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(RESEARCH ARTICLE)

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The effect of aqueous extract of *Moringa oleifera* seeds on the eyes of diabetic rabbits

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Abstract

The study aimed to induce, diabetes with Alloxan in local. white rabbits, study its effect on eye tissue, and treat it with aqueous extract of *Moringa* nibbar seeds at a concentration of 200 mg/kg, and observe ,the tissue changes.. After the end of the injection and dosing period after 30 days, the animals were killed and the eye was removed to perform histological techniques on it. 20 white rabbits were taken and divided into three groups: The first group: control group. This group ,was dosed with distilled water, The second group: The group, treated with Alloxan. This group was. injected, with Alloxan intraperitoneally at a dose of 175 mg/kg and, The third group: The group treated, with aqueous extract of *moringa* seeds at a concentration of 200 mg/kg for 30 days.

Keywords: Diabetes; White rabbits; Eye; *Moringa*

1. Introduction

Medicinal plants are one of God's blessings on creation, which is used as a source of food and medicine. Humanity has directed its thinking to obtain the full benefit from these blessings. Since ancient times, man has invented several ways to use these plants as a source of medicine. He has used the roots, leaves, seeds, and many other plant products as medicine. Medicinal plants contain some organic compounds that are Important for the human body, including: phenols, tannins, alkaloids, carbohydrates, flavonoids, and steroids (Edeoga, 2005; Mohammed and Ahmed,2024a).

The *moringa* tree is one of the most effective sources, of phytochemicals. It contains many biologically active compounds,. With antioxidant, antimicrobial, and immunomodulatory activities, that can improve the productive, and reproductive, performance and health ,of animals. Hence, this plant has gained, great interest for use as ,a feed additive or feed in the livestock industry.(Mohammed and Ahmed,2024b).

It is a disease that occurs, when the pancreas is unable' to produce enough insulin, or when the body is unable to use the insulin, it produces effectively. (MS, 20020a). It is also defined as chronic increase in the ,level of sugar in the blood., which is accompanied by thirst and abundant blood sugar. Urine, weakness, weight loss or obesity Sudden episodes of loss of consciousness that often lead to a fatal coma [Buysscher & Hermans , 1998; Muhammad bin Saad Al-Hamid ,2008). also defined diabetes as an imbalance in the process of sugar metabolism that leads to a high sugar level Glucose (glucose) is abnormally high in the blood, for various reasons; which ,may be psychological, muscular, due to excessive, sugar intake, or due to genetic factor. It occurs as a result of, a defect in the secretion; of insulin from the pancreas. The amount of insulin, that is secreted may be, less than what is required, or there may ,be a complete cessation ,of its production. This condition is insulin insufficiency.

During 1997, a new classification of diabetes was proposed by a group of experts (ADA) to replace the one developed in 1979 by (NDDG) which was confirmed in 1980 by the World Health Organization. The expression for insulin-dependent and non-insulin-dependent diabetes was deleted. Insulin-dependent, a term that the expert committee

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considered to be a somewhat therapeutic classification rather than an etiological classification. The new classification of diabetes mellitus defines both type 1 diabetes, type 2 diabetes, gestational diabetes and others with specific etiologies (Buysschaert & Hermans., 1998).

1.1. Type 1 diabetes

Previously: Insulin-dependent diabetes represents about 10% of all diabetes, and usually begins in childhood after autoimmune destruction of insulin-producing cells known as B cells of the pancreatic islets. There are two forms of this type of diabetes: autoimmune, the most common, and idiopathic, which is rare (Buysschaert.,1998).

1.2. Type 2 diabetes

Which was previously called (non-insulin dependent): This is the most common, form of diabetes, representing approximately, 90% of diagnosed cases,. This type of diabetes, usually manifests itself in adulthood, and it is a chronic disease that develops over time. Gestational diabetes: This type of diabetes is a transient condition that disappears in the weeks after childbirth , Women who have gestational diabetes; are at greater risk of, developing type 2 diabetes later. (Naylor et al., 1997).

1.3. Botanical description of *moringa*

Moringa is a fast-growing tree. It is one of the species of the *Moringacea* family. It is an angiosperm plant. It is small to medium-sized tree ,with a height of approximately 3-12 meters, and a diameter of 20-40 centimeters. It has an upright, brittle stem and a soft, white bark tending to grey. It contains pendulous branches. dropping branches picture (2a-1).

