International Journal of Enhanced Research in Science, Technology & Engineering
ISSN: 2319-7463, Vol. 12 Issue 8, August-2023, Impact Factor: 7.957

A Mini Review on Effect of Aniline on Liver and Spleen

Pallavi Mohurle[1], Varsha Dhurvey[2], Rashmi Urkude[3], Harshada Pawar[4], Dharita Joshi[5]

[1]Research student, Department of Zoology, RTM Nagpur University, Nagpur, Maharashtra, India
[2]Professor and Head, Department of Zoology, RTM Nagpur University, Nagpur, Maharashtra, India
[3]Professor, Department of Chemistry, Shivaji Science College, Nagpur, Maharashtra, India
[4]Research student, Department of Zoology, RTM Nagpur University, Nagpur, Maharashtra, India
[5]Research student, Department of Zoology, RTM Nagpur University, Nagpur, Maharashtra, India

ABSTRACT

The effects of certain environmental contaminants, such as compounds like aniline, on human health are constant. Aniline is amino aromatic substance. The liver is the primary location of aniline metabolism. According to several in-depth investigations, frequent exposure to aniline causes alterations that have an impact on the normal metabolism of the liver. Adding aniline also caused spectral abnormalities in liver microsomes, oxidative damage from aniline, and apoptosis in hepatocytes. As a result of chronic in vivo exposure to aniline, the spleen experiences a toxic reaction that is characterized by splenomegaly, hyperplasia, fibrosis, and tumor formation. Due to iron overload, oxidative DNA damage, and accelerated cell proliferation brought on by aniline exposure, the spleen may eventually develop tumors. The effects of aniline exposure on the liver and spleen were further discussed in this brief review.

Keywords: Aniline, liver, hepatocytes, oxidative damage, spleen, splenomegaly.

INTRODUCTION

For society, human health is a very critical issue. Pollutants including pesticides, herbicides, chemicals, and industrial waste always have an impact on health. In an environment that contains organic, inorganic substances, and their derivatives, chemicals constitute a major contaminant. The majority of chemicals are used in industry and released as solid, liquid, and gaseous wastes into the environment. Such substances have harmful impacts on all living things, including people. The most widely used chemical is Aniline.

Aniline is organic substance has the chemical formula C6H5NH2. It is the most basic aromatic amine, consisting of an amino group and a phenyl group. 8.4 million tons of aniline will be sold worldwide in 2020. The synthesis of dyes like fuchsine, safranin, and induline, plasticizers, pharmaceuticals like paracetamol and acetanilide, pesticides, herbicides, polymers including polyurethane and rubber additives, photographic chemicals as well as varnishes, and precursors of amino aromatic derivatives are just a few examples of the many different operational fields where aniline and its derivatives are used as intermediaries [1,2]. They are frequently observed in the environment because of their extensive use. Numerous industrially valuable compounds are synthesized using aniline and chloroaniline. As a result of their widespread use, these compounds are frequently found in the geo-ecological system. Because of their long-term persistence in the environment, they could be hazardous to aquatic life, people, and animals. [3]. If aniline is dispersed in the soil, it may swiftly contaminate groundwater or evaporatively evaporate to a moderate degree. In light of this, it may have an effect on living systems due to biological accumulation, long-term deposit, and carcinogenic qualities [4,5]. Male rats were gavaged 0.5, 15, and 60 mg/kg parachloro aniline and 100 mg/kg aniline daily for 28 days in a study [6]. In both cases, micronuclei increased at concentrations of 100 mg/kg aniline and 15 mg/kg parachloro aniline. In comparison to the control, there were statistically significant increases in mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and MPV at the same dose levels as there were statistically significant decreases in hemoglobin (HGB), absolute reticulocytes (ABRET), and methemoglobin levels. The mean corpuscular hemoglobin concentration (MCHC) and hematocrit (HCT) both decreased statistically significantly in the aniline group. These hematological findings support the observation that anemia was seen in the PCA-treated animals at dose levels of 15 mg/kg and 100 mg/kg aniline. In a another experiment, 30 male Wistar rats were exposed to concentrations of 9.2, 32.4, 96.5, and 274.9 mg/m^3 of aniline for six hours per day, five days per week, for two weeks. The findings of this

investigation indicate that hematological parameters at 90 and 270 m3 were significantly influenced. Red blood cell count and hemoglobin levels decreased [7]. Therefore, exposure to aniline can change certain hematological markers.

