cancer incidence can reduce the percentage of patients with this disease and treat more patients in the community (Sun et al., 2011).

Most herbal research today relates to the healing and therapeutic effects of traditional herbs that have long been used in societies. One of these plants, which has multiple scientific articles on its properties and importance in published scientific journals, is the saffron plant from the *Iridaceae* family with various compounds and antioxidant properties, one of the compounds of interest to researchers in saffron is crocin (Bakshi et al., 2017). Crocin is a carotenoid chemical compound with very high antioxidant properties (Bakshi et al., 2017) and has anti-inflammatory properties (Bakhtiary et al.,

2014; Bakshi et al., 2017). It has the most anticancer effect among saffron compounds, and has been proven to have proapoptotic properties. Due to its powerful anticancer properties, it can be a good candidate for cancer treatment and prevention (Hoshyar et al., 2017).

Several mechanisms have been proposed for the anti-cancer effects of crocin, including: effect on cell proliferation, induction of apoptosis, inhibition of angiogenesis (Hoshyar et al., 2017), inhibition of DNA and RNA synthesis however, the precise mechanism of crocin on DNA and RNA synthesis is unclear (Sun et al., 2011). Crocin induces cell death in p53-dependent and non-dependent pathways (Amin et al., 2015).

Crocin probably affects the mitochondrial membrane by affecting the expression of apoptotic and anti-apoptotic proteins and causes the release of cytochrome C, resulting in mitochondrial-dependent apoptosis in cancer cells. On the other hand, this compound affects the G2 stage of the cell cycle by affecting cadherin protein (Cdc25a / b / c) and then cyclin-B-cdk2 and accelerates the process of apoptosis (Amin et al., 2015).

Based on the scientific evidence on the effect of crocin on the expression of various genes in the apoptotic pathway in various cancers, in this project, the effect of crocin on the expression of PKHD1 and KLLN genes in damaged kidney tissue in cadmium-deficient rats was investigated.

2. Materials and methods

Animals

To do this experiment, forty healthy adult male Wistar rats (160-200 g) were purchased from the Pasteur Institute. All rats were housed under standard conditions ($21 \pm 2^\circ\text{C}$; 12/12h light/dark cycle, relative humidity 40-60%) in plastic cages. The animals had free access to water and fed special commercial food during the experiment. They were kept in the laboratory for seven days before the start of the test. In this examination, all the Ethics of working with animals were observed.

Chemicals

In this study, cadmium chloride was purchased from the Azmiran company (Germany, Alfa Aesar), And crocin was obtained from Sigma Aldrich chemical (Sigma- Aldrich, USA). These doses were chosen base on previous reports (Kilic & Kilic 2017; Ghotbeddin et al., 2017).

Animal grouping

Forty male Wistar rats were randomly divided into four groups (n= 10 per group):

Control group (1cc of normal saline (d. w.) /day), cadmium group (15 mg/ kg), crocin group (20mg/ kg) and Cadmium-Treated group (20 mg crocin/kg).

Injections of cadmium in single dose intraperitoneally and crocin were gavaged to rats for 8weeks.

Cadmium in single-dose (15mg/kg) and Crocin(20mg/kg) was gavaged for 8 weeks. These doses were chosen based on previous reports (Kilic et al., 2017; Ghotbeddin et al., 2017).

At the end of the experiment, the animals were anesthetized with ether and after dissection of animals and incision in the abdominal region of the separated kidney tissue, in washed physiological serum and to evaluate gene expression PKHD1 and KLLN at -70°C in liquid nitrogen were kept.

Measurement of genes expression

To evaluate the expression of PKHD1 and KLLN genes in kidney tissue, RNA was extracted with TRIZOL solution (CinnaGen Inc., Iran). In order to qualitatively evaluate the expression of the genes studied, PCR products were analyzed by 1.5% agarose gel electrophoresis also, the concentration and purity of the extracted RNA were confirmed by examining the absorption ratio at 260 / 280 nm with the nano-drop (NanoDrop ND-1000 spectrophotometer; Thermo Fisher Scientific, Waltham, MA), and samples with a light absorption ratio of 1.8 -2 were used for DNA synthesis. In this research, cDNA was synthesized according to the instructions of the PrimScript RT Reagent Kit (Japan Takara Corporation with proprietary code RR037A). To synthesize cDNA in a soluble RNA microtube, 0.5 µl of hexamer random and 5 µL of deionized water were added. The microtubes were placed in a thermocycler for 15 minutes at 37 ° C. Subsequently, then 2 µL RT-specific buffer, 0.5 oligo dNTPs primer, 0.5 µL reverse transcriptase enzyme were added to the microtube to reach the final volume of 10 µl. Time required for cDNA synthesis based on the desired kit: 5 min 65 ° C, 85 °C for 5 min and finally the reaction was kept at 4 ° C.

