

**Effect of Aloe vera extract on ovaries development in during gestational age of 18 day embryo of diabetic rats**Mehrzaad Jafari Barmak<sup>1</sup>, Zabihollah Khaksar<sup>2</sup>, Asghar Sharifi<sup>3</sup>, Reza Mahmoudi<sup>4\*</sup>

<sup>1</sup> Assistant professor of Histology, Cellular and Molecular research center, Yasuj University of Medical Sciences, Yasuj, Iran.

<sup>2</sup> Associate professor of Comparative Anatomy and Embryology, Department of Anatomy, Faculty of Veterinary, Shiraz University, Shiraz, Iran.

<sup>3</sup> Assistant professor of Bacteriology, Cellular and Molecular research center, Yasuj University of Medical Sciences, Yasuj, Iran.

<sup>4\*</sup> Associate professor of Anatomy, Cellular and Molecular research center, Yasuj University of Medical Sciences, Yasuj, Iran.

\*Corresponding: [rmahmoudi40@yahoo.com](mailto:rmahmoudi40@yahoo.com)

**Abstract:** Ovary is a part of the reproductive system can be affected by diabetes. Aloe vera is a plant that reduces hypoglycemic. Thus, in present study, its aloe vera effect on ovaries development in during gestational age of 18 day embryo of rat. **Material and methods:** Two Groups of female Sprag dawly rats were injected by streptozotocin (50 mg/kg/IP) and mating by male rat, vaginal plaque is objected. One groups of rats received aloe vera extract (400mg/kg) by oral in gestational age during and other group giving distilled water. After 18 day, rats sacrificed and their embryo were removed. Abdominal sections of embryo were processed and embedding by paraffin. The 5-6  $\mu$  sections were made and staining by hematoxyline- eosin and mason trichorome. Cellular of ovary change is evaluated. **Results:** Mean body weight do not show significantly ( $p>0.05$ ). Mean blood sugar were decreased significantly ( $p<0.05$ ). The mean of primordial germ cells were seen significant ( $p<0.05$ ). **Discussion:** This study showed that Aloe Vera can reduce the blood sugar, also cause be increased primordial germ cells in embryos of diabetic mothers. [Mehrzaad Jafari Barmak, Zabihollah Khaksar, Asghar Sharifi, Reza Mahmoudi. **Effect of Aloe vera extract on ovaries development in during gestational age of 18 day embryo of diabetic rats.** *Life Sci J* 2013;10(2s):323-327] (ISSN:1097-8135). <http://www.lifesciencesite.com>. 55

**Keywords:** Aloe vera, Ovaries development, Diabetic

**Introduction:**

Diabetes is a complex and multifarious group of disorders characterized by hyperglycemia (1). Diabetes is associated with increased risk of disease such as neuropathy and cardiovascular disorders, but it is also linked to reproductive problems such as spontaneous abortion, neonatal morbidity and mortality, congenital malformation and poor embryo development (2, 3). Diabetes causes alterations in the estrous cycle, associated with modification in ovarian function in spermatogenesis and oogenesis phases, which induces a decrease of primordial germ cells (3, 4). Some chemical drugs such as biguanides and sulfonylureas are currently available to reduce hyperglycemia in diabetes mellitus (7). These drugs have side effect and thus search for new drug or new compound is essential (1, 8). Many herbs and plant products have been shown to have hypoglycemia action. This leads to increasing demand for herbal products plant with antidiabetic activity and lower side effects (9, 12). Aloe vera is one of these antidiabetic plants (13). This plant has stiff gray-green lance shaped leaves containing clear gel in a central mucilaginous pulp. Clinical evaluations have revealed that the pharmacologically active ingredients are concentrated in both the gel

and rind of Aloe vera leaves (14). Polysaccharide containing plants which Aloe barbadensis is also among are used in various diseases as anti-inflammatory, antiulcer, antineoplastic and in wound healing and against hepatitis(15). It is known that it is activating macrophages and has also antiviral effect (16). In some studies it is shown that Aloe has an antioxidative effect. Its antigenotoxic and chemopreventive effect are also proven (14). The level of blood glucose was significantly decreased after oral administration of ethanolic extract of Aloe vera gel in STZ- induced diabetes (14). The aim of present study was evaluation of protect effect of Aloe vera gel on ovaries parenchyma 18 day embryo of female rats.

**Materials and Methods:**

Twenty female Sprague-Dawley rats (with weight 180-200 grams) were obtained from animal house of Shiraz University (Shiraz, Iran). The animals were kept in an experimental room for one week, for acclimatization to experimental conditions with 12 hour light and dark cycle. The animals were fed at laboratory chow and water ad libitum.