Its scientific name is *Moringa oleifera*. Lam. The name that is synonymous with it is *Moringa* pterygosperma Gaert, the origin of the *moringa* tree.

Oleifera is from India, where it grows in tropical and subtropical region and is grow all over the world with a rainfall rate of (7600-2500) mm annually, a temperature of 18 to 40 degrees Celsius, and a pH between (5.4-8). Its cultivation has spread nowadays in the Middle East. In Asian countries and Africa, and it is still spreading in other regions of the world (Leone *et al*., 2015).

The tree has a tuberous tab root, and this helps it withstand lack of water and drought conditions. This plant may tend to be sensitive to winds because it does not have a good and deep root system, as it was mentioned that the roots resulting from planting cuttings (pens) are less deep than those resulting from planting Seeds (Choudhary, 2016).

As for the leaves, they are alternate, of the type of decompound leaf, as they contain a main axis 30-75 cm long and a common branch, and they are long petioles with 8-10 pairs of leaflets. Each pair consists of two opposite leaves, oval and a single leaflet at the apex, which is inverted oval and is the most common. Long, the lower pair of leaflets is triangular, and the edge of the leaflets is not serrated, picture (c2-1). It is often mistakenly believed that this tree belongs to legumes because of its leaves that resemble the leaves of leguminous plants(Qureshi & Solank .,2015).

As for the flowers, they are small, white, bisexual. They contain five sepal leaves and five petals surrounding five stamens and five sterile staminodes. The flowers are carried on a floral stand in inflorescences 10-25 cm long and 2.5 cm in diameter. They have a fragrant smell and are drooping. They also contain an ovary. (Chaudhary&Chaurasia., 2017).

2. Materials and working methods

The animals used in this study were local white rabbits, Oryctolagus cuniculus, whose ages ranged between 10-14 month and their weights ,ranged between ;(1150-1850) grams. They were, raised in the animal house, of the College-of- Veterinary Medicine - Tikrit University,. The rabbits placed in iron cages, covered with; metal covers. It has a floor, the cleanliness of the cage must be taken into account by replacing the sawdust two to three times during the week.

The animals were fed, with a diet consisting ;of 35%, wheat, 35%, yellow corn, 20% soybeans, and 10% concentrated animal protein ,and vitamins, preservatives, and antifungal; materials. They were given water , food continuously throughout the study .(Balducci *et al*., 2001).

2.1. Inducing diabetes in experimental animals

The animals were inject with, Alloxan prepared at a concentration, of :175 mg/kg for ;a period of 3 days intraperitoneally. Immediately after the injection, the animals; were provided with food -and a 10% glucose solution to prevent ,a drop in the level of; blood glucose that occurs as a result of the destruction ,of pancreatic beta cells. As for the control animal, they were given food- and water in a regular manner. Sufficient without the use of alloxan .(Dubey and Dixit 1994).

Animals with a concentration- higher than (300 mg per 100 ml), were considered diabetic, as they showed signs of extreme fatigue, frequent ,urination, and lethargy. (Witek et al., 2001).

2.1.1. Glucose solution 10%

It was prepared by taking 10 grams of glucose powder and dissolving it in 100 ml of drinking water and keeping it in the refrigerator until use (Al-Ghabsha and Al-Abayji 1989).

2.2. Preparation of cold water extract of *moringa* seeds

Moringa seeds were obtained from private shops that sell medicinal herbs in Baghdad Governorate. They were diagnosed by professors specialized in plant classification at the University of Karbala. They were then cleaned, leaves and foreign materials removed from them, then dried in the shade with constant stirring until dry seeds were obtained. The seeds were ground with a grinder. Laboratory medicinal herbs until a fine powder is obtained. Take (20) gram of dry powder ,and mix with (400) ml; of distilled water ,using an electric blender; and leave for 24 hour at- room temperature. Then filter the mixture using several layers, for the purpose of getting rid of plankton, and placing In the, centrifuge at a speed -of (3000) rpm for (10) minutes, then filter- the extract using Whatman ;NO type 101 filters papers to obtain a clear solution ,and dry it in the oven after placing it in clean, sterile glass dishes at 40°C and store it in the refrigerator in sterile glass containers. And tightly closed until use (Hernandezi et al., 1994).

control group. This group was dosed with distilled water.