Physical And Chemical Properties Of Aniline

An organic substance known as aniline has the chemical formula C6H5NH2. The simplest aromatic amine is made up of an amino group and a phenyl group. Additionally, benzyl-amine, phenyl-amine, and amino benzene are some of the many aliases for aniline. It reacts quickly through an electrophilic substitution process because it is an electron-rich benzene derivative. Aromatic chemicals are known for easily igniting and burning with a smoky flame[8]. There are numerous analogues of aniline that have additional phenyl substitutions. These include, among many others, toluidines, xylidines, chloroanilines, aminobenzoic acids, and nitroanilines.

Historical Background

Otto Unverdorben destroyed indigo to extract aniline, which he did for the first time in 1826 [9]. He gave it the name Crystallin. FriedliebRunge discovered a compound from coal tar in 1834 that, when exposed to lime chloride, took on a lovely blue hue. He called it Cynol or Kynol[10]. Carl Julius Fritzsche (1808–1871) processed indigo with caustic potash in 1840 to produce an oil that he termed aniline after the plant *Indigoferasuffruticosa*, which produces indigo [11]. NikolayNikolaevichZinin converted nitrobenzene into a base in 1842, which he called benzidam. This compound came to be known as phenylamine or aniline when August Wilhelm von Hofmann demonstrated its identity in 1843 [12].

EFFECT ON HEPATIC TOXICITY

The liver is the primary location of aniline metabolism. After repeatedly administering male albino wistar rats 5 and 50 mg/kg of aniline for a month,examined the liver microsomal enzyme activity[13]. After one month of exposure to the higher dose of the substance (50 mg/kg body weight), it was discovered that the aniline boosts the activity of aniline p-hydroxylase and speeds up the N-demethylation of aminopyrine. The biotransformation of aniline in the perfused liver of male Sprague-Dawley rats at a dose of 50 mg/kgbw revealed in the study that aniline is hydroxylated in the liver to produce a variety of metabolites, some of which are trapped inside red blood cells. The liver is a detoxifying organ that breaks down harmful compounds and regulates the metabolism of medications[14]. The hepatic cell suffers substantial damage as a result of this breakdown, which is carried out by the endoplasmic reticulum of the hepatocytes[15]. The reported that liver of the male Swiss albino mice showed aggregation of lymphocyte infiltration, the large size of kupffer cells, some inflammatory cells, dilatation of portal vain, and proliferation of bile ductules after receiving 1/8 of 3,4 Dichloro aniline for 14,30,35 days. The liver displayed damaged hepatocytes, karyolysis of many nuclei, proliferation of bile ductules, dilatation, and congestion of the portal and portal vein with medium-dose therapy (1/16 LD50) [16]. One of the studysheds light on the mechanism of aniline-induced liver cell damage and apoptosis. Male Sprague-Dawley rat primary cultured hepatocytes were exposed to aniline at concentrations of 0, 1.25, 2, 50, 5.0, and 10.0 g/mL for 24 hours in the presence or absence of N-acetyl-L-cysteine (NAC). Aniline significantly reduced the levels of GSH (glutathione), CAT (catalase), SOD (superoxide dismutase) activity, and mitochondrial membrane potential in hepatocytes when compared to the negative control group, while significantly increasing the levels of reactive oxygen species and MDA (malondialdehyde). Aniline decreased cell viability and, in a concentration-dependent way, caused apoptosis[17]. Aniline was given to SPF Kunming mice intragastrically in a single dose of 100 mg/kg body weight. A single cell gel electrophoresis assay revealed that the tail length and tail movement of the hepatocyte DNA increased gradually from 8 hours after aniline administration, reached a maximum at 16 hours, and started to recover at 32 hours. The hepatocytes and peripheral blood lymphocytes were obtained at 3, 8, 16, 24, and 32 hours after aniline administration, respectively. As a result of these observations, aniline may have the potential to be genotoxic to hepatocytes and peripheral lymphocytes[18]. Their studies demonstrate that the hepatocytes and peripheral lymphocytes of mice have potent DNA repair machinery that guards against aniline toxicity.If male Big Blue F344 rats treated with parachloro aniline at doses of 15 mg/kg and aniline at a dose of 100 mg/kg experienced statistically significant increases in liver weight, while calcium and cholesterol levels were significantly higher compared to controls[6].

Aniline exposure has been shown in numerous studies to seriously harm the liver and affect normal liver metabolism, including an increase in liver size, changes in enzyme function, apoptosis, and DNA damage.