**Plant material and extraction:**

Fresh and healthy Aloe vera leaves weighting between 550-650 gr with approximate length 50-70

cm were collected from matured healthy plant and washed thoroughly with fresh running tap water. The leaves were dissected longitudinally and fleshy mucilaginous pulp (parenchymatous tissues) was selectively scraped out, leaving behind the thick epidermis layers. The scraped pulp was homogenized with ethanol 96% and distilled water (50/50, v/v). Thereafter solution filtrated, afterward its were evaporated till dryness under a rotary evaporator.

#### Induction of diabetes

Streptozotocin (STZ), purchased from Sigma Chemical Co, was dissolved in Distilled water immediately before use. The animal received STZ (50 mg/kg) intraperitoneal. Fasted blood glucose levels were assessed 48 hours after STZ injection as well as glycosuria to confirm the diabetic states. The rats were kept for 10 days to stabilize the diabetic condition. Only rats with a fasting blood glucose level of at least 250 mg/dl were used in the experiment. The rats were divided randomly in two groups.

Group A: diabetic control group that received distilled water once a day.

Group B: diabetic rats were treated orally with Aloe vera gel (400 mg/kg) for 18 days, once a day. Female rats were caged with males overnight. Females were considered 1 day pregnant on demonstrating vaginal plaque. At the end of 18 day,

blood glucose is measured and afterward, the animals were sacrificed and embryo removed and weight. Embryos were fixed in 10% formalin. Abdominal piece of whole embryo removed. The 5-6  $\mu$  serial sections were made using paraffin embedding techniques and stained by hematoxyline-eosin.

The numbers of primordial germ cell were measured in 200x magnifications. Histometrical studies were done using Olympus microscope and Olysia software.

#### Statistical Analysis:

Spss version 11.5 was used for statistical analysis. Group's variance was analyzed by student-t test for significant differences.  $P < 0.05$  was considered statistically significant.

#### Results:

The result showed that the STZ (50 mg/kg) induced diabetic mellitus in rats and blood glucose reached to  $471.67 \pm 19.2$  mg/dl. Administration of Aloe vera (400 mg/kg) reduced the blood glucose significantly so that reached to  $126.6 \pm 4.6$  mg/dl on the end of 18 days ( $p < 0.05$ ), (Figure.1). The mean of weight embryo in group A were increased  $5 \pm 0.28$  gram and group B were decreased  $4.36 \pm 0.18$  gram. The weights of embryo diabetic rats in two groups not significant (Figure.2- Table1).

Table1: The mean ( $\pm$  S.E) of parameter ovary in two groups

	Group A	Group B	P-value
Weight(gr)	$5 \pm 0.28$	$4.36 \pm 0.18$	$> 0.05$
Blood glucose	$471 \pm 19.22$	$126.6 \pm 4.6$	$< 0.05$
Primordial germ cell	$432 \pm 23.09$	$476 \pm 17.32$	$< 0.05$

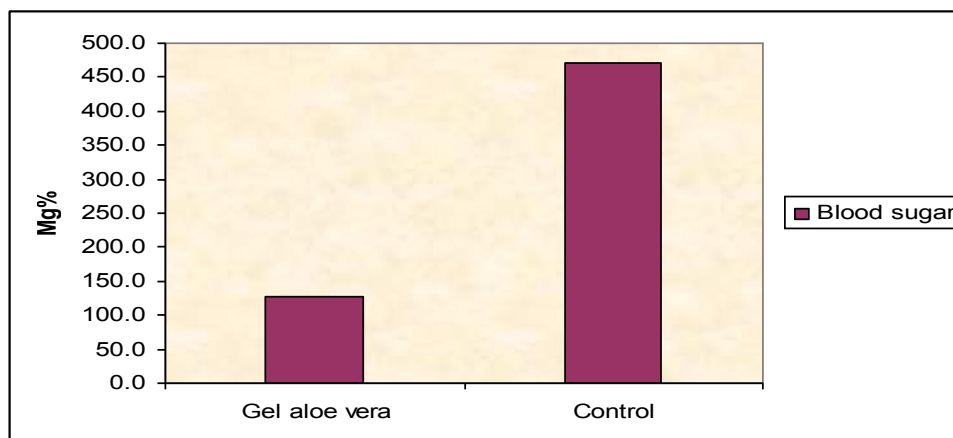


Figure.1: Mean ( $\pm$  S.E) of blood glucose embryo of rats after 18 days in two groups ( $p < 0.05$ ),  $n=5$

