Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats

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Article ID: WMC001758

Article Type: Research articles

Submitted on: 14-Mar-2011, 09:04:22 PM GMT Published on: 16-Mar-2011, 10:19:53 PM GMT

Article URL: http://www.webmedcentral.com/article_view/1758

Subject Categories: CYTOLOGY

Keywords: Tested Plants, Cleome Droserfolia (sommo), Juniperus Phoenicia (araar), Anostatica Hierochuntica (kafta) and Hyphened Thebaica (doum), Alloxan, Pregnant Females Albino Rats, Diabetes Mellitus

How to cite the article: Salah S H, Abdou H S, Abd El Raouf A . Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats . WebmedCentral: CYTOLOGY 2011;2(3):WMC001758

Source(s) of Funding:

No funding
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Abstract

The influences of medicinal plants *Juniperus Phoenicea* (Araar), *Hyphaene thebaica* (Doum), *Anastatica hierochuntica* (Kafta) and *Cleome droserifolia* (Sammo) as antidiabetic agents were investigated using female pregnant albino rats. Female rats were injected with 60 mg/kg b.w. alloxan to induce diabetes. Diabetic rats treated orally with the methanol extracts of tested plants till the 19 day of gestation. The present studies include the frequencies of chromosomal aberrations and nucleic acid system of liver in the female pregnant rats and their embryos. The results showed that injection of alloxan caused highly significant increase in chromosomal aberrations as well as in blood glucose levels as a result of diabetes in pregnant females. It also caused a high incidence of chromosomal deviation in embryos and decreased the liver soluble protein contents of female rats and their embryos. These effects in alloxanized animals were treated and improved by ingestion of the methanol extracts of the tested plants (Araar, Doum, Kafta and Somma) in which under their treatments, the increased level of blood glucose of diabetic rats was deceased. Ingestion with the plants methanolic extracts improved and normalized the effects of diabetes in nucleic acids values of liver tissues. These were accompanied with nucleases (RNAase and DNAase) activities. The inhibited activities of both DNAase and RNAase of pregnant rats and their embryos were stimulated and readjusted around the normal values. Also administration of the plants methanol extracts decreased the percentage of chromosomal aberrations in the female rats and embryos. It is concluded that there are some biochemical dynamics which might occur in the metabolism of glucose, nucleic acids and proteins in order to prevent or to reduce the oxidative stress of diabetes by flavonoids treatment.

Introduction

Diabetes mellitus has become one of the most frequent complications – accompanying chronic diseases with high morbidity and mortality (Adeghate, 1999 b). It is a metabolic disorder characterized by hyperglycemia resulting from lack of insulin, lack of insulin effect, or both. Two types of diabetes are recognized; type 1, known as insulin – dependent diabetes (IDDM), and type II, known as non – insulin – dependent diabetes (NIDDM) (Hansen, 1998). In type I diabetes, the absolute deficiency of insulin attributed to β–cell dysfunction leads to hyperglycemia, enhanced lipolysis, protein catabolism and ketosis (Hansen, 1998). Type II diabetes impairs the ability of insulin to stimulate both muscle glucose uptake and splanchnic glucose uptake, thus contributes to induce hyperglycemia (Basu et al., 2000). The severity and frequency of the late degenerative complications are high in patients with either type I or type II diabetes mellitus (Adeghate, 1999 a).

The available methods for treatment of diabetes mellitus include diet or diet and oral hypoglycemic drugs or diet and insulin (Boon et al., 1999). In traditional practices, medicinal plants are used to control diabetes mellitus in many countries. This caused an increase in the number of experimental and clinical investigations directed toward the validation of the hypoglycemic (El–Ridi, 2001) and / or anti – diabetic (Alarcon–Aguilara et al., 1998) properties of different medicinal plants. It has been reported that the chard (*Beta Vulgaris* L.var. cicla) extract increased pancreatic beta cells in Streptozotocin (STZ) – diabetic rats (Bolkent et al., 2000). Water extract of *S总监anthus Sonchifolius* (yacon) leaves fed to normal and STZ diabetic rats also showed hypoglycemic effects (Aybar et al., 2001). Moreover, a long – term administration of olive leaves or *Cinnamon bark* caused significant improvement in tissue injury induced by STZ treatment (Onderoglu et al., 1999). The water extract of *Anastatica hierochuntica*, in the dose given and rout of
administration used by Tarek and Mamdouh (2002) has hypoglycemic effect in both normoglycemic and diabetic rats.

The objectives of the present study are to evaluate the hypoglycemic effect of the methanolic extracts of J. phoenicea, H. thebaica, A. hierochuntica and Cleome droserifolia in normal and Alloxan – diabetic pregnant female rats and their embryos.

Materials and Methods

2.1. Animals
Adult diabetic and normal albino rats (72 rats) weighing (120 – 150 g) were maintained in a well ventilated animal house. Animals were housed in large spacious polypolyethylene cages with free access to food and ad libitum during the course of experiment (Lane – patter and Pearson, 1971). Males and diabetic females were orally administered with the different treatments once daily, for one week, then two females were housed with one male overnight, and successful mating was confirmed by vaginal plug. The presence of vaginal plug was considered as the zero gestational day. The pregnant female's administration with the different treatments continuous during pregnancy till the 19th day of gestation.

