RESEARCH OPINIONS IN ANIMAL & VETERINARY SCIENCES
PRINT ISSN 2221-1896, ONLINE ISSN 2223-0343
www.roavs.com

Acute toxicity study of alkaloids of *Datura stramonium* seeds in rat

Bouzidi Abdelouahab[1], Mahdeb Nadia[1,2] and Kara Nabila[2]

[1]Department of Biochemistry, Faculty of natural sciences and life, Ferhat Abbas University, Setif 19000 Algeria
[2]Department of Agronomy, Faculty of natural sciences and life, Ferhat Abbas University, Setif 19000 Algeria

Abstract

The effects of acute administration of total alkaloids of *Datura Stramonium* seeds were studied in male and female Albino-Wister rats. After acute i.p administration of dose 100 mg/kg (1/4 DL_{50}) of total alkaloids of *Datura stramonium*, there were no remarkable changes in general appearance and no deaths occurred in any experimental group in both sexes. The weight of treated animals has seen a normal evolution in comparison with that of control animals. Twenty four hours after the treatment, a significant reduction in the relative weight of liver was observed in treated rats. The RBC, HCT, HGB and WBC were significantly higher in the treated male rats than the control group. However, the treated female rats showed a significant decrease in RBC, HCT and HGB. There were no statistical differences in GOT, GPT and ALP between male rats at different intervals. However, an increase of transaminases was observed in female rats treated after the 1st day. The results showed that the acute toxicity of Datura *stramonium* had no deleterious effect on these parameters in male and female rats.

Keywords: *Datura stramonium*, Rat, Liver, Alkaloids, Toxicity

Introduction

The solonacies plant, *Datura stramonium*, is locally known as "Sikrane" and is prevalent in north Algeria, usually matures between May and October (Bouzidi et al., 2000). *Datura stramonium* is known to contain toxic tropane alkaloids, including the pharmacologically active compounds atropine and scopolamine and their concentrations are variable with part of the plant and harvest times (Dugan et al., 1989; Friedman and Levin, 1989; Miraldi et al., 2001; Berkov et al., 2006; Chollet et al., 2010). The plant is used in traditional medicine to treat headaches and asthma (Gidado et al., 2007; El Bazaoui et al., 2009).

Datura stramonium (known also as thorn apple, Jimson weed and Jamestown weed), is mostly abused by younger teens (Djibo and Bouzou, 2000; Cohen et al., 2003; Prado, 2004; Forrester, 2006; Benghezala et al., 2011). There are also a few reports of accidental poisoning in children (Salah and Awad, 1984; Bouzidi et al., 2000; Al-Shaikh and Sablay, 2005) The adverse clinical effects of Datura intoxication are varied and may consist of dry mouth and extreme thirst, dryness of the skin, pupil dilation and impaired vision, urinary retention, rapid heart beat, confusion, restlessness and hallucinations (Chan, 2002; Marc et al., 2007).

The *Datura stramonium* has been involved in accidental poisoning of animals of farm livestock and poultry. The plant was mistakenly gathered together with corn intended for animal feeds (Nelson et al., 1982; Kara et al., 2009; Botha and Penrith, 2009; Guitart et al., 2010) and companion animals (Tostes, 2002; Berny et al., 2010).

Moreover few experimental studies in domestic and laboratory animals have been performed to evaluate the effect of this plant in different species such as horses (Binev et al., 2006; Gerber et al., 2006) and rats (Dugan et al., 1989; Gidado et al., 2007). Therefore, a study was design to find the effect of this plant on some important parameters in male and female rats.

Materials and Methods

Seeds of *Datura stramonium* were collected in south Setif (east Algeria) between August and September (Fig. 1). The seeds were stored at room temperature in dry place. After drying, the seeds were kept in tightly–closed containers prior to use.

100 g of air dried powdered of seeds was defatted with petroleum ether under reflux and then the seeds were witted with150 ml of NH_4OH (25%, m/m) for 4 hours and were extracted to exhaustion with $CHCl_3$ using a soxhlet apparatus for 6 hours. The organic extract (containing free alkaloids + lipophilic impurities) was shaken three times with 150 ml aqueous sulphuric acid (2 %). The acid extracts (alkaloids salts)

were treated three times with 50 ml NH_4OH (25 %) to pH 10 to liberate the free alkaloids which were separated by extraction with 150 ml CH_2Cl_2, and then dried with Na_2SO_4 and concentrated under reduced pressure to obtain crude alkaloids, according to the method described by Bruneton (1999). The yield of this extract was approximately 0.089 ± 0.02 % (w/w).