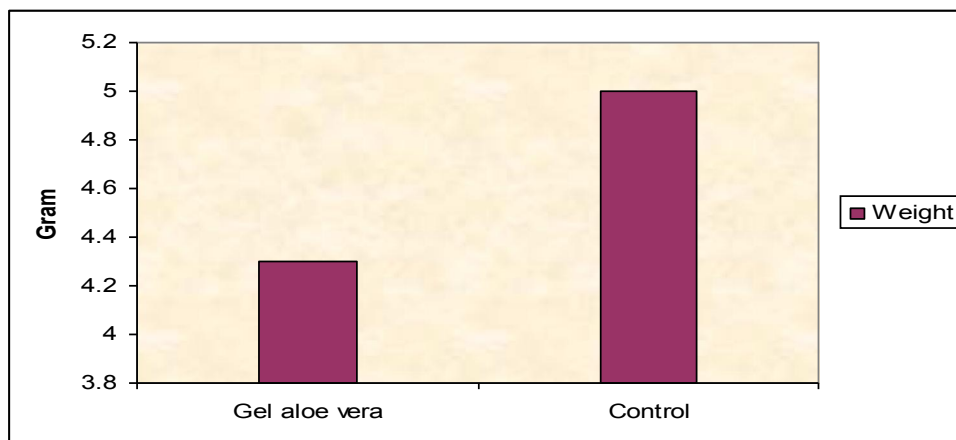


Figure.2: Mean ( $\pm$  S.E) of weight embryo of rats after 18 days in two groups ( $p>0.05$ ), $n=5$

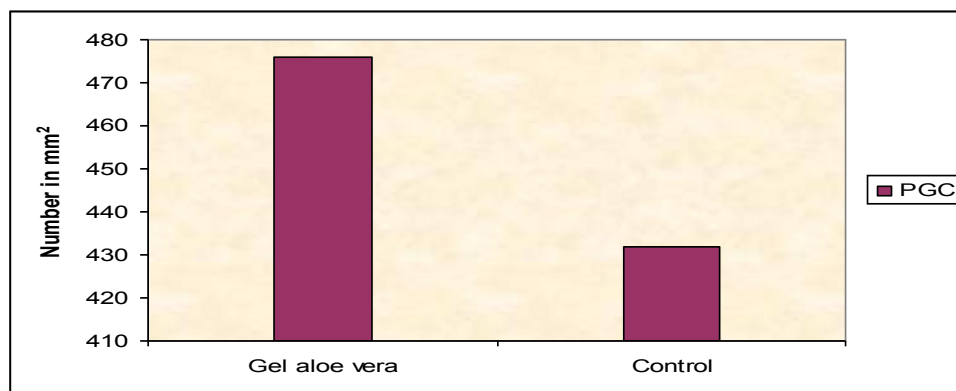


Figure.3: Mean ( $\pm$  S.E) of primordial germ cell embryo of rats after 18 days in two groups ( $p>0.05$ ), $n=5$

#### Histometrical Change:

Histological study showed that total numbers of primordial germ cell were increased in group B. It is significant ( $P>0.05$ ), (Table.1). Blood sugar is decrease ( $p<0.05$ ) but weight is not significant (Table1, Figure1, 2, 3,4,5).

#### Discussion:

In recent years, various plant extract have been claimed to be useful for the cure of diabetes mellitus, but few of them tested for their effects on body tissue of diabetic patient. In present study, we investigated the antidiabetic effect of Aloe vera gel on embryo of ovaries structure in STZ- induced female diabetic rats. STZ is a compound commonly used for the induction of type I diabetes in experimental rats. STZ caused diabetes by rapid depletion of B cell in pancreas Langerhance Island, which leads to a reduction of insulin release. In our studies, oral feeding of Aloe vera gel reduced blood

glucose level by 26.8%. Noor et al.(1) and Bolkent et al.(12) reported the antidiabetic effect of Aloe vera in diabetic mice induced by alloxan (500mg/kg,twice daily)(12). Helal et al. (17) and Rehman et al. (18) reported that the blood glucose is decreased in alloxan diabetic rats which treated by Aloe vera for 30 days (17, 18). The period of these treatments were similar to present study. Rajasekaran et al.(14) and Noor et al.(1)[2008] were observed that oral receiving of Aloe vera (300 mg/kg,daily) to diabetic rats reduced the blood glucose level (1,14). These results confirmed our results. AV treatment for 18 days showed potential hypoglycemic activity in oral glucose tolerance test and antidiabetic activity in alloxanized rats (18). Aloe vera may exert its antidiabetic effect by supporting and maintenance the death of B cells or it may permit recovery of partially destroyed B cells (1). Also, the hypoglycemic action of the extract of herbal plants may be possible

through the insulinomimetic action or by other mechanism such as stimulation of glucose uptake by peripheral tissue, inhibition of endogenous glucose production or activation of gluconeogenesis in liver and muscles (19).