2.2. Chemicals:
Alloxan: recrystallized, used to produce insulin – dependent diabetes experimentally.

2.3. Plant materials:
The methanol extracts of the four tested plants: C. droserfolia (sommo), J. Phoenicia (araar), A. hierochuntica (kafta) and H. thebaica (doum) were used in the present studies as antidiabetic agents.

2.4. Experiments
Pregnant females albino rats were divided into six groups (36 rats) Group 1 control group (normal pregnant females) was orally administered with distilled water. Group 2 animals were injected with alloxan 60 mg/kg (Lazaro, 1949) for only one injection and orally administered with distilled water (pregnant diabetic control). Group 3 pregnant diabetic rats were administered orally with 150 mg/kg/day with methanolic extract of sommo. Group 4 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of kafta. Group 5 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of araar and Group 6 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of doum. Another six groups (36 rats) of nonpregnant females albino rats were used as controls for previous six groups, respectively for blood glucose values comparison. Females were sacrificed at day 19th of gestation and the chromosomes prepared from bone marrow cells of mothers according to Yosida et al. (1977). In case of embryo, cells were prepared according to the method of Romagnano et al. (1985) to study any genotoxic effect. For biochemical studies blood and livers of the females and their embryos were collected. Livers chilled up for analysis and blood plasma was prepared by centrifugation at 3000 rpm and chilled up for analysis (Astrwov, 1974).

2.5. Slide preparation:
To study the chromosomal aberrations in bone marrow cells of females (mothers), 50 metaphases from each animal were examined. In the case of embryo cells, chromosomal aberrations of liver in 900 cells per each group were examined.

Determination of blood glucose was adopted using the method of Trinder (1969). Liver tissues were homogenized by the method of Peares (1985) for determination of RNA and DNA. The DNA concentrations were determined in the supernatant according to Dische method (1955) using diphenylamine reaction which produced blue color when heated with DNA. The RNA concentrations were determined using orcinol method according to Schneider method (1957). Total soluble protein in liver was determined according to Lowary et al. (1951) method. The activity of liver nucleases was determined according to Bergmeger (1974).

2.6. Data analysis
Intergroup differences of cytogenetic and biochemical studies were analyzed statistically by using standard T-test according to Sokall and Rohlf (1995).

Results

3.1. Cytogenic results:
The present result in Table (1) showed the frequencies of structural and numerical chromosomal aberrations as well as the mitotic activity induced by alloxan and also the effect of administration of the methanol extracts of the plants studied in female bone marrow cells. Injection with alloxan increased the frequencies of individual and total chromosomal aberrations in female bone marrow cells when compared with the control. This increase was found to be statistically significant at (P<0.001).

Administration of the plants methanol extracts under test after injection with alloxan indicated that these
plants methanol extracts significantly decreased the frequencies of chromosomal aberrations induced by alloxan ($P<0.001$, $P<0.01$), but still higher than that of the normal pregnant control significantly.

The methanol extract of *H. thebaica* (Doum) ingestion showed the lowest percentage of total structural aberrations in female bone marrow cells when compared with those of Sammo, Araar and Kafra methanol extracts ingestion. Numerical aberrations also showed a significant decrease in animals injected with alloxan (diabetes) after treatment with methanolic extracts of the medicinal plants, but also still higher than the control. The mitotic activity increased after treatment with the extracts of the four plants to approximately reach the control group.

Table (2) showed that the results of structural and numerical chromosomal abnormalities in embryo cells of treated mothers in all experimental groups as well as the mitotic activity. The observed types of abnormalities were breaks, deletions, centromeric attenuations, endomitosis and gaps. Diabetic animals (alloxanized) showed a significant increase (at level of $P<0.01$) for total structural aberrations. The treatment with the methanolic extracts of the four tested plants inhibited the percentage of total aberrations significantly (at level of $P<0.01$) although this percentage of abnormalities in treated groups remain significantly increased than the normal control group. Numerical aberrations were recorded and the results showed slight deviation from the normal group. The mitotic activity showed highly significant decrease ($P<0.001$) after injection with alloxan (diabetes). Treatment with the plants extract increased the number of divided cells significantly to be near the normal group. The statistical analysis showed no significant difference between animals treated with different methanolic extracts of the medicinal plants.

### 3.2. Biochemical results:

The influences of Araar, Doum, Kafta and Sammo as hypoglycemic or antidiabetic agents were studied using their methanol extracts against alloxanized diabetes of female pregnant albino rats. The results in Table (3) summarized the blood glucose levels of the present experimental female normal and pregnant albino rats. The average level of blood glucose of the alloxanized normal female diabetic rats raised from 80.80 ± 2.25 mg/dl for normal female (healthy without pregnancy) to 379.40 ± 14.72 mg/dl for normal female diabetic rats, but for female pregnant rats, the values were increased from 88.00 ± 0.79 to 421.60 ± 8.88, respectively. These elevated blood glucose levels of diabetic normal female and pregnant female albino rats were greatly decreased to around that of normal control of normal and pregnant female animals which were ranged from 106 to 107% and from 102 to 105%, respectively to their controls by ingestion of the methanol extracts of the four tested medicinal plants. The variation between the effects of four different plants was insignificant.