A Pyeunicam HPLC was used to quantify atropine and scopolamine in total alkaloids of *Datura stramonium* seeds. The HPLC system consisted of a isocratic pump, a rheodyne injector equipped with a 20 µl sampling loop and photodiode array detector. Separation was achieved by a Varian/Chrompack column C_{18} (125 x 4 mm i.d, particle size 5 µm) proceeded by a guard column, at temperature 35°C. The mobile phase (delivered at a flow rate of 1 ml/min) consisting of acetonitrile – phosphate buffer (pH 3.8) (15/85, v/v), was filtered through a 0.45 µm membrane and degassed before use, as designed by Kirchhoff et al. (2004).

Male and female Albino-wistar rats weighing between 200–250 g were purchased from animal center of Pasteur's Institute (Algiers – Algeria). Animals were housed in hanging transparent plastic cages (55 x 33 x19cm) in the animal room of Faculty of Sciences University Ferhat Abbas Setif Algeria and acclimated for 3 weeks prior to experiment. The litter was renewed every 3 days. They were fed with a standard pellet and tap water *ad libitium.* All animals were kept in standard environmental conditions. Each animal was identified by body marks using 1% picric acid solution. Animals were deprived of food but not water 16 hours prior administration of the test substances. All experimental procedures were conducted in accordance with the guide for care and use of laboratory animals and in accordance with the scientific council of the Faculty of Natural Sciences and Life of the University Ferhat Abbas, Setif – Algeria.

Two groups of 10 albino-wistar rats of each sex were given single dose of 100 mg/kg (1/4 DL_{50}) body weight of total alkaloids of seeds of *Datura stramonium* by intraperitoneal route (Antov et al., 1991). The control group (10 rats) received saline water with few drops of ethanol at the same volume. Animals were observed and recorded systematically at 1, 2, 3, 4, 5, and 6 hour and daily after test substance administration. The visual observations included changes in skin and hair, eyes, mucous membranes and also respiratory, circulatory, autonomic and central nervous system.

At the end of all experimental periods, the first group (of each sex) was sacrificed after 24 hour of treatment and the second group after 5 days. Animals were anaesthetized with urethane at the dose 760 mg/kg. Two blood samples were obtained from the retro-orbital vein: a sample for hematology containing ethylenediaminetetraacetic acid with apparatus Medonic (Beckman Coulter – USA) and sample for serum and used for measurement of activities Glutamic-oxaloacetic transaminase (GOT), Glutamic-Pyruvic Transaminase (GPT) (using commercial Kits – SGM Rome-Italy) and alkaline phosphatase (ALP)(using commercial Kits–Cypress Diagnostic Langdrop– Belgium) with apparatus Technicon RA-1000-USA.

After blood collection, the animals were sacrificed by cervical dislocation. After autopsy, all tissues were examined grossly and major's organs (liver, brain, heart, kidneys, Spleen, testicles, and lung) were weighted. The relative organ weight of control and treated was calculated. Tissues from liver of all animals were fixed in 10% buffered formalin solutions then embedded in paraffin and cut with a microtome set at 5 µm, stained with hematoxylin and eosin and examined by light microscopy for histopathological changes.

Statistical analysis

The statistical significance of differences between means was calculated using one-way ANOVA followed by Turkey's test for multiple comparisons with control group.

Results

The representative chromatograms for tropane alkaloids of *Datura stramonium* are shown in Fig. 3. The retention times for scopolamine and atropine were 8.96 and 15.98 min, respectively. The concentrations of atropine and scopolamine were 4 and 2 mg/100g of *Datura stramonium* seeds respectively.

The results of the study of animals administered with single dose of 100 mg/kg body weight of total alkaloids did not show any toxic symptoms such as immediately after injection or at the end of 5 days.

There were increases in all the groups, but significant differences were detected in the weight gain of female and male rats treated with plant, as compared to control groups during acute toxicity (Table1 and 2). The weight gain of female rats was lower compared with that of male rats. The effects of total alkaloids of *Datura stramonium* on relative organ weights are presented in tables 3 and 4. Female treated rats showed a significant reduction in liver relative weight on the 1st day and lungs relative weight on the 5th day. However, there was significant decrease in the relative weight of liver, brain and spleen on the 1st day of male rats treated with plant as compared to control group.