Table 1: Studies on effect of aniline on liver

Model organism	Dosage	Duration	Inference	Reference
Male albino rat (Wistar)	5 and 50 mg/kgbw	1 month	Increse activity of aniline p-Hydroxylase enzyme	Justyna*et al*., 1975
Male Sprague-Dawley rats	50 mg/kgbw	30 days	Formation of metabolites	Eyer*et al*., 1980
Male Swiss albino	1/32, 1/16 and 1/8	14,30,35 days	large size of kupffer	Eissa*et al*., 2012

mice	mg/kgbw of LD value		cells	
Male Sprague-Dawley rats	0, 1.25, 2.50, 5.0, and 10.0 μg/mL	24 h	apoptosis of hepatocytes	Wang *et al*., 2016
SPF Kunming mice	100 mg/kg body weight	3, 8, 16, 24, and 32 hours	Aniline is potential genotoxicant to hepatocytes	Gaopeng*et al*., 2016
Male Big Blue F344 rats	0.5, 15, or 100 mg/kgbw	28 days	increases in liver weight	Koenig *et al*., 2018

Effcet On Splenotoxicity

A hazardous aromatic amine known as aniline has been linked to both human and animal hemopoietic toxicity. As a result of chronic in vivo exposure to aniline, the spleen experiences a toxic reaction that is characterized by splenomegaly, hyperplasia, fibrosis, and tumor formation. Due to iron overload, oxidative DNA damage, and accelerated cell proliferation brought on by aniline exposure, the spleen may eventually develop tumors. In aniline-treated rats, the hematological system is largely damaged, resulting in methemoglobinemia, hemolysis, and hemolytic anaemia, which destroy erythrocytes [19,20,21]. After being administered in large doses to rats, aniline and a number of structurally similar aromatic amines cause spleen tumors. It is hypothesized that erythrocyte toxicity caused by compounds causes the spleen to accumulate toxic metabolites carried there by erythrocytes, deposit erythrocytes debris, particularly iron, which may catalyze tissue-damaging free-radical reactions, and induce splenic hyperplasia as a result of erythrocyte overload[2].

The spleen, particularly phagocytes, would be anticipated to play a significant role in eliminating aniline-damaged erythrocytes. Damaged erythrocytes will build up and then break down, releasing aniline and/or its metabolites and causing iron to build up in the spleen [23,24]. The splenic phagocytes, particularly the macrophages themselves, can activate during the scavenging of damaged erythrocytes and release reactive oxygen species (ROS) [24,25], which may further contribute to the oxidation of biomolecules resulting in further injury. Red pulp cellularity, erythrophagocytosis, and cellular fragmentation were all associated with dose-related histopathologic growth of the splenic red pulp[26]. Iron deposition in red pulp also increased significantly with dose. Aniline indicates lipid peroxidation and protein oxidation in the spleen, and Khan's 1997 work supports the idea that oxidative stress contributes to aniline's splenic toxicity. The dose-response relationship of the tumor incidence in male Fischer 344 rats at doses of 300, 400, or 500 mg/kg body weight was elucidated[27].

In male Fischer 344 rats, 300 mg/kg of aniline caused carcinogenicity in the spleen. In Fischer 344 rats, long-term feeding trials using aniline hydrochloride have shown a propensity for the formation of spleen tumors. As a result of the spleen being overloaded with erythrocyte debris at doses of 0.03 to 3.0 mM for 1, 2, and 4 h, the spleen tumours seen in rats are growing on chronic spleen injury[28]. Six male rats were treated to aniline hydrochloride at doses of 10, 30, or 100 mg/kg bw/day, After 1 and 4 weeks, the spleen was substantially enlarged in all of the animals in the high dose group, and histopathologically, this was linked to vascular congestion in the spleen at 100 and 30 mg/kg[29]. It was established that red pulp's sinus/venosus blood vessels had dilated, causing vascular congestion.