Table (4) also showed that diabetes decreased liver total soluble protein of both pregnant females and their embryos. The decreased values were 71% and 80%, respectively relative to the normal control of pregnant (167.7 ± 3.16 mg/g) and embryo (128.1 ± 2.12 mg/g) liver. But ingestion of the studied medicinal plants methanol extracts improved the lower values of diabetic animals and normalized them to around the normal levels which ranged between 102% and 105% for pregnant liver and from 101% to 102% for embryo relative to their normal control.

Concerning to nucleic acids metabolism, the contents of DNA and RNA (as shown in Table 5) in pregnant and embryo liver tissues of diabetic rats were decreased significantly relative to their normal control. The nucleic acid values of diabetic pregnant liver amounted 47% and 72% relative to normal pregnant control, respectively for DNA and RNA contents (the normal values of DNA was 0.403 ± 0.017 mg/g and for RNA was 0.190 ± 0.007 mg/g tissue). These abnormal levels of nucleic acids produced by diabetes were improved by the administration of methanolic extracts of Kafta, Sammo, Araar and Doum which contained flavonoid compounds. These four extracts ingestion into diabetic animals readjusted DNA and RNA content in range of 103% - 106% and 103% -107%, respectively relative to those of normal pregnant control. In case of embryo, the contents of liver DNA and RNA were 0.210 ± 0.004 and 0.157 ± 0.007 mg/g tissue of normal control. Diabetes reduced these values to 70% and 79% respectively relative to normal control. The tested medicinal methanol extracts treatments improved these effects of diabetes and normalized the level values of DNA and RNA of 123% - 126% and 101% - 105% relative to those of normal control, respectively. It means that methanol extracts (or flavonoids extract) of the four studied medicinal plants normalized and treated the harmful effects of diabetes on nucleic acid contents of diabetic pregnant female rats and their embryos.

The data present in Table (6) showed that the activity of nuclease (DNAase and RNAase) of pregnant liver tissue and their embryo liver of diabetic animals which were inhibited significantly steadily when compared with those of normal pregnant healthy female control. The activity was inhibited to values ranged from 52% to 56% relative to those of normal control. It means
that about half of the liver nucleases activity was lost by diabetes. The effects of diabetes oxidative stress on nucleases activity was improved and readjusted to around those of normal control by treatments of the methanol extracts of the present tested medicinal plants, in which the normalized values were 101 – 103% and 103 -105%, respectively for diabetic pregnant females and their embryo DNAase activity and 102 -105% and 103%, respectively for diabetes pregnant female and their embryo RNAase activity relative to those of normal pregnant healthy females and their embryos. The normal healthy pregnant control had values of 60.10 ± 1.03 IU/g tissue and 58.32 ± 0.01 IU/g tissues for DNAase activity and 36.24 ± 0.65 IU/g tissue and 30.11 ± 0.74 IU/g tissue for RNAase activity, respectively in pregnant females liver and their embryos.

The present results suggested that the treatment of diabetic pregnant females with the methanolic extracts of Araar, Doum, Kafta and Sammo which contain flavonoids compounds inhibited the induced chromosomal aberrations in the bone marrow cells and embryo cells and improved and readjusted the harmful and disturbed effects of diabetes on blood glucose and liver total soluble protein, RNA and DNA contents as well as liver nucleases (DNAase and RNAase) activity.

**Discussion**

The increase in number of diabetic patients have motivated scientists to find new method to cure diabetes (Adeghate, 1999 b). In the present study, the effect of the metabolic extracts of *J. phoencea* (Araar), *H.thebaica* (Doum), *A. hierochuntica* (kafta) and *C. droserifolia* (Sammo) were tested on normoglycemic as well as alloxan diabetic female pregnant rats and their embryos. The diabetogenic effect of alloxan and STZ was reported by several investigators (Choi et al. 1991; Adeghate, 1999 b; Wang et al., 2000; Mohamed Bnouham et al., 2002). It was indicated in the present study by the significant increase in blood glucose level in alloxan injected rats. It was also indicated by the highly significant increase in somatic chromosomal aberrations of pregnant female and their embryos. The effect of STZ and alloxan relate to their structure as a DNA alkylating agent, which leads to necrosis of pancreatic beta cells and thus to a state of insulin - dependent diabetes mellitus (Elsner et al., 2000). As alloxanized diabetes may either increase the enteanance rate of glucose into blood from liver (stimulated hepatic glycogenolysis or gluconeogenesis) and may be reduced the rate of removal of glucose from blood by different tissues which decreased storage or utilization of glucose. These may be due to the absent of insulin. Alloxan acted directly promptly and specifically on the β-cells of pancreas (Chatterjee and Shinde, 2002).