The haematological parameters of the rats treated with total alkaloids are presented in Table 5 and 6. The RBC (red blood cells), HCT (Haematocrit), HGB (Haemoglobin) and WBC (white blood cells) were significantly higher in all the treated male groups than the control group. However, there was significant

Fig. 1: Plant of *Datura stramonium*

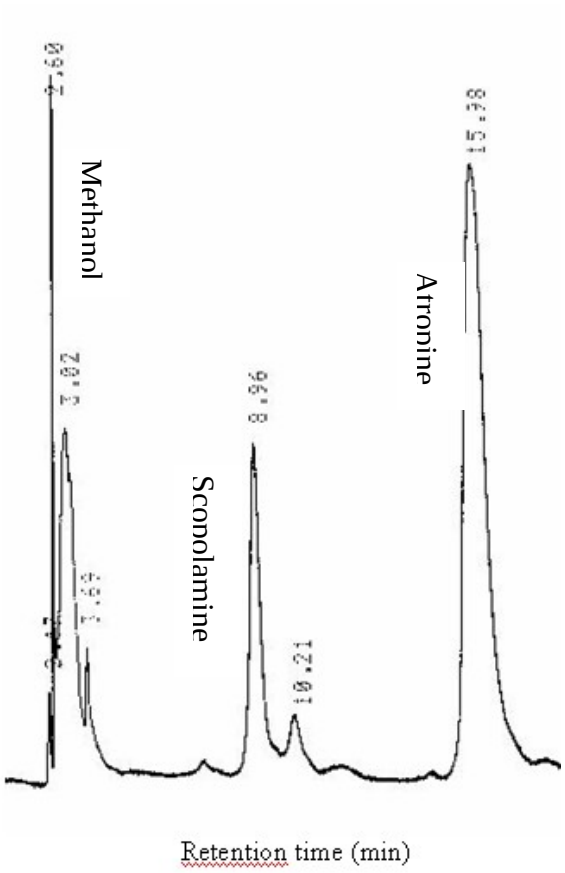

Fig. 2: HPLC chromatogram of the total alkaloids from the seeds of *Datura stramonium*

decrease in the RBC, HCT and HGB of female rats treated with total alkaloids *Datura stramonium* seeds.

It was observed that the values of GOT, GPT and ALP on day 1 and 5 were comparable with the values of the control and treated male groups. There were no statistical differences in GOT, GPT and ALP observed between groups. However, female rats showed a significant increase in GOT and elevated but not significant GPT after 1st day, and return to normal values after the 5th day (Fig. 3 & 4).

The histological examination of liver from rats was performed in both control and treated groups (not shown). All the sampling tissue sections showed no histopathological changes. They were within normal limits. Neither degenerative nor infiltrative lesions were observed.

Discussion

The *Datura stramonium* has been used in folklore and traditional systems of medicine, as a cure for the asthma (Pretorius and Marx, 2006), stimulation of central nervous system**,** treatment of dental and skin infections (Gidado et al., 2007). *Datura stramonium* (called Jimson weed) has been reported as a drug of abuse (Chan, 2002; Birmes et al., 2002; Aroukou et al., 2003). *Datura stramonium* is known to contain highly toxic tropane alkaloids, including the pharmacologically active compounds atropine and scopolamine (Dugan et al., 1989; Desachy et al., 1997).

HPLC is one of the most commonly used techniques to analyze alkaloids (Steenkamp et al., 2004). HPLC enabled to evaluate the content of the two mains alkaloids. The atropine concentration was higher than that of scopolamine. The separation and values of scopolamine and atropine were in agreement with the ranges previously reported in available literature (Miraldi et al., 2001; Friedman, 2004).

The toxic effects of a test substance can be basically determined by physical examination, daily observation, visual examination, food and water consumption, body and organ weight, haematology, urinalysis, biochemical organ function tests and pathology studies (Stevens and Gallo, 1989).