Real time PCR was performed by Rotor Gene 6000 (Corbett Research, Australia) for genes expression analysis. For real-time PCR, 2x master mix buffer, forward and reverse primer, cDNA and deionized water were used. The thermal cycling conditions were as follows: 15 min at 95°C, and 40 cycles at 95°C for 20 seconds, 60°C for 20 seconds and 72°C for 5 min. The expression of the gene was assayed using $2^{-\Delta\Delta C_t}$ formula (Jalili et al., 2017). The primers required were designed using Oligo 7 software and the blasts were examined to confirm the sequence of primers designed from the NCBI site (<http://www.ncbi.nlm.nih.gov/blast>). The primers sequence is presented in Table 1, the GAPDH gene was used as housekeeping.

Table 1.

The sequence of primers used for real-time PCR analysis.

Gene	Forward	Reverse
GAPDH	5`-ATCACTGCCACTCAGAAGAC -3`	5`-ACATTGGGGGTAGGAACAC -3`
PKHD1	5`-AGCTGGAGGAGACAAATGGC-3`	5`-CCAGGCTTCCTTATGCACCA-3`
KLLN	5`-CCCAGGGAGTGCAAAGAGAG-3`	5`-TCTTCGGGTAGCTGGAGTGA-3`

Statistical data analysis

Statistical analysis was performed by **GraphPad Prism statistical software package version 8.4**. The data were expressed as mean±SEM and analyzed by one-way analysis of variance (ANOVA) followed by post hoc Tukey's test was used to assess the statistical significance of data between different groups. Significance level P<0.05 was considered.

3. Results

Quantitative expression analysis PKHD1 and KLLN genes

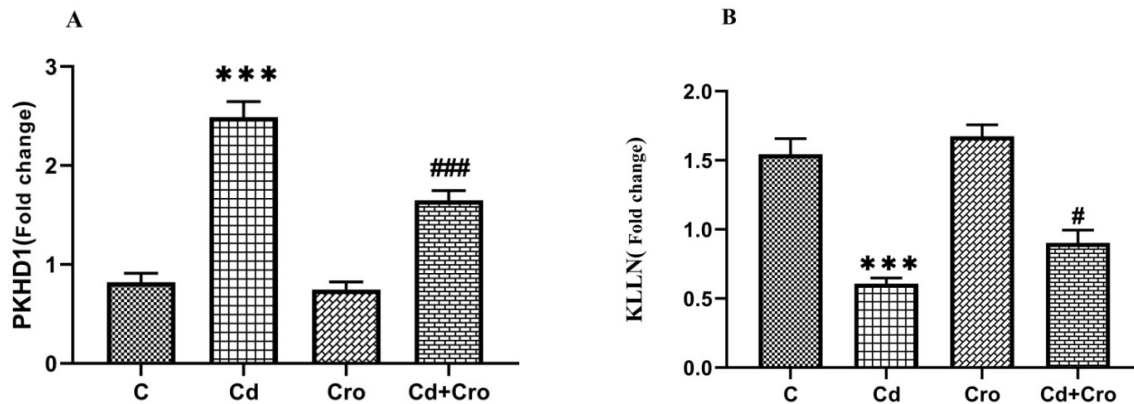
Comparison of the expression of PKHD1 and KLLN genes between different groups of the current study is presented in Figure 1A and B. There was a significant increase in PKHD1 gene expression in the cadmium group compared to the control group (P<0.001). The expression of PKHD1 gene was significantly increased in cadmium groups treated with crocin compared to the cadmium group (P<0.001).

In Figure 1B, the difference in expression of the KLLN gene is observed in different groups.

KLLN gene expression was significantly decreased in the cadmium group in compared to the control group ($P<0.001$). Statistical analysis also illustrated a significant increase of KLLN gene expression in the cadmium group gavaged with crocin for 8 weeks compared to the cadmium group ($P<0.05$).