In a different investigation, 30 male Wistar rats were given conc. of 9.2, 32.4, 96.5, and 274.9 mg aniline/m3 through their noses for 6 hours per day, five days per week for two weeks. The findings of this study indicate that spleen weights at 90 and 270 mg/m3 considerably increased. At 30 mg/m3, extramedullary hematopoiesis in the spleen was noted[30]. Male Sprague-Dawley rats were subchronically exposed to aniline (0.5 mmol/kg/day by drinking water for 30 days), while controls received nothing but water[31]. Aniline therapy significantly increased the amount of splenic oxidative DNA damage, which was shown by an ELISA-measured 2.8-fold rise in 8-OHdG (8-Hydroxy-2-deoxyguanosine) levels. In a different experiment, male rats were gavaged with 1 mmol/kg/day of aniline in water for 1, 4, or 7 days. In the spleen, exposure to aniline significantly increased the expression of (heme oxygenase-1) HO-1 mRNA (2 and 2.4-fold at days 4 and 7, respectively), which was supported by ELISA and Western blot analysis. According to a related study, aniline poisoning may cause splenic toxicity and HO-1 up-regulation may cause oxidative damage. We concentrated on characterizing the nitrated proteins in the spleen of aniline-exposed rats in the study utilizing proteomic techniques. By using 2D Western blotting and an anti-3-nitrotyrosine-specific antibody, it was found that aniline exposure increased the tyrosine nitration of proteins in comparison to controls[32].

This result supports the idea that protein nitration contributes to aniline's ability to cause splenic toxicity[33]. Treatment with PCA at levels below 15 mg/kg and with aniline at a dose of 100 mg/kg led to statistically significant increases in spleen weights as compared to the vehicle control[34]. The weight of the spleen increased following aniline treatment in a statistically significant manner. Aniline-induced splenic toxicity is linked to oxidative DNA damage, and the production of DNAglycosylases is stimulated to treat oxidative DNA lesions in rats[34]. The transcriptional up-regulation of fibrogenic/inflammatory factors (cytokines IL-1, IL-6, and TNF-) is brought on by oxidative stress. Mitogen-activated protein kinases (MAPKs) and kinase phosphorylation are examples of upstream signalling events. A

fibrogenic and tumorigenic reaction in the spleen could be triggered by any of these occurrences. Aniline is harmful or carcinogenic to the blood, liver, and spleen, among other organs and systems of the human body[35].

Numerous studies have shown that exposure to aniline may have an impact on the spleen, resulting in methhemiglobinomia, splenomegaly, hyperplasia, iron accumulation in the spleen, and DNA damage in the spleen, which in turn causes the development of spleen tumors.

Table 2: Studies on effect of aniline on spleen

Model organism	Dose	Duration	Inference	Reference
Male Sprague-Dawley rats	600 ppm	30, 60, and 90 days	increases in Aniline hydroxylase and iron content of the spleen	Khan *et al*., 1993
Male Sprague-Dawley rats	1 mmol/kgbw	24 h	increases in the iron content of the spleen	Khan *et al*., 1995
Male Sprague-Dawley rats	0.7 mmol/ kgbw	14 days	iron accumulation in the spleen	Khan *et al*., 1995
Male SD rats	0.25, 0.5, 1, and 2 mmol/kgbw	24 hr	increased splenicred pulp cellularity, and cellular fragmentation	Khan et al., 1997
Fischer 344 rats	300, 400 or 500 mg/ kg body weight	24 and 48 h	induction of spleen tumors	Bomhard 2003
Male rats	10, 30 or 100 mg/kg bw/day	1 or 4 weeks	spleen was enlarged	Mellert*et al*., 2004
Male Wistar rats	9.2, 32.4, 96.5, and 274.9 mg aniline/m^3	6 h/day, 5days/week for two weeks	significantly increased spleen weights	Pauluhn 2004
Fischer 344 rats	0.03 to 3.0 mM	1,2,1 and 4 h	spleen damage with erythrocyte debris with DNA damage	Bomhard and Herbold 2005
Male Sprague–Dawley rats	0.5 mmol/kg/day	30 days	increase in splenic oxidative DNA damage	Ma *et al*., 2008
Male Sprague–Dawley rats	1 mmol/kg/day	1, 4, or 7 days	(heme oxygenase-1) HO-1 up-regulation in aniline-induced splenic toxicity	Wang *et al*., 2010
Rats	-	-	role of protein nitration in aniline-mediated splenic toxicity	Fan 2011
Wistar rats	50 and 100 mg/kgbw -	30 days	Reduced glutathione (GSH), and nitric oxide (NO) content) of spleen	Khan *et al*., 2014
Male Wistar rats	100 ppm	28 days	splenic phagocytes, and release reactive oxygen species (ROS)	Khairnar *et al*., 2016
Male Big Blue F344 rats	0.5, 15, 60, and 100 mg/kgbw	28 days	increases in spleen weights	Koenig *et al*., 2018
Rats	-	-	up-regulation of inflammatory factors (cytokines, IL-1, IL-6 and TNF-α)	Paurane*t al*., 2019

CONCLUSION

According to the previously mentioned mini-review on aniline's impact on the liver and spleen, aniline is toxic to both organs and causes serious harm to them, including apoptosis, genotoxicity, and changes in the liver microsome, splenomegaly, hyperplasia, fibrosis, and eventually the development of tumors in the spleen.