The liver is known to be key organ in the metabolism and detoxification of xenobiotic, is vulnerable to damage induced by a huge variety of chemicals as reported by Udem et al. (2009). An obvious sign of

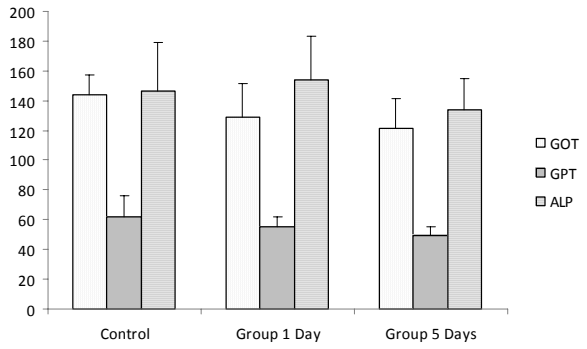

Fig. 3: Effect of acute administration of *Datura stramonium* (100 mg/kg) on GOT, GPT and ALT in male rats

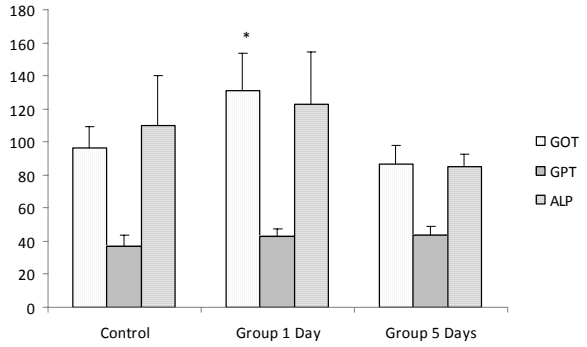

Fig. 4: Effect of acute administration of total alkaloids of seeds of *Datura stramonium* (100 mg/kg) on GOT, GPT and ALP in female rats. *indicate significant difference at P<0.05

hepatic injury is leakage of cellular enzyme into plasma. When the liver cell membrane is damaged, a variety of enzymes normally located in the cytosol are released into blood stream. The estimation of the GPT (glutamic-pyruvic transaminase) and GOT (glutamic-oxaloacetic transaminase) in the serum is a useful quantitative marker for the extent and type of hepatocellular damage reported by Kumar et al. (2004). An increase in the level ALP (alkaline phosphatase) is an indication of biliary obstruction (Udem et al., 2009). The male group treated with 100 mg/kg total alkaloids did not show change in the level of these enzymes after the 1st and 5th day. However, female rats showed a significant increase in GOT and elevated but not significant GPT after 1st day and returned to normal values after the 5th day. This could be explained by rapid metabolism and excretion of alkaloids and their metabolites (Hardman et al., 1998).

Hematological studies showed a significant increase in red blood cells, hematocrit and hemoglobin after the 1st and 5th day in male rats, this could be explained by a need of oxygen to repair the injury as a result of the toxicity of total alkaloids fro*m Datura stramonium* and a significant decrease in female rats during the same period of experimentation.The lower of RBC may be attributed to the increase in GPT and GOT. Increased GPT and GOT are indicative of liver damage. If that is the case, serum protein synthesis in the liver will be suppressed and the bone marrow may not have enough proteins to synthesize RBC. Also, these haematological alterations suggest possible dehydration as reported by Dugan et al. (1989). Ours resultants show that the total alkaloids of *Datura stramonium* are able to produce alterations on erythrocyte parameters.

Microscopic examinations of treated-groups show no histopathological changes. It is likely that rat quickly eliminates the alkaloids of *Datura stramonium.*

This study shows that female rats are slightly more sensitive than male rats to total alkaloids of *Datura stramonium* seeds under conditions of acute toxicity.

Acknowledgements

We would like to think ANDRS, Oran Algeria, for financial support of the project. We also thank Prof Touabti Director of Central Laboratory and the staff of the laboratory of anatamo-pathology of the Hospital of Setif Algeria.

Table1: Effect of acute administration of *Datura stramonium* (100 mg/kg) on body weight of male rats

Group	1st Day	5th Day	Difference
Group sacrificed after 1 day	208.1 ± 37[b]		
Group sacrificed after 5 days	210.1 ± 10.81[a]	235.2 ± 8.43	25.1 ± 2.83[a]
Group control	239.8 ± 22.58[a]	254.5 ± 15.39	14.70 ± 10.97[b]

Mean in the columns with different superscripts are significantly different at (P<0.05)

Table 2: Effect of acute administration of *Datura stramonium* (100 mg/kg) on body weight of female rats