On the other hand, the results indicated that administration of the methanol extracts of the present studied plants improved, but not completely normalized, the diabetogenic action induced by alloxan. They showed hypoglycemic effects on blood glucose level in plant – treated diabetic females and embryos when compared with untreated diabetics. They also caused a significant decrease in the percentage of chromosomal aberrations caused by alloxan in both mothers and embryos. The four methanolic extracts had a great amount of flavonoid compounds which did not alter the low level of insulin (Ismail et al., 2007). This means that the reducing effect of methanol extracts (flavonoid compounds) on blood glucose of diabetic animals was not related to the insulin content in blood. Flavonoids ingestion into diabetic animals increased blood adiponectin level, which produced a hypoglycemic action of insulin sensitivity (Comds et al., 2002; Yamauchi et al., 2003; Ismail et al., 2007). Similarly, it is believed that the presence of flavone glycosidic components in the plant extract of *Anastatia hierochuntia* (Khalifa, 1980), is responsible for reducing the blood glucose level in STZ – diabetic rats. Such hypoglycemic effect could be through increased serum insulin levels provided by repair/ regeneration of the endocrine pancreas.

Results of liver total soluble protein nucleic acids and nucleases showed that diabetes caused a great harmful and disturbance in liver metabolism including either in liver metabolites content or in enzymes activity, but the medicinal plants methanol extracts (included falvonoids) improved and normalized these harmful and disturbed effects of diabetes on livers metabolism as well as their metabolites. The present results are in agreement with those of Salah et al. (2010). They found that flavonoids of the metabolic extracts of Kafta, Araar, Somma and Doum improved and treated diabetes in case of blood glucose, and liver contents of total soluble protein, RNA and DNA as well as the activity of enzymes related to nucleic acid such as nucleases (RNAase and DNAase). They also found that it inhibited the percentage of chromosomal aberrations in male somatic and germ cells. It means that, the effects of diabetes oxidative stress on liver protein and nucleic acids as well as their nucleases activity was readjusted and improved by ingestion of alcoholic extracts of the four present medicinal plants.
On the other hand, there is no any available data was observed in literature about these items in pregnant animals and their embryo, except those of our work on male albino rats. The present findings of DNA content in liver of pregnant females and their embryos are confirmed by the results of chromosomal aberrations in which DNA content was deceased and chromosomal aberrations were increased by diabetes. Also, RNA content data confirmed the results of protein content which is in agreement with the findings of nuclease activity. This means that the present results confirmed each other. The results showed that plants methanol extracts (including flavonoids) might cause an adaptation of certain enzymes system in the animal tissues of diabetic animals which provide the necessary enzymatic machinery to cope with the increased flow of nucleotides during the experimental period by converting it rapidly into DNA and RNA (m-RNA, r-RNA and t-RNA). However, flavonoids may play a role in the improvement of replication and transcription enzymes activity to produce DNA and RNA. In addition, the efficiency of flavonoids against the oxidative stress of diabetes is not completely known (Coskun et al., 2005 and Ismail et al., 2007). Flavonoids were in conjugation with adiponectin may play a role in binding of insulin with its receptor. Moreover, these compounds may interfere with signal sequences of insulin inside the cell (Anton et al., 2007 and Abdel-Magid, 2007). The induction of flavonoids extracts elevated the adiponectin levels in the diabetic rats, which showed glucose lowering effects and improvement in insulin resistance and sensitivity and diabetic oxidative stress as well as the whole body metabolism (Abdel-Magid, 2007).

From the present results, it can be concluded that there are some biochemical dynamics which might occur in the metabolism of glucose, nucleic acids and proteins in order to prevent or to reduce the oxidative stress of diabetes by flavonoids treatment. The decrease of blood glucose with increase soluble protein and nucleic acids content as well as stimulation of nuclease activity of liver tissues in diabetic female pregnant rats and their embryos by flavonoids induction subsequently is considered as results of diabetes oxidative stress which was treated by flavonoids compounds. Also, the results suggested that, the stimulation of protein biosynthesis processes was marked by increase the RNA content as well as stimulation of nuclease activity. The enzymes may supply transcription stage of protein biosynthesis with the needed precursors nucleotides (Adams et al., 1993).

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Chromosomal aberrations in the bone marrow of female rats treated with Sammo, Araar, Kafta and Doum after injection with alloxan.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Number of examined cells</th>
<th>Chromosomal aberrations</th>
<th>Numerical aberrations</th>
<th>Mitotic Index (MI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>structural gap</td>
<td>chromatid break</td>
<td>fragment deletion</td>
</tr>
<tr>
<td>Control</td>
<td>250</td>
<td>1.20%</td>
<td>0.00%</td>
<td>0.00%</td>
</tr>
<tr>
<td>Alloxan</td>
<td>250</td>
<td>6.80%</td>
<td>3.00%</td>
<td>2.00%</td>
</tr>
<tr>
<td>Sammo</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alloxan</td>
<td>250</td>
<td>3.6%</td>
<td>0.8%</td>
<td>0.4%</td>
</tr>
<tr>
<td>Araar</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alloxan</td>
<td>250</td>
<td>5.2%</td>
<td>1.2%</td>
<td>0.4%</td>
</tr>
<tr>
<td>Kafta</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alloxan</td>
<td>250</td>
<td>4.4%</td>
<td>0.8%</td>
<td>0.8%</td>
</tr>
<tr>
<td>Doum</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alloxan</td>
<td>250</td>
<td>2.8%</td>
<td>2.0%</td>
<td>0.8%</td>
</tr>
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</table>

* Significant at P<0.05, ** at P<0.01 and *** P<0.001
Chromosomal aberrations in the embryos of female rats treated with Sammo Araar, Kafta and Doum after injection with alloxan.