REFERENCES

[1]. H. Yen, W. Tsai, C. Chen, and S. Wang, "Fate of anilide and aniline herbicides in plant-materials-amendedsoils," Journalof Environmental ScienceHealth B,43(5): 382-389, 2008.
[2]. Y. Jiang,Y. Shang,J. Zhou,K. Yang, and H. Wang"Characterizationandbiodegradationpotentialofananiline-degradingstrainofPseudomonasJA1atlowtemperature," Desalinationand WaterTreatment,57: 25011-25017, 2016.
[3]. A. Edali, K. Patil, andS.Mulla, "An overview of aniline and chloroaniline compound as environmental pollutant," Bioscience and Bioengineering Communication, 1(4): 1-2, 2018.
[4]. X. Li, X. Jin, N. Zhao, I.Angelidaki, and Y. Zhang,"Efficient treatment of anilinecontaining wastewater in bipolar membrane microbial electrolysis cell-fenton system," WaterResearch,119: 67-72, 2017.
[5]. V. Mohonta and B.Mishra, "Integration of cancer and non-cancer human health riskassessmentforanilineenrichedgroundwater:Afuzzyinference system-basedapproach,"Environmental Geochemistry and Health, 42,3, 2020.
[6]. C. Koenig, C. Beevers, and K.Pant, "Assessment of the mutagenic potential of para-chloroaniline and aniline in the liver, spleen, and bone marrow of Big Blue® rats withmicronuclei analysis in peripheral blood," Environmental and Molecular Mutagenesis, 59(9): 785-797,2018.
[7]. J.Pauluhn, "Sub-acute inhalation toxicity of aniline in rats: Analysis of time-dependence and concentration-dependence of hematotoxic and Splenic Effects" Toxicological Sciences, 81(1): 198-215,2004. doi:10.1093/toxsci/kfh187
[8]. T.Kahl,K.Schroder,R.Lawrence,W.Marshall,H.Hoke, and R.Jackh, "Aniline," Ullmann's Encyclopedia of IndustrialChemistry,67: 1137,2007.
[9]. U.Otto, "Ueber das Verhalten der organischenKörper in höherenTemperaturen" [On the behaviour of organic substances at high temperatures], Annalen der Physik und Chemie., 8(11): 397-410,1826.
[10]. F.Runge, "UebereinigeProdukte der Steinkohlendestillation" (On some products of coal distillation), Annalen der Physik und Chemie, 31: 65-77,1834.
[11]. J. Fritzshe, "Uber das Anilin, einneuesZersetzungs product des Indigo" (On aniline, a new decomposition product of indigo). Bulletin Scientific, 7(12): 161- 165,1840.
[12]. A. Hofman, "Chemical investigation of organic basis in coal tar oil,"Annalen der Chemie und Pharmacie, 47: 37-87, 1843.
[13]. M.Justyna, " Effectofrepeatedexposuretoaniline,nitrobenzene,andbenzeneonlivermicrosomalmetabolism inthe rat,"BritishJournal ofIndustrialMedicine, 32: 42-48,1975.
[14]. P.Eyer, H.Kampffmeyer, and H.Maister,"Biotransformation of nitrobenzene, phenylhydroxylamine,andanilineintheisolatedperfusedratliver,"XenobioticatheFateofForeignCompound in Biological System, 10(7): 499-516,1980.
[15]. S.Sharma, G.Goyal, and A.Sharma,"Toxicity of tomato red, a popular food dye blend on male albino mice," Experimental and Toxicologic Pathology, 60: 51-57,2008.
[16]. F.Eissa, A.Makawy, M.Badr, and O.Elhamalawy, "Assessment of 3, 4-Dichloroaniline toxicity as environmental pollutant in male mice," European Journal of Biological Sciences 4 (3): 73-82,2012.
[17]. Y.Wang, H.Gao, X.Lin, S.Dong, H.Dong, J.Jia and Y.Wu, "Anilineinduced oxidative stress and apoptosis of primary cultured hepatocytes," International JournalofEnvironmental Research and PublicHealth, 13(12): 1188,2016.
[18]. B. Gaopeng, L.Ruixiang, S.Baozhong, and J.Haihua,"DNA damage in mouse hepatocytes and lymphocytes induced by aniline and their repair dynamics,"ActaLaboratoriumAnimalisScientinaSinica, 6: 139-144,2016.
[19]. J.Harrison andD.Jollow, "Roleofanilinemetabolitesinaniline-inducedhemolyticanemia,"TheJournal of Pharmacology and ExperimentalTherapeutics, 238(3): 1045-1054,1986.
[20]. Y.Kim and GCarlson. "The effect of an unusualworkshift on chemical toxicity," Toxicoogical Sciences, 7(1): 144–152,1986. doi:10.1093/toxsci/7.1.144
[21]. F.Khan, S.Kaphalia, J. Boor and A.Ansari, "Subchronic toxicity of aniline hydrochloride in rats," Archives of Environmental Contamination and Toxicology,24(3): 368-374,1993. http://dx.doi.org/10.1007/BF01128736.
[22]. S.Bus and A.Popp,"Perspectives on the mechanism of action of the splenic toxicity of aniline and structurally-related compounds," Food Chemical Toxicology, 25(8): 619-626,1987.
[23]. F.Khan, S.Kaphalia, J.Boor and A. Ansari, "Subchronic toxicity of aniline hydrochloride in rats," Archives of Environmental Contamination and Toxicology,24(3): 368-374,1993. http://dx.doi.org/10.1007/BF01128736.
[24]. F. Khan, S.Kaphalia, and A. Ansari, "Erythrocyte-aniline interaction leads to their accumulation and iron deposition in rat spleen," Journal of Toxicology and Environmental Health, 44(4): 415-421, 1995. http://dx.doi.org/10.1080/15287399509531970. PMID: 7723074.
[25]. U.Khairnar, A.Upaganlawar and C.Upasani, "Ameliorative Effect of Chronic Supplementation of Protocatechuic Acid Alone and in Combination with Ascorbic Acid in Aniline Hydrochloride Induced Spleen Toxicity in Rats," Scientifica,1-9,2016. doi:10.1155/2016/4306984
[26]. F.Khan,J.Boor,Y.Gu,W.Alcockand A.Ansari,"Oxidativestressinthesplenotoxicityof aniline,"Fundamental andApplied Toxicology, 35: 22-30,1997.
[27]. M.Bomhard,"High-dose clastogenic activity of aniline in the rat bone marrow and its relationship to the carcinogenicity in the spleen of rats," Archives of Toxicology, 77: 291-297,2003.