Figure 1.

Effects of crocin on the genes expression PKHD1(A) and KLLN (B) of in male rat expose cadmium. Data are expressed as mean \pm SEM (n=10).



C: control, Cd: cadmium (15mg/kg), Cro: crocin (20mg/kg), Cd+Cro: cadmium (15mg/kg) + crocin (20mg/kg).

***: $P<0.001$ as compared with the control group. ###: $P<0.001$, #: $P<0.05$ as compared with the cadmium.

4. Discussion

Cadmium has specific toxic effects on kidney tissue cells and reacts with many cellular transport proteins to cause kidney tissue disruption (Mollaie et al., 2000). In addition to the pathological effects of cadmium such as genotoxicity, protein degradation, and DNA damage, apoptosis has recently attracted much research. Recent studies have shown that cadmium is likely to interfere with apoptosis in cells through several mechanisms (Chen et al., 2014).

The evaluation of expression of two genes in this research and their changes in cadmium group compared with control group is in line with previous evidence. Venza et al. (2014), suggested that cadmium induces kidney cancer in humans through induction of epigenetic changes, especially changes in DNA methylation patterns (Venza et al., 2014). Rafati Rahimzadeh and worker indicated that cadmium has direct detrimental effects on cell life, which can be adversely affected by induction of protease activity and impaired protease/antiprotease balance, ultimately leading to cell membrane damage, inflammation and increased risk (Rafati Rahimzadeh et al., 2017).

Some studies attributed the increase in oxidative stress due to the production of free radicals as well as the induction of inflammation as a possible mechanism of cadmium in causing cellular damage as well as induction of cancer (Liu et al., 2011; Kim et al., 2017). Given the pathological effects of cadmium on renal cancer induction (Kun Song et al., 2015) as well as the pivotal role of KLLN and PKHD1 genes

in the incidence of kidney disease, in the present study, for the first time, the effect of crocin on the expression of PKHD1 and KLLN genes in renal tissue of male rats were tested for cadmium.

The use of compounds that can counteract the toxic effect can greatly help to improve the destructive effects of this toxic element. The results of our finding indicated the effect of crocin on the expression of both PKHD1 and KLLN genes. Statistical analysis of these two genes revealed a decrease in PKHD1 gene expression and an increase in KLLN gene expression in kidney tissue of rat treated with crocin compared to the cadmium group ($p < 0.05$).

According to research, crocin is a compound with high antioxidant and anti-inflammatory properties that, due to its positive properties, can reduce the production of free radicals and greatly protects the cells and tissues involved (Bakshi et al., 2017). So, since the phenomenon of apoptosis abolished many in cancer cells, the use of crocin is likely to induce the apoptotic process to cause premature death of cancer cells (Hoshyar et al., 2017).

It has been reported that the PKHD1 gene plays an important role in the control of apoptosis and the development of cancer cells (Ward et al., 2002). Ward et al. demonstrated that PKHD1 gene expression is increased in colorectal cancer cells. They also reported that mutations in the PKHD1 gene are closely linked to colorectal cancer (Ward et al., 2011).

Sjöblom et al. (2006), studying 14661 colorectal cancer patients, found that increased expression of PKHD1 after mutation was one of the most common somatic the mutation associated with cancer cell proliferation and metastasis (Sjöblom et al., 2006).

Evidence suggested that increased expression of PKHD1 is associated with an increased risk of colorectal cancer and the severity or stage of the disease. (Ward et al., 2002). In our finding, the expression of PKHD1 in the kidney of rats receiving cadmium was also increased, which is in line with the results of previous studies.

Concerning the other gene in the present finding, the results of our analysis suggested that consumption crocin induces KLLN gene expression in kidney tissue of rats, so that crocin had the most significant effect on increasing KLLN expression. However, cadmium significantly decreased KLLN expression in the kidney tissue of rats. On the other hand, co-administration of cadmium and crocin significantly increased KLLN expression compared to the cadmium-treated group. It has been reported that the KLLN gene plays an important role in controlling apoptosis and preventing the progression of cancer cells. It seems that impaired expression may be one of the leading causes of the development and progression of kidney cancer (Nizialek et al., 2013).