<table>
<thead>
<tr>
<th>Groups</th>
<th>No. of embryos</th>
<th>No. of cells</th>
<th>Structural chromosomal aberration</th>
<th>Total chromosomal aberration excluding gap</th>
<th>Numerical index</th>
<th>Mito tic index</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>break</td>
<td>C.A</td>
<td>del.</td>
<td>6.56%</td>
<td>1.1%</td>
</tr>
<tr>
<td>Control</td>
<td>15</td>
<td>900</td>
<td>0.44%</td>
<td>1%</td>
<td>0.33%</td>
<td>0.56%</td>
</tr>
<tr>
<td>Alloxan</td>
<td>15</td>
<td>900</td>
<td>2.11%</td>
<td>4%</td>
<td>1.89%</td>
<td>2.56%</td>
</tr>
<tr>
<td>Sammo alloxan</td>
<td>15</td>
<td>900</td>
<td>0.78%</td>
<td>1.4%</td>
<td>1.1%</td>
<td>6.89%</td>
</tr>
<tr>
<td>Araar alloxan</td>
<td>15</td>
<td>900</td>
<td>0.67%</td>
<td>1.2%</td>
<td>0.09%</td>
<td>0.78%</td>
</tr>
<tr>
<td>Kafta alloxan</td>
<td>15</td>
<td>900</td>
<td>0.56%</td>
<td>0.56%</td>
<td>0.78%</td>
<td>1%</td>
</tr>
<tr>
<td>Doum alloxan</td>
<td>15</td>
<td>900</td>
<td>0.56%</td>
<td>1.3%</td>
<td>0.56%</td>
<td>0.78%</td>
</tr>
</tbody>
</table>

Significant at $P<0.05$, ** at $P<0.01$ and *** $P<0.001$
Table 3

Effect of the experimental medicinal plants as antidiabetic agents on blood glucose of diabetic female albino rats.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Normal female</th>
<th></th>
<th>Pregnant female</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mg/100 ml</td>
<td>%</td>
<td>mg/100 ml</td>
<td>%</td>
</tr>
<tr>
<td>Normal control</td>
<td>80.80 ± 2.25</td>
<td>100</td>
<td>88.00 ± 0.79</td>
<td>100</td>
</tr>
<tr>
<td>Diabetic control</td>
<td>379.40 ± 14.72</td>
<td>470</td>
<td>421.60 ± 8.88</td>
<td>479</td>
</tr>
<tr>
<td>Araar</td>
<td>86.0 ± 2.15</td>
<td>106</td>
<td>90.00 ± 2.89</td>
<td>102</td>
</tr>
<tr>
<td>Sammo</td>
<td>85.40 ± 2.41</td>
<td>106</td>
<td>89.40 ± 1.79</td>
<td>102</td>
</tr>
<tr>
<td>Kafta</td>
<td>85.80 ± 2.63</td>
<td>106</td>
<td>92.00 ± 4.96</td>
<td>105</td>
</tr>
<tr>
<td>Doum</td>
<td>86.40 ± 1.79</td>
<td>107</td>
<td>91.20 ± 2.41</td>
<td>104</td>
</tr>
</tbody>
</table>
Effect of the experimental medicinal plants as antidiabetic agents on liver soluble protein of diabetic female albino rats and their embryos.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Pregnant liver</th>
<th></th>
<th>Embryo liver</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mg/g tissue</td>
<td>%</td>
<td>Mg/g tissue</td>
<td>%</td>
</tr>
<tr>
<td>Normal control</td>
<td>167.70 ± 3.16</td>
<td>100</td>
<td>128.10 ± 2.12</td>
<td>100</td>
</tr>
<tr>
<td>Diabetic control</td>
<td>118.60 ± 2.99</td>
<td>71</td>
<td>102.10 ± 2.00</td>
<td>80</td>
</tr>
<tr>
<td>Araar</td>
<td>170.2 ± 2.87</td>
<td>103</td>
<td>129.10 ± 1.94</td>
<td>101</td>
</tr>
<tr>
<td>Sammo</td>
<td>171.11 ± 3.21</td>
<td>102</td>
<td>128.41 ± 2.11</td>
<td>102</td>
</tr>
<tr>
<td>Kafta</td>
<td>175.50 ± 3.42</td>
<td>105</td>
<td>130.20 ± 1.95</td>
<td>102</td>
</tr>
<tr>
<td>Doum</td>
<td>176.60 ± 3.33</td>
<td>105</td>
<td>130.7 ± 1.96</td>
<td>102</td>
</tr>
</tbody>
</table>
Table 5