- **The group treated with Alloxan**. This group was injected with Alloxan intraperitoneally at a dose of 175 mg/kg.
- **The group treated with aqueous extract of *moringa*** seeds at a concentration, of 200 mg/kg for 30 days.

3. Results

3.1. The first group (control group)

3.1.1. Eye

The retina contains several nervous tissue layers, the inner nerve fiber layer extending from the ganglion cells, which were found scattered and with prominent nuclei, as well as the inner plexiform layer, in which some apparent delamination was found, with a spongy pattern for that wide layer adjacent to the, inner nuclear layer, which was found two rows of The nuclei of nerve cells are aligned with each other. The outer plexiform layer contains fine nerve fibers extending from the outer nuclear layer, which has hyperplasia and the alignment of its nuclei, which are adjacent to the layer of rods and cones, as they appear normal (fig. 1).

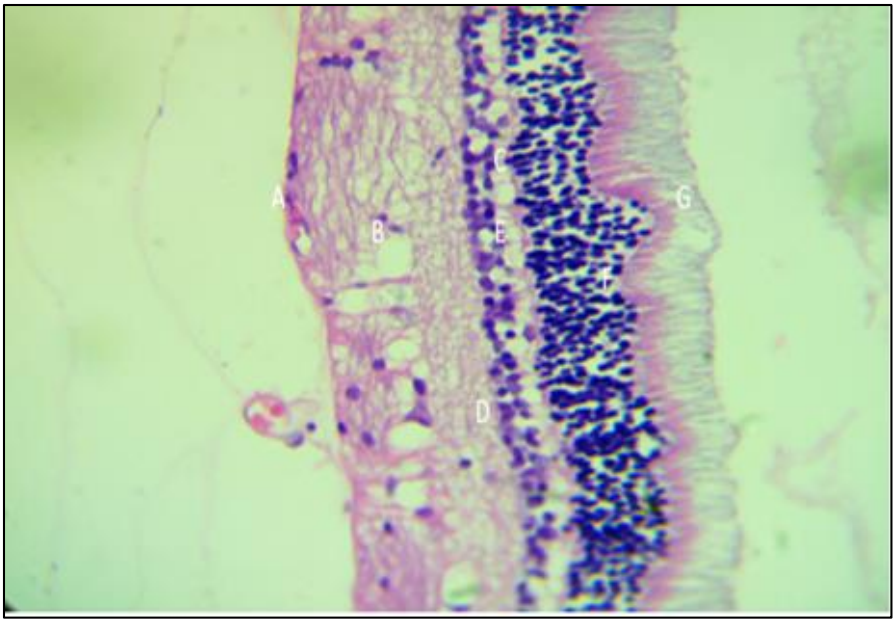

Figure 1 Peripheral neural layer (A) Neuroganglionic layer (B) Vascularity of the internal neuronal plexiform layer. (C) Inner nuclear layer. (D) Narrow outer tegmental layer. (E) Hyperplasia of the outer nuclear layer. (F) Orthomorphic layer of rods and cones. (G)(H&EX40).

3.2. The second group (the group treated with alloxan).

The retina contains a fine bundle of nerve fibers surrounding the retina, with a layer of some ganglion cells in which there are many gaps interconnected, and the inner plexiform layer also contains many small, foam-shaped gaps. As for the inner nuclear layer, it contains many cell nuclei. The disassembled layer, around which a number of spaces and gaps were found, was aligned with the outer plexiform layer adjacent to the outer nuclear layer, which had hyperplasia, compactness, and condensation of the nuclei of the neurons in it. The layer of light-receiving cells (rods and cones) was wide, with dispersion and irregularity in its presence in the layer of receiving cells. The light has fig. (2).