Group	1st Day	5th Day	Difference
Group sacrificed after 1 day	213.30 ± 18.31		
Group sacrificed after 5 days	215.4 ± 9.68	235.2 ± 8.43	4.6 ± 3.95[a]
Group control	210.2 ± 20.43	222.7 ± 19.11	12.50 ± 6.55

Mean in the columns with different superscripts are significantly different at (P<0.05)

Table 3: Relative organ weights of male rats treated with 100 mg/kg of *Datura stramonium*

Group	Liver	Brain	Kidney	Lungs	Heart	Spleen	Testes
Group control	0.0396± 0.00241	0.00889± 0.00117[a]	0.00705± 0.00076	0.00818± 0.00223	0.00379± 0.00047	0.00452± 0.00071[a]	0.0087± 0.00209
Group sacrificed after 1 Day	0.0358± 0.00421	0.00763± 0.00063[b]	0.00695± 0.00749	0.00936± 0.00417	0.00388± 0.00036	0.00384± 0.00034[b]	0.00738± 0.00222
Group sacrificed after 5 Days	0.0398± 0.00371	0.00889± 0.001[a]	0.00749± 0.00254	0.00805± 0.00136	0.00395± 0.00057	0.00566± 0.00162[a]	0.0106± 0.00205

Mean in the columns with different superscripts are significantly different at (P<0.05)

Table 4: Relative organ weights of female rats treated with 100 mg/kg of *Datura stramonium*

Group	Liver	Brain	Kidney	Lungs	Heart	Spleen
Group control	0.0381± 0.00456[a]	0.00853± 0.00097	0.00734± 0.00107	0.00967± 0.00241	0.00393± 0.000298	0.00469± 0.000767
Group sacrificed after 1 Day	0.0342± 0.00239[b]	0.00858± 0.000538	0.0073± 0.001	0.00764± 0.00565	0.00391± 0.000295	0.00447± 0.000593
Group sacrificed after 5 Days	0.0398± 0.00282[a]	0.00891± 0.000645	0.00702± 0.000585	0.00738[a]± 0.00155	0.00390± 0.000332	0.00477± 0.000766

Mean in the columns with different superscripts are significantly different at (P<0.05)

Table 5: Effect of acute administration of *Datura stramonium* (100 mg/kg) on some haematological parameters in male rats

Group	RBC 10^6/mm^3	MCV	RDW	HCT%	PLT 10^6/mm^3	MPV	WBC 10^6/mm^3	HGB g/l
Control	7.83±0.56[b]	52.75±3.23	16.11±1.86	41.26±2.30[b]	431±73.92	6.6±0.31[b]	8.56±2.69[b]	13±0.8[b]
Group 1st day	8.46±0.56[a]	52.09±0.98	14.76±1.14	44.67±2.29[a]	487.88±85.59	7.22±0.14[a]	10.51±1.42[a]	14.14±0.63[a]
Group 5th day	8.47±0.66[a]	53.77±2.69	16.13±0.95	46±2.51[a]	433±73.29	6.81±0.23[b]	10.00±2.40[a]	14.1±0.44[a]

Mean in the column with different superscripts are significantly different at (P<0.05). RBC, red blood cell; MCV, mean corpuscular volume; RDW, red cell distribution width; HCT, hematocrit; PLT, platelets; MPV, mean platelet volume; WBC, white blood cell; HGB, hemoglobin.

Table 6: Effect of acute administration of *Datura stramonium* (100 mg/kg) on some haematological parameters in female rats

Group	RBC 10^6/mm^3	MCV	RDW	HCT%	PLT 10^6/mm^3	MPV	WBC 10^6/mm^3	HGB g/l
Control	8.10± 0.46[a]	54.06± 2.06	13.18± 1.06	43.71± 2.06[a]	464.12± 51.39	7.29± 0.17	10.37± 1.04	14.35± 0.43[a]
Group 1st day	6.51± 0.30[b]	53.15± 1.91	12.03[a]± 0.85	34.62± 1.71[b]	411.78± 37.67	6.6[a]± 0.25	10.02± 1.6	12.1[b]± 0.57
Groupe 5th day	7.11± 0.72[b]	54.18± 0.6	14± 0.99	38.54± 3.95[b]	536.87± 136.62	7.28± 0.27	9.53± 2.46	13.22± 1.00[b]

Mean in the column with different superscripts are significantly different at (P<0.05) RBC, red blood cell; MCV, mean corpuscular volume; RDW, red cell distribution width; HCT, hematocrit; PLT, platelets; MPV, mean platelet volume; WBC, white blood cell; HGB, hemoglobin.