[28]. E.Bomhardand B.Herbold, "Genotoxic activities of aniline and its metabolites and theirrelationshipto thecarcinogenicity," CriticalReview inToxicology, 35(10): 783-835,2005.
[29]. W.Mellert, K.Deckardtand, C.Gembardt, "Aniline: Early indicators of toxicity in malerats and their relevance to spleen carcinogenicity," Human and Experimental Toxicology,23(8): 369-419,2004.
[30]. J.Pauluhn, "Sub-acute inhalation toxicity of aniline in rats: Analysis of time-dependence and concentration-dependence of hematotoxic and Splenic Effects," Toxicological Sciences, 81(1): 198-215,2004. doi:10.1093/toxsci/kfh187
[31]. H.Ma, J.Wang, Z.Abdel-Rahman, J.Boor and F. Khan, "Oxidative DNA damage and its repair in rat spleen followingsubchronic exposure to aniline," Toxicology and Applied Pharmacology, 233(2): 247-253,2008. doi:10.1016/j.taap.2008.08.010
[32]. J.Wan, H.Ma, P.Boor, S.Ramanujam, Ansari G A S, and Khan M F. 2010. Up-regulation of heme oxygenase-1 in rat spleen after aniline exposure. Free Radical Biology and Medicine, 48(4): 513-518.
[33]. X.Fan, J.Wang, V.Soman, S.Ansari and F.Khan, "Aniline-induced nitrosative stress in rat spleen: Proteomic identification of nitrated proteins," Toxicology and Applied Pharmacology, 255(1): 103–112,2011. doi:10.1016/j.taap.2011.06.005
[34]. M.Pouran, H.Hossini and G.Ashraf, "Molecular mechanism of aniline induced spleentoxicityandneurontoxicityinexperimentalratexposure:AReview,"CurrentNeuropharmacology,17(3): 201-213,2019.
[35]. Y.Feng, L.Chen, T.Zhang, H.Wu, L.Song and Y. Wang, "Research progress on health effects of aniline,"Chinese Journal of Preventive Medicine**,** 54(2): 213-218,2020.