Given the relationship between p53 and the KLLN gene, and on the other hand, the effect of crocin on p53 (Hoshyar et al., 2017), it is hypothesized that crocin can affect the expression of the KLLN gene and induce apoptosis, and it may have a powerful effect in treating and preventing kidney cancer. According to the finding of Nizialek et al., (2013) a mutation in the KLLN gene impaired its protein function and, as a result, made breast cancer cells immature, increased metastatic properties, proliferative potential, and differentiation (Nizialek et al., 2013).

Wang and colleagues reported that increased KLLN gene expression induces apoptosis, decreased breast cancer cell viability, decreased metastatic properties and angiogenesis (Wang et al., 2013).

In another study, Chen et al. (2015) Showed that crocin induced apoptosis and susceptibility of cancer cells and eventually their premature death by stopping the cell cycle and increasing the Bax / Bcl2 ratio, as well as increasing P53 expression.

Therefore, increasing the expression of KLLN gene and decreasing the expression of PKHD1 gene in the cadmium group treated with crocin will probably be very helpful in promoting apoptosis

and reducing the adhesion and metastasis properties of cancer cells due to the proapoptotic properties of crocin.

5. Conclusion

The results of the effect of crocin on gene expression of PKHD1 and KLLN kidney tissue in the treated cadmium group, can show the effective effect of crocin of saffron plant in line with previous studies. It is possible to use additional studies of crocin as a natural herbal compound in supportive and therapeutic methods for people with or suffering from kidney cancer, although the requirement of this proposal definitely requires histological studies, biochemical factors, pathways and other genes are in the process of apoptosis in kidney tissue.

References

- Amin, A., Bajbouj, K., Koch, A., Gandesiri, M., Schneider-Stock, R. (2017) Defective Autophagosome Formation in p53-Null Colorectal Cancer Reinforces Crocin-Induced Apoptosis. *Int J Mol Sci.* , 16(1):1544-1561. <https://doi.org/10.3390/ijms16011544>
- Bakhtiary, Z., Shahrooz, R., Ahmadi, A., Malekinejad, H., Mostafavi, M. (2014) Study of protective effects of crocin on testicular histomorphometry and serological parameters in cyclophosphamide on treated adult mice. *Stud Med Sci.*, 25(7):663-673. URL: <http://umj.umsu.ac.ir/article-1-2417-en.html>
- Bakshi, H.A., Hakkim, F.L., Sam, S., Javid, F., Rahan, L. (2017) Dietary Crocin Reverses Melanoma Metastasis. *J Biomed Res.*, 32(1):39-50. <https://doi.org/10.7555/JBR.31.20160120>
- Bennett, K.L., Campbell, R., Ganapathi, S., Zhou, M.; Rini, B., Ganapathi, R, et al. (2011) Germline and somatic DNA methylation and epigenetic regulation of KILLIN in renal cell carcinoma. *Genes Chromosomes Cancer.*, 50(8):654-61. <https://doi.org/10.1002/gcc.20887>
- Chen, C., Xun, P., Nishijo, M., He, K. (2016) Cadmium exposure and risk of lung cancer: a meta-analysis of cohort and case-control studies among general and occupational populations. *J Expo Sci Environ Epidemiol.*, 26 (5):437-444. <https://doi.org/10.1038/jes.2016.6>
- Chen, Sh., Zhao, Sh., Wang, X., Zhang, L., Jiang, E., Guet, Y. et al. (2015) Crocin inhibits cell proliferation and enhances cisplatin and pemetrexed chemosensitivity in lung cancer cells. *Transl Lung Cancer Res.*, 4(6): 775-783. <https://doi.org/10.3978/j.issn.2218-6751.2015.11.03>
- Chen, Y.Y., Zhu, J.Y., Chan, K.M. (2014) Effects of cadmium on cell proliferation, apoptosis, and proto-oncogene expression in zebrafish liver cells. *Aquat Toxicol.*, 157:196-206. <https://doi.org/10.1016/j.aquatox.2014.10.018>
- Chow, W.H., Dong, L.M., Devesa, S.S. (2010) Epidemiology and risk factors for kidney cancer. *Nat Rev Urol.*, 7(5):245-257. <https://dx.doi.org/10.1038/nrurol.2010.46>
- Ghotbeddin, Z., Fatemi-Tabatabaei, S.R., Tabandeh, M.R., Mirzabeigi, M., Badripour, N., Amiri R. (2017) Effect of crocin on inhibitory avoidance memory, balance and explorative behaviours following cisplatin administration in rat. *Feyz.*, 21(2):118-25. <https://doi.org/10.3390/ijms16011544>
- Hartwig, A., Schwerdtle, T. (2002) Interactions by carcinogenic metal compounds with DNA repair processes: toxicological implications. *Toxicol Lett.*, 127(1-3):47-54. [https://doi.org/10.1016/S0378-4274\(01\)00482-9](https://doi.org/10.1016/S0378-4274(01)00482-9)