Effect of the experimental medicinal plants as antidiabetic agents on liver DNA and RNA content of diabetic female albino rats and their embryos.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Pregnant liver</th>
<th>Embryo liver</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mg/g tissue</td>
<td>%</td>
</tr>
<tr>
<td>Normal control</td>
<td>0.403 ± 0.017</td>
<td>100</td>
</tr>
<tr>
<td>Diabetic control</td>
<td>0.190 ± 0.014</td>
<td>47</td>
</tr>
<tr>
<td>Araar</td>
<td>0.432 ± 0.004</td>
<td>107</td>
</tr>
<tr>
<td>Sammo</td>
<td>0.430 ± 0.004</td>
<td>107</td>
</tr>
<tr>
<td>Kafta</td>
<td>0.434 ± 0.004</td>
<td>108</td>
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<tr>
<td>Doum</td>
<td>0.429 ± 0.006</td>
<td>106</td>
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</table>

<table>
<thead>
<tr>
<th>RNA</th>
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<tbody>
<tr>
<td>Normal control</td>
<td>0.190 ± 0.007</td>
<td>100</td>
<td>0.157 ± 0.007</td>
<td>100</td>
</tr>
<tr>
<td>Diabetic control</td>
<td>0.137 ± 0.004</td>
<td>72</td>
<td>0.124 ± 0.004</td>
<td>79</td>
</tr>
<tr>
<td>Araar</td>
<td>0.199 ± 0.008</td>
<td>105</td>
<td>0.158 ± 0.002</td>
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<tr>
<td>Sammo</td>
<td>0.195 ± 0.007</td>
<td>103</td>
<td>0.159 ± 0.005</td>
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<tr>
<td>Kafta</td>
<td>0.202 ± 0.005</td>
<td>106</td>
<td>0.161 ± 0.005</td>
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<tr>
<td>Doum</td>
<td>0.204 ± 0.004</td>
<td>107</td>
<td>0.162 ± 0.003</td>
<td>103</td>
</tr>
</tbody>
</table>
Illustration 6

Table 6

Effect of the experimental medicinal plants as antidiabetic agents on liver nuclease activity of diabetic female albino rats and their embryos.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Pregnant liver</th>
<th>Embryo Liver</th>
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<tbody>
<tr>
<td></td>
<td>mg/g tissue</td>
<td>%</td>
</tr>
<tr>
<td>DNAase</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal control</td>
<td>60.10 ± 1.03</td>
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</tr>
<tr>
<td>Diabetic control</td>
<td>32.47 ± 1.11</td>
<td>54</td>
</tr>
<tr>
<td>Araar</td>
<td>61.12 ± 2.16</td>
<td>102</td>
</tr>
<tr>
<td>Sammo</td>
<td>62.12 ± 1.97</td>
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<tr>
<td>Kafta</td>
<td>62.00 ± 2.42</td>
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<tr>
<td>Doum</td>
<td>60.97 ± 1.86</td>
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<td>Normal control</td>
<td>36.24 ± 0.65</td>
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<td>Diabetic control</td>
<td>20.22 ± 0.50</td>
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<tr>
<td>Araar</td>
<td>37.01 ± 0.54</td>
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<td>Sammo</td>
<td>37.98 ± 0.92</td>
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</tr>
<tr>
<td>Kafta</td>
<td>36.94 ± 0.71</td>
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</tr>
<tr>
<td>Doum</td>
<td>37.00 ± 0.83</td>
<td>102</td>
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</table>
Reviews

Review 1

Review Title: Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats

Posted by Karim Hmadcha on 24 Mar 2011 10:05:40 AM GMT

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<td>11</td>
<td>Are the keywords and abstract or summary informative?</td>
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</table>

Rating: 4

Comment:

Dear Editor

In the manuscript titled: “Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats”, the authors provide evidence and discussed the importance of the use of methanol extract of the medicinal plants Juniperus Phoenicea (Araar), Hyphaene thebaica (Doum), Anastatica hierochuntica (Kafta) and Cleome droserifolia (Sammo), to reduce blood glucose level in alloxan induced female pregnant diabetic albino rats and to prevent chromosomal aberrations. The topic is interesting and aims to the therapeutic potential of medicinal plants to prevent diabetes; however, the present manuscript definitely needs to be substantially revised

Comments to the authors:

1. Language: In addition to spell check using a common word text processor, or similar, correction of English grammar and syntax is required.
2. Also, numerous errors of capitalization of words are present throughout the article.
3. Lack of conclusion in the abstract section.
4. The timing need to be included (e.g. diabetes/glucose recovery in hours, days???)
5. Discuss or state:
   - Chromosome aberrations in other organs.
   - The effect of medicinal plants on normal animals.
   - The effect of medicinal plants on male rats.
   - DNA and RNA content in other organs.