References

Al-Shaikh, A.M. and Sablay, Z.M. 2005. Hallucinogenic plant poisoning in children. *Saudi medical journal*, 26: 118-121

Antov, G., Zaikov, C., Bouzidi, A., Mitova, S., Michaelova, A., Halkova, J. and Choumkov, N. 1991. Biochemical and histological changes after acute oral poisoning with the acetanilide herbicide acetochlor. *Journal de toxicologie clinique et experimentale*, 11: 349-356.

Aroukou, H., Matray, M.D., Bragança, C., Mpaka, J.P., Chinello, L., Castaing, F., Bartou, C. and Poisot, D. 2003. Voluntary poisoning by ingestion of *Datura stramonium*. Another cause of hospitalization in youth seeking strong sensations.

Annales de médecine interne, 154:1S46-1S50.

Ben Ghezala, H., Kouraichi, N., Brahmi, N., El Ghord, H., Thabet, H. et Amamou, M. 2011. Intoxication aiguë par les plantes. *Journal européen des urgences*, 21: 266.

Berkov, S., Zayed, R. and Doncheva, T. 2006. Alkaloid patterns in some varieties of *Datura stramonium*. *Fitoterapia*, 77: 179-182.

Berny, P., Caloni, F., Croubels, S., Sachana, M., Vandenbroucke, V., Davanzo, F. and Guitart, R. 2010. Animal poisoning in Europe. Part 2: Companion animals. *The Veterinary Journal*, 183: 255-259.

Binev, R., Valchev, I. and Niklov, J. 2006. Hematological studies on Jimson weed (Datura stramonium) intoxication in horses. *Trakia Journal of Science*, 4: 43-48.

Birmes, P., Chounet, V., Mazerolles, M., Cathala, B., Schmitt, L. and Lauque, D. 2002. Self-poisoning with *Datura stramonium*: 3 case report. *Presse médicale*, 30: 69-72.

Botha, C.J. and Penrith, M.L. 2009. Potential plant poisonings in dogs and cats in southern Africa. *Journal of the South African Veterinary Association*, 80: 63-74

Bouzidi, A., Mahdeb, N., Allouche, L. and Houcher, B. 2002. Etudes épidémiologiques sur les plantes toxiques dans les régions de Sétif et Bordj Bou Arreridj. Algérie. *Bulletin d'information Toxicologique.* 18: 5-10.

Bruneton, J. 1999. Pharmacognosie, phytochimie, plantes médicinales. 1ère édition. Lavoisier, Paris.

Chan, K. 2002. Jimson Weed Poisoning–A Case Report. *The Permanente journal*, 6: 28-30.

Chollet S, Papet Y, Mura P. and Brunet B. 2010. Détermination des teneurs en atropine et scopolamine de différentes espèces sauvages et ornementales du genre *Datura*. *Annales De Toxicologie Analytique*, 22:173-179.

Cohen, S., Berny, C., Meyran, S., Mialon, A. and Manchon, M. 2003. Intoxication volontaire par une tisane de feuilles de Datura. *Annales De Toxicologie Analytique*, 15: 287-291.

Desachy, A., Frois, B., Vignon, P., Roustan, J. and Gay, R. 1997. Uncommon poisoning by *Datura stramonium*; two case reports. *Réanimation et Urgences*, 6: 51-53.

Djibo, A. and Bouzou, S.B. 2000. Acute intoxication with "sobi-lobi" (Datura). Four cases in Niger. *Bulletin de la Société de Pathologie Exotique*, 4: 294-297.

Dugan, G.M., Gumbmann. M.R. and Friedman, M. 1989. Toxicological evaluation of Jimson Weed (*Datura stramonium*) seed. *Food and Chemical Toxicology*, 27: 501-510.

El Bazaoui, A., Stambouli, H., Bellimam, M.A. and Soulaymani, A. 2009. Détermination des alcaloïdes tropaniques des graines du *Datura stramonium* L. par CPG/SM et CL/SM. *Annales de toxicologie analytique*, 21: 183-188.

Forrester, M.B. 2006. Jimsonweed (*Datura stramonium*) exposures in Texas, 1998-2004. *Journal of Toxicology and Environmental Health*, 69: 1757-1762.