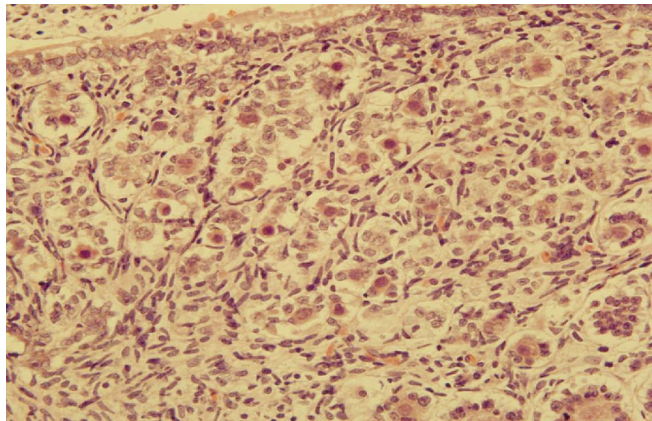


Figure.4: Histological feature of ovary in group A (X200, H&E).

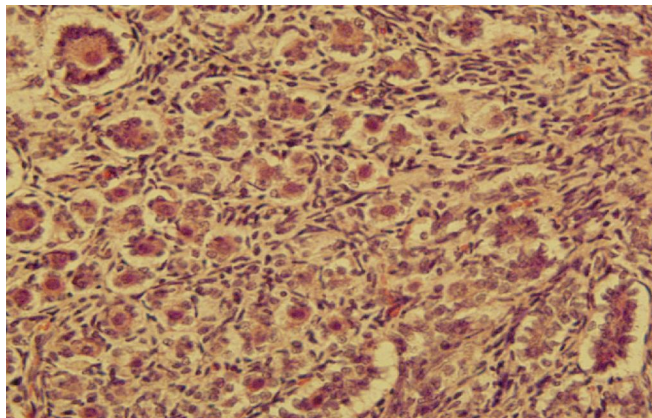


Figure.5: Histological feature of ovary in group B (X200, H&E).

Rajasekaran et al., (2006) reported that, the increased levels of plasma insulin indicate that the A. vera gel extract stimulates insulin secretion from the remnant B cells or from degenerated B cells (20). Diabetes is characterized by weight loss (5, 19) and it was also seen in this study. The body weight decreasing was prevented by administration of Aloe vera. This effect of Aloe vera was reported in limited studies including (1, 18). The mean primordial germ cell increased in rats that fed Aloe vera extract compared to control group, but not significantly. Ovaries dysfunction in diabetes mellitus may be associated with imbalance glucose utilization, follicular atrophy and impaired steroidogenesis (6).

Tatewakil et al. (21) observed the percentages of the primary follicles decreased in diabetic mice (21). Ballester et al.(5) reported that some degenerative signals of ovarian structure caused to observed small cells, with picnotic nuclei spread in the ovary, scavenger cells in follicles, an increasing the intracellular vacuoles in follicles and corpora lutea in diabetic rat ovary(5). Deficiency of insulin is one of the determinant factors that influence ovary structure in diabetes and, thus, the Aloe vera – induced recovery of serum insulin levels would be one of the mechanisms involved in the improvement ovary structure and function (20). Oxidative stress, through the production of free radices has been implicated in the progression of long-term diabetes complications including microvascular and macrovascular dysfunction (8). Anti – oxidants such as phenolic compounds and saponins in the gel extract of Aloe vera may be responsible, in part, for its protective effect on ovary structure in diabetic rats. The results of present study showed that Aloe vera brings back the blood glucose and body weight to normal in the diabetes rats. After treatment with Aloe vera, the numbers of normal primordial germ cell were increased. It is shown that A. Vera could have a beneficial and supporting effect on ovarian tissue and folliculogenesis if used as a hypoglycemic agent in diabetes.

#### Acknowledgements:

This work was supported by Shiraz University and Cellular and Molecular research center of Yasuj University of Medical Sciences which is greatly appreciated.

#### Corresponding Author: Reza Mahmoudi

Cellular and Molecular research center, Yasuj University of Medical Sciences, Yasuj, I.R.Iran.