Competing interests: No
Invited by the author to make a review on this article? : No

Experience and credentials in the specific area of science:
Experience in stem cell biology, diabetes and its complication, molecular biology and epigenetics.

Publications in the same or a related area of science: No

How to cite: Hmadcha K. Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats [Review of the article 'Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats' by ], WebmedCentral 1970;2(3):REVIEW_REF_NUM623
Review 2

Review Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

Posted by Dr. Adam Baldinger on 19 Mar 2011 01:58:29 PM GMT

1. Is the subject of the article within the scope of the subject category? Yes
2. Are the interpretations / conclusions sound and justified by the data? Yes
3. Is this a new and original contribution? Yes
4. Does this paper exemplify an awareness of other research on the topic? No
5. Are structure and length satisfactory? Yes
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9. Are the illustrations and tables necessary and acceptable? Yes
10. Are the references adequate and are they all necessary? Yes
11. Are the keywords and abstract or summary informative? Yes

Rating: 8

Comment:

Article ID: WMC001758

Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

The title of the paper sufficiently and clearly the contents

The abstract contains a sufficient summary of the paper’ conten notation, nomenclature and method used conform to standards

Line 4,11,17 change Methanolic to Methanol

MATERIALS AND METHODS

add

2.1. Animals , 2.2. Chemicals, 2.3. Plant materials, 2.4, Experiments, 2.5. Slide preparation, 2.6. Data analysis

Result

Change Table (2) presented the results of ..... To Table (2) showed that the results of

Change Table (6) in connection showed that to

The data present in Table (6) showed that

Result and Discussion change Methanolic to Methanol

acceptable in the area

* the organization of the paper is satisfactory
* italic writing often does not follow rules
* there are some Grammar problem

Recommended for acceptance conditionally; the author must make revisions and modifications as indicated in this section
Prof. Dr. Adam A. Baldinger  
Museum of comparative zoology, Harvard university, Cambridge, USA  
baldingeradan@yahoo.com

Invited by the author to make a review on this article? : No 
Experience and credentials in the specific area of science:  
there is my experience in cell biology and physiology 

Publications in the same or a related area of science: No 

How to cite: Baldinger A. Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats [Review of the article ‘Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats ’ by J]. WebmedCentral 1970;2(3):REVIEW_REF_NUM603
Review 3

Review Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

Posted by Dr. Fatma Said on 19 Mar 2011 12:52:46 PM GMT

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<tr>
<td>11</td>
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</table>

Rating: 7

Comment:
My Dear. Prof. Dr EDITOR

Manuscript Number WMC001758

Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

General comment: This paper may be interest to the readers of the journal.

Title: sufficiently and clearly

The abstract: sufficient summary of the paper

- Key words: ok

- Introduction: Enough

Result and Discussion are accurate and supported by the content

- References: Follow the style of the journal

- All figures and tables: Sufficient

Introduction
In the present study, the hypoglycemic effect of the methanolic extracts of Juniperus phoencea (Araar), Hyphaene thebaica (Doum), Anastatica hierochuntica (kalla) and Cleome droserifolia (Sammo) were tested in normal and Alloxan – diabetic pregnant female rats and their embryos

Change to

The objectives of the present study are to evaluate the methanolic extracts of J.phoencea (Araar), H.thebaica (Doum), A.hierochuntica (kalla) and C.droserifolia (Sammo) were tested in normal and Alloxan – diabetic pregnant female rats and their embryos

- MATERIALS AND METHODS
2.4. Experiments

Pregnant females albino rats were divided into six groups (36 rats) Group I control group (normal pregnant females) was orally administered with distilled water, Group 2 animals were injected with alloxan 60 mg/kg (Lazaro, 1949) for only one injection and orally administered with distilled water (pregnant diabetic control), Group 3 pregnant diabetic rats were administered orally with 150 mg/kg/day with methanolic extract of sammo, Group 4 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of kafta, group 5 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of araar and Group 6 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of doum. Another six groups (36 rats) of nonpregnant females albino rats were used as controls for previous six groups, respectively for blood glucose values comparison.

Result.

Cytogenetic results: Table (1) showed

Recommended for acceptance and the author must make minor revisions

Prof. Dr. Fatma A said
Professor of parasitology.
Zoology Department, Faculty of Science, Cairo university, Egypt.

Invited by the author to make a review on this article? : No

Experience and credentials in the specific area of science:

MATERIALS AND METHODS

Pregnant females albino rats were divided into six groups (36 rats) Group I control group (normal pregnant females) was orally administered with distilled water, Group 2 animals were injected with alloxan 60 mg/kg (Lazaro, 1949) for only one injection and orally administered with distilled water (pregnant diabetic control), Group 3 pregnant diabetic rats were administered orally with 150 mg/kg/day with methanolic extract of sammo, Group 4 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of kafta, group 5 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of araar and Group 6 pregnant diabetic rats administered orally with 150 mg/kg/day with methanolic extract of doum. Another six groups (36 rats) of nonpregnant females albino rats were used as controls for previous six groups, respectively for blood glucose values comparison.