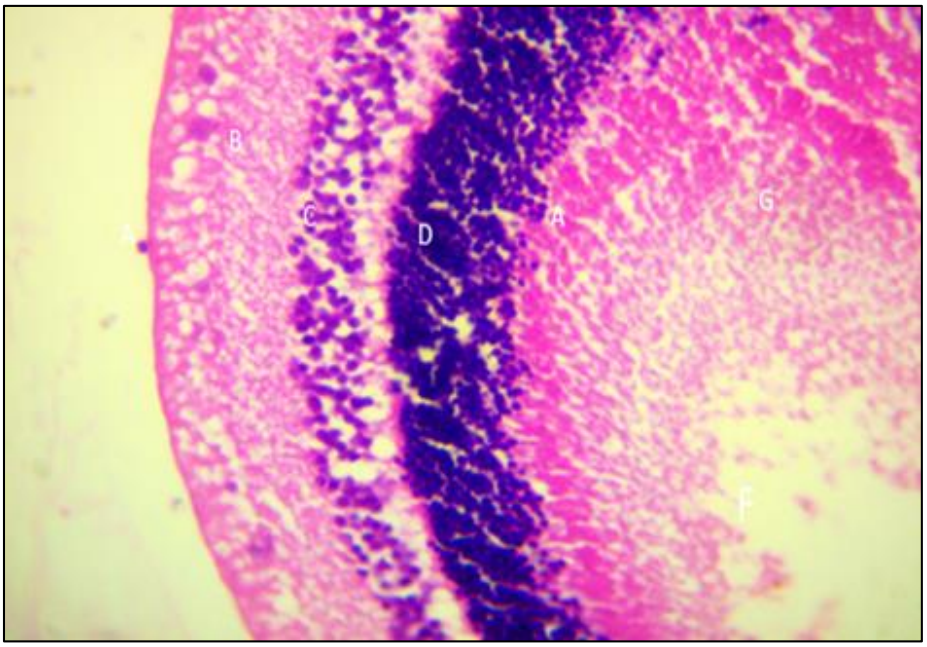

Figure 2 Retina, nerve fiber layer (A), ganglion neuron layer with gaps around it, (B), foamy vacuoles in the inner plexiform (C), dispersion and rupture in the inner nuclear layer (D), necrosis of the outer nerve plexiform (E).), hyperplasia of the outer nuclear layer (F) scattering and irregularity of the photoreceptor layer (G) (H&EX40)

3.3. The third group: (the group treated with aqueous extract of *moringa* seeds at a concentration of 200 mg-kg)

The retina was characterized by the presence of the outer plexiform layer, which contained some ganglion neurons, as this layer appeared in a spongy shape, in addition to the presence of the nuclear layer, as the nuclei of nerve cells appeared dispersed and continuous with the neural plexiform layer. The nuclear layer had a density in the number of nuclei (hyperplasia). This layer appeared in a stratified manner, and in front of it was the layer of photoreceptor cells (rods and cones), as some disintegration appeared on it.

The vascular layer behind the retina has severe blood congestion and the scleral layer consists of a bundle of loose fibrous colloid fibers (fig. 3).

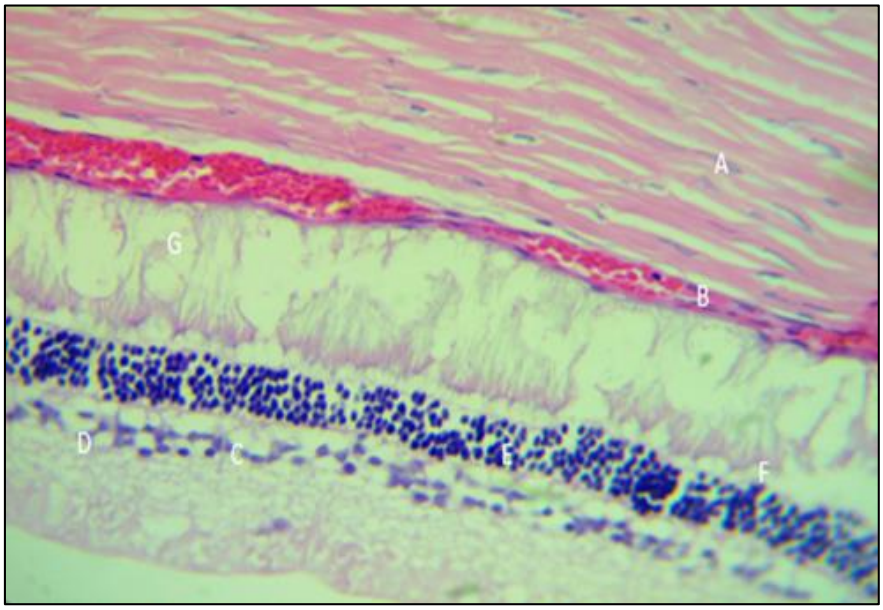

Figure 3 Sclera (A), choroid (B) with blood vessels, inner pterygoid layer (C), inner nuclear layer (D), outer pterygoid layer (E), outer nuclear layer (F), rods and cones layer (G)