Friedman, M. and Levin, C.E. 1989. Composition of Jimson Weed (*Datura stramonium)* Seeds. *Journal of Agricultural and Food Chemistry*, 37: 998-1005.

Friedman, M. 2004. Analysis of biologically active compounds in potatoes *(Solanum tuberosum)*, tomatoes (*Lycopersicon esculentum*), and jimson weed (*Datura stramonium*) seeds. *Journal of Chromatography*, 1054: 143 – 155.

Gerber, R., Naudé, T.W. And De Kock, S.S. 2006. Confirmed Datura poisoning in a horse most probably due to *Datura ferox* in contaminated tef hay. *Journal of the South African Veterinary Association*, 77: 86-9.

Gidado, A., Zainab, A.A., Hadiza, M.U., Serah, D.P., Anas, H.Y. and Milala, M.A. 2007. Toxicity studies of ethanol extract of leaves of *Datura stramonium* in rats. *African Journal of Biotechnology*, 6: 1012-1015.

Guitart, R., Croubels, S., Caloni, F., Sachana, M., Davanzo, F., Vandenbroucke, V. and Berny, P. 2010. Animal poisoning in Europe. Part 1: Farm livestock and poultry. *The Veterinary Journal*, 183: 249-254.

Hardman, J.G., Limbird L.E., Molinoff Ruddon R.W. and Gilman A.G.1998. Les bases pharmacologiques de l'utilisation des médicaments. McGraw-Hill, London.

Kara, N., Bouzidi, A., Mahdeb, N. and Djellal, F. 2009. Bilan de l'intoxication des animaux d'élevage par *Datura stramonium* dans la région de Sétif, Algérie. *Livestock Research for Rural Development*, 20 (11).

Kirchhoff, C., Bitar, Y., Ebel, S. and Holzgrabe, U. 2004. Analysis of atropine, its degradation products and related substances of natural origin by means of reversed-phase high-performance liquid chromatography. *Journal of Chromatography A*, 1046: 115-120.

Kumar, G., Sharmila B.G., Vanitha P., Sundararajan, M. and Rajasekara, P. M. 2004. Hepatoprotective activity of *Trianthema portulacastru rats* against paracetamol and thioacetamide intoxication in albino. *Journal of Ethnopharmacology*, 92: 37-40.

Marc, B., Martis, A., Moreau, C., Arlie, G., Kintz, P. and Leclerc, J. 2007. Intoxications aiguës

à *Datura stramonium* aux urgences. *La Presse Médicale*, 36 :1399-1403.

Miraldi, E., Masti, A., Ferri, S. and Barni C.I. 2001. Distribution of hyoscyamine and scopolamine in *Datura stramonium. Fitoterapia,* 72: 644-648.

Nelson, P.D., Mercer, H.D., Essig, H.W. and Minyard, J.P. 1982. Jimson weed seed toxicity in cattle. *Veterinary and Human Toxicology*, 24: 321-325.

Prado, P. 2004. Le Jilgré (*Datura stramonium*). Une plante hallucinogène, marqueur territorial en Bretagne morbihannaise. *Ethnologie Française*, 34: 453-461.

Pretorius, E. and Marx, J. 2006. *Datura stramonium* treatment and possible effects on prenatal development. *Environmental Toxicology and Pharmacology*, 2: 331-337

Salah, A.T. and Awad, H.M.1984. Datura intoxication in Riyadh**.** *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 78:1134-135.

Steenkamp, P.A., Harding, N.M., Van Heerden, F.R. and Van WYk, B.E. 2004. Fatal Datura poisoning: identification of atropine and scopolamine by high performance liquid chromatography/photodiode array/mass spectrometry. *Journal Forensic Science International*, 145: 31–39.

Stevens, K.R. and Gallo, M.A. 1989. Practical considerations in the conduct of chronic toxicity studies: In A.W. Hayes (Editor), Principles and Methods of Toxicology. Raven Press, New York, pp. 237-250.

Tostes, R.A. 2002**.** Accidental *Datura stramonium* poisoning in a dog. *Veterinary and Human Toxicology*, 44: 33-34.

Udem, S.C., Obidoa, O. and Asuzu, I.U. 2009. Acute and chronic toxicity studies of *Erythrina senegalensis* DC stem bark extract in mice. *Comparative Clinical Pathology*, 19: 275-282.