- Hoshyar, R.; Mollaei ,H. (2017) A comprehensive review on anticancer mechanisms of the main carotenoid of saffron, crocin. *J Pharm Pharmacol.*,69(11):1419-1427. <https://doi.org/10.1111/jphp.12776>
- Liu, T., He, W., Yan, C., Qi, Y., Zhang, Y. (2011). Roles of reactive oxygen species and mitochondria in cadmium-induced injury of liver cells. *Toxicol Ind Health.*,27(3):249-56. <https://doi.org/10.1177/0748233710386408>
- Jalili, C., Salahshoor, M.R.,Moradi, M.T., Ahookhash, M., Taghadosi, M., Sohrabi, M. (2017) Expression Changes of Apoptotic Genes in Tissues from Mice Exposed to Nicotine. *Asian Pac J Cancer Prev.*,18(1):239-244. <https://doi.org/10.22034/apjcp.2017.18.1.239>
- Kilic, S., Kilic, M. (2017) Effects of cadmium-induced stress on essential oil production, morphology and physiology of lemon balm (*Melissa officinalis* L., Lamiaceae). *Appl Ecol Environ Res.*, 15(3):1653-69. http://dx.doi.org/10.15666/aecr/1503_16531669
- Kim, J.,Song, H., Heo, H.R., Kim, J.W., Kim, H.R., Hong ,Y.,et al. (2017) Cadmium-induced ER stress and inflammation are mediated through C/EBP-DDIT3 signaling in human bronchial epithelial cells. *Exp Mol Med.*,49(9):e372. <https://doi.org/10.1038/emm.2017.125>
- Song, KJ., Luo, H., Yin, Xh., lei Huang, G., yang Luo ,S., Yuan ,D.B.,et al. (2015) Association between cadmium exposure and renal cancer risk: a meta-analysis of observational studies. *Sci rep*,5:17976. <https://doi.org/10.1038/srep17976>
- Mahdavi, L.S .,Khodarahmi, P .,Hayati, R.N .(2017)Effects of intraperitoneal administration of cadmium on apoptotic bcl-2 and Bax genes expression in rat hippocampal cells. *MGj.*,12 (1) :81-89. http://mg.genetics.ir/browse.php?a_id=1470&sid=1&slc_lang=en
- Mollaei, H., Safaralizadeh, R., Babaei, E., Abedini, M.R., Hoshyar, R.(2017) The anti-proliferative and apoptotic effects of crocin on chemosensitive and chemoresistant cervical cancer cells. *Biomed Pharmacother.*,94:307-316. <https://doi.org/10.1016/j.biopha.2017.07.052>
- Nizialek ,E.A., Peterson, C., Mester, J.L., Downes-Kelly, E.; Eng, C. (2013) Germline and somatic KLLN alterations in breast cancer dysregulate G2 arrest. *Hum Mol Genet.*,22(12):2451-2461. <https://doi.org/10.1093/hmg/ddt097>
- Sá, I., Semedo, M., Cunha, M.E. (2016) Kidney cancer. Heavy metals as a risk factor. *Porto Biomed J.*,1(1):25-28. <https://doi.org/10.1016/j.pbj.2016.03.006>
- Siegel, R.L., Miller, K.D., Jemal ,A.(2016) Cancer statistics. *CA Cancer J Clin.*,66(1):7-30. <https://doi.org/10.3322/caac.21332>
- Sjöblom, T.,Jones, S.,Wood, L.D., Parsons, D.W., Lin J., Barber, T.D.,et al. (2006) The consensus coding sequences of human breast and colorectal cancers. *Science.*,314(5797):268-274. <https://doi.org/10.1126/science.1133427>
- Wang, J., Ke ,Y., Shu, T.(2009) Crocin has pharmacological effects against the pathological behavior of colon cancer cells by interacting with the STAT3 signaling pathway. *Exp Ther Med.* 2020;19(2):1297-1303. doi:10.3892/etm.2019.8329Thévenod, F.Cadmium and cellular signaling cascades: to be or not to be?. *Toxicol Appl Pharmacol.*,238(3):221-239. <https://doi.org/10.1016/j.taap.2009.01.013>
- Thompson, E.R., Gorringer, K.L., Choong, D.Y., Eccles ,D.M., kConFab, Mitchell, G.,et al. (2012) Analysis of KLLN as a high-penetrance breast cancer predisposition gene. *Breast Cancer Res Treat.*,134(2):543-547. <https://doi.org/10.1007/s10549-012-2088-3>