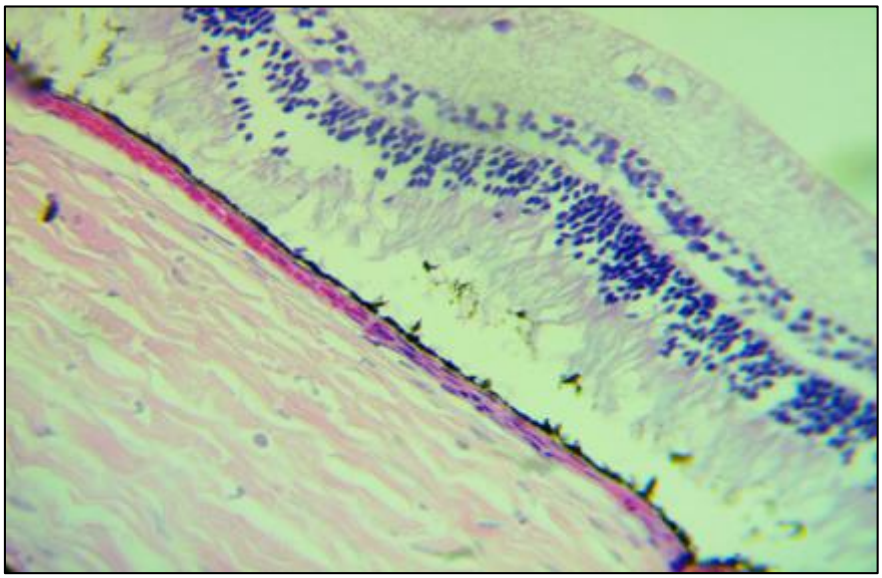

Figure 4 Retina and sclera (A), choroid with blood vessels and pigment carriers (B), ganglion cell layer (C), inner pterygoid layer (D), inner nuclear layer (E), outer pterygoid layer (F), outer nuclear layer (G) Layer of rods and cones. (H)(H&EX40)

The retina contained nerve fibers at the periphery of the layer with the presence of a number of ganglion neurons, and that plexiform layer appeared in a spongy shape adjacent to the inner nuclear layer, as it was found in it disintegration of a number of its nuclei. The outer plexiform layer contained nerve fibers extending from the outer nuclear layer with accumulated and continuous nuclei. With the light-receiving layer composed of rods and cones.

The choroid layer contained congested blood vessels with a band of dark melanocytes.

The scleral layer contained numerous loose bundles of colloidal fibers (Fig. 4).

The layer of nerve fibers is continuous with large ganglion cells, as their nuclei are spherical, pale in color, with bulging cytoplasm, and the layer of inner plexiform nerve fibers appears spongy and widespread, united with the inner nuclear layer, which has multiple dispersed nuclei and is parallel narrow outer plexiform layer, the outer -nuclear layer. It appeared as hyperplasia of the cells of this layer and its many normal-shaped nuclei. Rods and cones appeared in form long, pale-pigmented threads united with the choroid layer, which is composed of blood vessels and pigment-bearing cells and adjacent to the sclera, which is composed of bundles of colloidal fibers. (fig. 5).