E-mail: [rmahmoudi40@yahoo.com](mailto:rmahmoudi40@yahoo.com)

#### References:

- 1- Noor, A., S.Gunasekaran, A.Sooai Manickeam and M.A. Antidiabetic activity of Aloe vera and histology os organs in Streptozotocin induced diabetic rats. *Current Sci*;2008;94: 8-25.
- 2- Sharpe RM.Regulation of spermatogenesis. In: Knobil E,Neill JD.(eds).The physiology of reproduction. 2<sup>nd</sup> ed. New York (NY): Raven Press; 1994:1363-434.
- 3- Abdollahnejad, A. Gol, A. Dabiri, S. Garlic effects on reproductive complications of diabetes mellitus in male rats. *Physiology and Pharmacology*; 2009; 13 (3): 297 -307.
- 4- Borland, K. Mita , M.The actions of insulin- like growth factor I and II on cultured sertoli cells.*Endocrinology*;1984;114: 240-246.

- 5- Ballester, J, Dominguez, J, et al. Tungstat administration improves the sexual and reproductive function in female rats with Streptozotocin-induced diabetes. *Human Reproduction*; 2007; 22: 2128-2135.
- 6- Griswold MD. The central role of sertoli cells in spermatogenesis. *Semin Cell Dev Biol* 1998; 9(4):411-6.
- 7- Omotayo O, Siti A and etal. Antioxidant protective effect of Glibenclamid and metformin in combination with Honey in pancreas of streptozotocin induced diabetic rats. *Int. J.Mol.Sci*;2010;11:2056-2066.
- 8- Nirmala, A., S. Saroja, H.R. Vasanthi and G. Lalitha. Hypoglycemic effect of Basella rubra in Streptozotocin induced diabetic albino rats. *Journal of pharmacognosy and Phytotherapy*; 2009; 1: 025-030.
- 9- Alper Okyar, Ayse Can et al. Effect of alovera leaves on Blood glucose level in type I and type II diabetic rat models. *Phytherapy Research*;2001;15:157-161.
- 10- Philipps AF, Porte PJ, Stabinsky S, Rosenkrantz TS, and Raye JR, Effects of chronic fetal hyperglycemia up on oxygen consumption in the ovine uterus and conceptus. *J clin invest*; 1984; 74(1): 279-286.
- 11- Vetrichelvan, T. and M, Jegadeesan. Anti-diabetic activity of alcoholic extract of Aerva lanata (L.) Juss. Ex Schultes in rats. *J. Ethnopharmacol*;2002; 80:103-107.
- 12- Bolkent, S., N. Akev, A.Can, S. Bolkent, R. Yanardag and A. Okyar. Immunohistochemical studies on the effect of Aloe vera on the pancreatic- cells in neonatal Streptozotocin-induced type-II diabetic rats. *Egyptian J. Biol*;2005; 7: 14-19.
- 13- Can, A. and A. Nuriye. Effect of Aloe vera leaf gel and pulp extract on the liver in type-II diabetic rat models. *Biological and Pharmaceutical Bulletin*;2004;27:694-698.
- 14- Rajasekaran S, and etal. Antioxidant effect of Aloe Vera gel extract in Streptozotocin – induced diabetes in rats. *Pharmacol Reports*; 2005; 57:90-96.
- 15- Josias H. Composition and Application of Aloe Vera Leaf Gel. *Molecules*; 2008; 13:1599-1616.
- 16- Zhang, L. and I.R. Tizard. Activation of a mouse macrophage cell line by acemannan: the major carbohydrate fraction from Aloe vera gel. *Immunopharmacol*;1996; 35:119- 28.
- 17- Helal, E.G.E., M.H.A. Hasan, A.M. Mustafa and A. Al-Kamael. Effect of Aloe vera extract on some physiological parameters in diabetic albino rats. *Egyptian J. Hospital Medicine*;2003; 12: 53-61.
- 18- Rehman, S., S.A. Jafri, S. Hassan, I. Ahmed and M. Naim. Study on diabetic effect of Aloe vera extract on alloxan induced diabetic rats. *Libyan Agriculture Research Center Journal International*; 2011;2: 29-32.
- 19- Vestegard H, Studies of gene expression and activity of hexokinase, phosphofructokinase and glycogen synthase in human skeletal muscle in states of altered insulin stimulated glucose metabolism. *Dan Med Bull*; 1999;46:13-34.
- 20- Rajasekaran, S., K. Ravi, K. Sivagnanam and S. Subramanian. Beneficial effect of Aloe vera leaf gel extraction on lipid profile status in rats with streptozotocin diabetes, *Clinical and Experimental Pharmacology and Physiol*;2006; 33: 232-237.
- 21- Tatewakil, R., H. Otani, O. Tanaka and J. Kitadal. A morphological study on the reproductive organs as a possible cause of developmental abnormalities in diabetic NOD mice. *Histology and Histopathol*;1989;4: 343-358.

12/24/2012