Result.
Cytogenetic results: Table (1) showed To

3.1. Cytogenetic results: The present result in Table (1) showed

Publications in the same or a related area of science: No

How to cite: Said F. Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats [Review of the article ‘Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats’ by ]. WebmedCentral 1970;2(3):REVIEW_REF_NUM602
Review 4

Review Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

Posted by Prof. Fayez Bakry on 19 Mar 2011 12:27:15 PM GMT

1. Is the subject of the article within the scope of the subject category? Yes
2. Are the interpretations / conclusions sound and justified by the data? Yes
3. Is this a new and original contribution? Yes
4. Does this paper exemplify an awareness of other research on the topic? No
5. Are structure and length satisfactory? Yes
6. Can you suggest brief additions or amendments or an introductory statement that will increase the value of this paper for an international audience? No
7. Can you suggest any reductions in the paper, or deletions of parts? No
8. Is the quality of the diction satisfactory? Yes
9. Are the illustrations and tables necessary and acceptable? Yes
10. Are the references adequate and are they all necessary? Yes
11. Are the keywords and abstract or summary informative? Yes

Rating: 8

Comment:
I write you in regards to Manuscript Number WMC001758

Title: Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats

§ there are some Grammer problem
§ Abstract: Lacked conclusion
§ Introduction is ok
§ table and figure Sufficient
§ Results and Discussion: Sufficient
§ Conclusion: ok
§ References: Follow the style of the journal

§ Recommended for acceptance , the author must make revisions and modifications as indicated in this section

§ summarize all the mistakes in the manuscript

Abstract
Line 19. Add It is concluded that

Materials and Methods
Change The pregnant female administration to The pregnant female's administration
Change Methanolic to methanol in all text
Change The methanolic extracts of the four tested plants: Cleome droserfolia (sommo), Juniperus Phoenici(araar), Anostatica hierochuntica (kafta) and Hyphened thebaica (doum) were used in the present studies as antidiabetic agents. To
The methanol extracts of the four tested plants: *C. droserfolia* (sommo), *J. Phoenicia* (araar), *A. hierochuntica* (kafta) and *H. thebaica* (doum) were used in the present studies as antidiabetic agents.

**Prof. Dr. Fayez Ahmed Bakry**
Environmental Researches and Medical Malacology Department
Theodor Bilharz Research Institute (TBRI). Egypt.

My dear

Invited by the author to make a review on this article? : No

Experience and credentials in the specific area of science:
Recommended for acceptance, the author must make revisions and modifications as indicated in this section

Publications in the same or a related area of science: No

**How to cite:** Bakry F. Chromosomal aberrations and nucleic acids systems affected by some Egyptian medicinal plants used in treating female pregnant diabetic rats[Review of the article ’Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats ’ by ].WebmedCentral 1970;2(3):REVIEW_REF_NUM600
Review 5

Review Title: The prospect of alloxan in providing an animal model that is suitable for the screening of anti mutagenic agents: comment and review

Posted by Jeanne Adiwinata Pawitan on 18 Mar 2011 04:51:59 AM GMT

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Rating: 4

Comment:
The article by Salah SH and Abdou HS showed that methanol extract of four medicinal plants i.e. Juniperus Phoenicea (Araar), Hyphaene thebaica (Doum), Anastatica hierochuntica (Kafta) and Cleome droserifolia (Sammo) could reduce blood glucose level in alloxan induced diabetic rats. In addition, the methanol extracts decreased alloxan induced chromosomal aberration, though the reduction did not attain the normal level. Therefore, the four medicinal plants showed promising potentials to treat diabetes [1], and worth further testing.

Alloxan induced chromosomal aberrations were structural as well as numerical aberration. However, Table -1 that shows chromosomal aberrations in the bone marrow of female rats was not well arranged, as structural chromosomal aberration only consists of chromatid gap, while numerical aberrations include chromosomal gap, break, fragment, endomitosis and hypoploidy [1].

Alloxan structure is similar to streptozotosin that act as a DNA alkylating agent, which may cause mutagenesis that may be represented as chromosomal aberration in bone marrow and embryo of female pregnant rats.1 In pancreatic beta cells, chromosomal aberration may lead to necrosis, and thus insulin-dependent diabetes mellitus [1, 2]. Moreover, studies showed that alloxan caused uneuploidy in mice [3], and Neurospora crassa [4]. Therefore, in addition to develop diabetic animal models, alloxan may be used to develop mutations in animal models that is suitable for the screening of antimutagenic agents.

References


Competing interests: none declared

Invited by the author to make a review on this article? : No

Experience and credentials in the specific area of science:
Doing research in cytology, especially developing a simple method to detect chromosomal breakage

Publications in the same or a related area of science: Yes


How to cite: Adiwinata Pawitan J. The prospect of alloxan in providing an animal model that is suitable for the screening of anti mutagenic agents: comment and review[Review of the article 'Chromosomal Aberrations and Nucleic Acids Systems Affected by Some Egyptian Medicinal Plants used in Treating Female Pregnant Diabetic Rats ' by ].WebmedCentral 1970;2(3):REVIEW_REF_NUM595
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