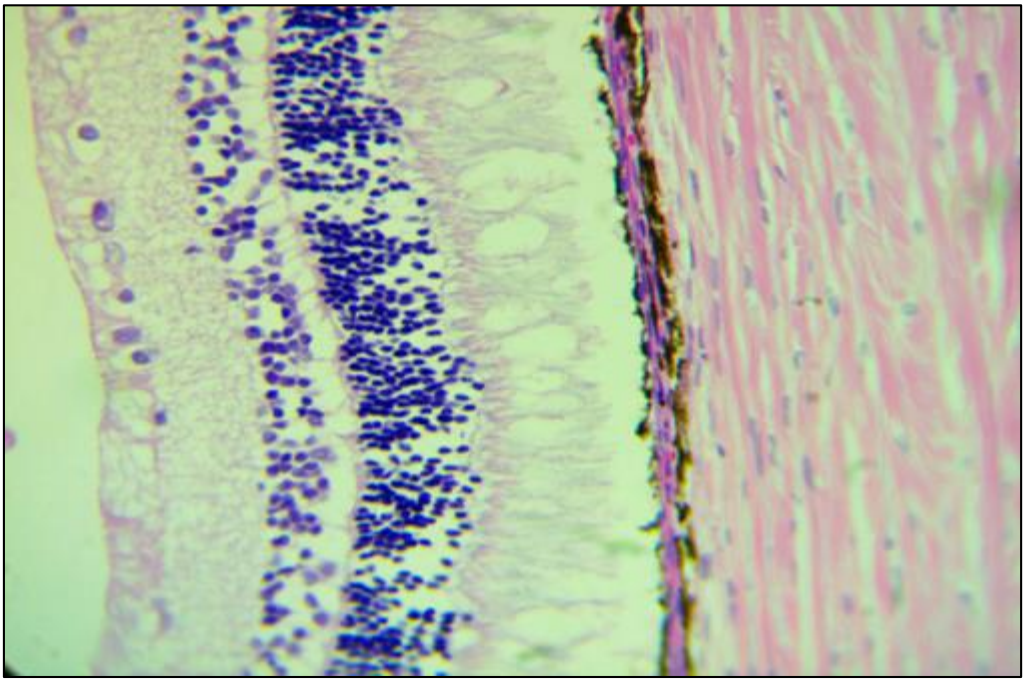

Figure 5 The scleral layer (A), the choroid with pigment carriers, (B), the ganglion cell layer, (C), the inner tegmental layer. (D), the inner nuclear layer. (E), the outer tegmental layer, (F), the outer nuclear layer, (G), the layer of rods and cones. (H)(H&EX40)

The retina contains a layer of nerve fibers continuous with the ganglion cells and with the plexus or inner nerve plexiform layer. It was noted that there was a small number of ganglion neurons, and there was dispersion and a decrease in the numbers of the inner nuclear layer.

The outer plexiform layer, is narrow communicates with the outer nuclear layer; which is composed of large numbers of neuronal nuclei. The light-receiving layer, which is composed of (rods and cones), has a shrinkage from the outer nuclear layer, while being adjacent to the choroid layer, which appeared away from the sclera (fig. 6,7).

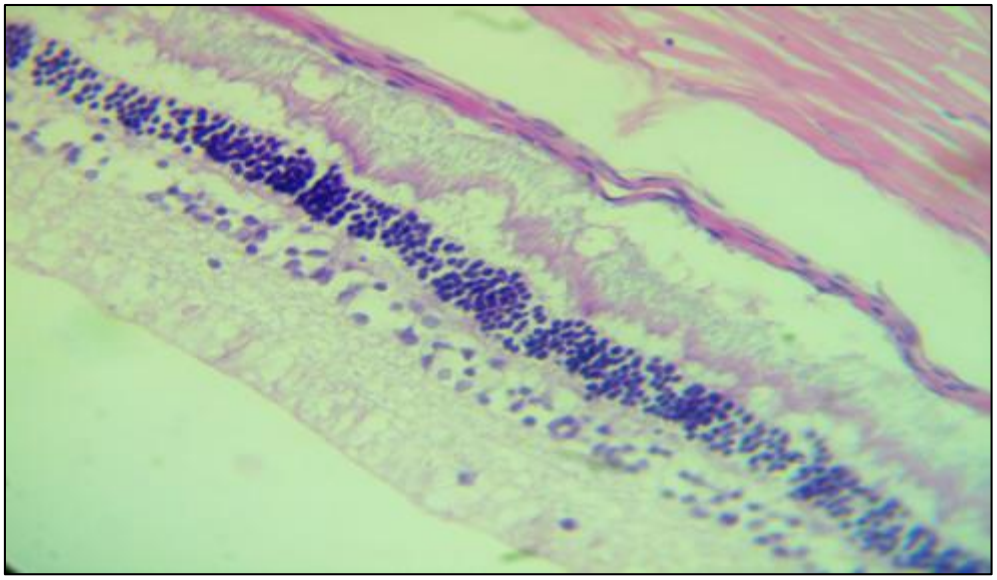

Figure 6 Ganglion neurons (A), inner plexiform layer, (B), inner nuclear layer, (C), outer plexiform layer, (D), outer nuclear layer ,(E), rods and cones layer, (F) (H&EX40)