- Venza, M., Visalli, M., Biondo, C., Oteri, R., Agliano, F., Morabito, S., et al. (2014) Epigenetic effects of cadmium in cancer: focus on melanoma. *Curr Genomics.*,15(6):420-435. <https://doi.org/10.2174/138920291506150106145932>
- Wang, Y., He, X., Yu, Q., Eng, C. (2013) Androgen receptor-induced tumor suppressor, KLLN, inhibits breast cancer growth and transcriptionally activates p53/p73-mediated apoptosis in breast carcinomas. *Hum Mol Genet.*,22(11):2263-2272. <https://doi.org/10.1093/hmg/ddt077>
- Ward, C.J.; Hogan, M.C.; Rossetti, S.; Walker, D.; Sneddon, T.; Wang, X.; et al. (2002) The gene mutated in autosomal recessive polycystic kidney disease encodes a large, receptor-like protein. *Nat Genet.*,30(3):259-269. <https://doi.org/10.1038/ng833>
- Ward, C.J., Wu, Y., Johnson, R.A., Woollard, J.R., Bergstralh, E.J., Cicek, M.S., et al. (2011) Germline PKHD1 mutations are protective against colorectal cancer. *Hum Genet.*,129(3):345-349. <https://doi.org/10.1007/s00439-011-0950-8>
- Rafati Rahimzadeh, M., Rafati Rahimzadeh, M., Kazemi, S., Moghadamnia, A.A. (2017) Cadmium toxicity and treatment: An update. *Caspian J Intern Med.* 8(3):135-145. <https://doi.org/10.22088/cjim.8.3.135>

RESUMEN

Diversos estudios sugieren que compuestos antioxidantes de hierbas tienen un papel útil en la reducción de los efectos nocivos de los contaminantes ambientales y los químicos tóxicos a los que está expuesta la mayoría de las personas. El cadmio es uno de los elementos tóxicos que se acumulan en muchos órganos, especialmente en los riñones. El objetivo de este estudio fue investigar el efecto de la crocina en la expresión de los genes PKHD1 y KLLN en ratas tratadas con cadmio.

En este estudio experimental, 40 ratas Wistar macho adultas (200-250 g) se dividieron aleatoriamente en los siguientes grupos: el grupo de control recibió solución salina normal, el grupo de cadmio (15 mg / kg), el grupo de crocina (20 mg / kg) y el grupo de cadmio alimentado diariamente con crocina a una dosis de 20 mg / kg.

Después de ocho semanas de tratamiento, se disecaron las ratas y se extrajeron los tejidos renales para evaluar la expresión de los genes PKHD1 y KLLN mediante un método en tiempo real. Los datos se analizaron mediante ANOVA de una vía y la diferencia significativa entre los grupos fue $P < 0,05$.

Nuestros resultados mostraron un aumento en la expresión del gen PKHD1 y una disminución en la expresión del gen KLLN en el tejido renal en el grupo de cadmio en comparación con el grupo de control ($P < 0,001$).

Además, se observó una disminución significativa en la expresión del gen PKHD1 ($P < 0,001$) y un aumento en la expresión del gen KLLN ($P < 0,05$) en los tejidos de todas las ratas tratadas con cadmio en comparación con el cadmio.

El consumo de crocina puede tener un efecto protector contra la expresión alterada de la vía apoptótica inducida por cadmio PKHD1 y KLLN.

Palabras clave: Cáncer renal; Cadmio; Crocina; PKHD1; KLLN.