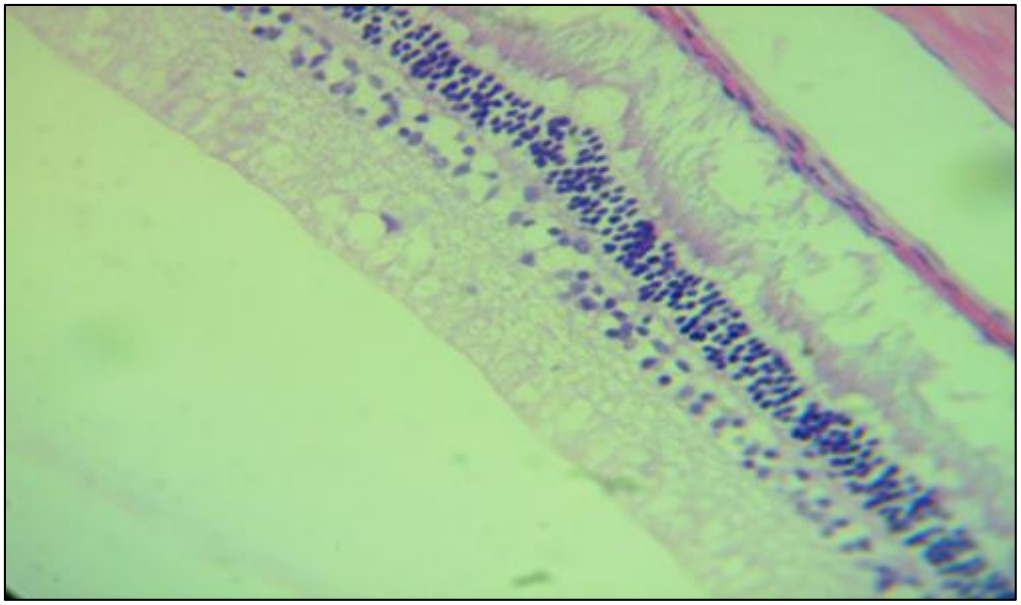

Figure 7 Ganglion neurons, (A), inner plexiform layer, (B), inner nuclear layer, (C), outer plexiform layer, (D), outer nuclear layer,(E), rods and cones layer, (F) (H&EX40)

4. Discussion

The *moringa* tree helps diabetics maintain blood sugar levels, as a result of it containing chlorogenic acid, which helps reduce the percentage of glucose and protein in the urine. The isothiocyanate compound found in *moringa* leaves helps protect against diabetes.

These studies: A small study published in the International Journal of Food Sciences and Nutrition indicated that adding *moringa* to meals contributed to reducing the high level of sugar; in the blood, but reason was not clear, and it is believed that this is due to some plant compounds found in the *moringa* plant. Which are called isothiocyanates (in English: Isothiocyanates), while one preliminary study indicated that taking *moringa* supplements along with a sulfonylurea type drug (in English: Sulphonylure) does not contribute to improving the levels of cumulative glucose in the blood, and

it should be noted that taking The *moringa* plant may lead to a severe decrease in blood sugar levels in diabetics. Therefore, attention must be paid to and monitoring of blood sugar levels if using *moringa*. It is always recommended to consult a doctor before taking any type of new herbs or nutritional supplements, as the *moringa* plant is a source of... Rich in amino acids, vitamins, and minerals, it also contains the compound isothiocyanates, which is a compound similar to sulforaphane found in cruciferous vegetables, which provides many health benefits. The *moringa* plant is also a rich source of dietary fiber and antioxidants. Including polyphenols, flavonoids, quercetin, and chlorogenic acid, which helps lower blood pressure, in addition to their good content of vitamin A, vitamin C, and other vitamins. B, which contributes to improving the digestion process. In addition, the *moringa* plant possesses antibacterial, antifungal and antimicrobial properties. It helps fight infection and inhibits the growth of many microbiology.

5. Conclusion

We conclude through research that the *moringa* plant has the ability to lower blood sugar levels and also had a significant impact on causing histological changes in the eyes.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

statement of ethical approval were optained.

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