Histological effect of melatonin hormone on adult rat's prostate

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Abstract

Background: The prostate is the largest of the accessory glands of the male reproductive tract. Its secretion serves as a diluent and vehicle for transport of sperms from male to female, so its function is very important for the normal fertility. Melatonin is the basic neuro-hormone of the pineal gland, regulates the sexual and reproductive activities in all mammals including man.

Objective: This work aimed to study the effect of different doses of dietary melatonin on adult rat's prostate, *"histologically*".

Methods: Melatonin was supplied to adult Wister albino rats, for successive 30 days. Rats were divided into 6 groups. Group I was the control. Group II, III, IV, V and VI were given (mixed with their diet) a daily dose of 125, 250, 500, 750 and 1000 µg / kg body weight, respectively. The dietary melatonin was supplied to rats mixed with their food. After the last day of treatment, animals were killed under effect of anesthesia; prostate was removed for histological study.

Results: The results showed significant beneficial effects on prostate by normal therapeutic dosages, whereas significant damaging effects were seen with further stepping up doses.

Conclusion: Dietary melatonin has good effects on the rat's prostate within therapeutic doses, whereas it had highly damaging changes in overabundance.

Keywords: Prostate, melatonin, and fertility.

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Introduction

The prostate is the largest of the accessory glands of the male reproductive tract. Its secretion serves as a diluent and vehicle for transport of sperms from male to female, so its function is very important for the normal fertility [(1)]. Melatonin is the basic neuro-hormone of the pineal gland. This hormone evidently plays an important regulatory role in the sexual and reproductive activities in all mammals including man [(2 & 3)].

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Melatonin limits human prostate cancer cell growth by a mechanism which involves the regulation of androgen receptor function but it is not clear whether other mechanisms may also be involved [(4)], hence, it would be of great interest to study the relationship between melatonin and prostate function.

Materials and methods

Adult male Wister albino rats (8 weeks old), 48 in number, were used in this work. They were kept in an animal room, with a temperature of 22±2Cº, the light - dark cycle was 12:12. Water was offered *ad libitum*. They fed a control diet with free access to food, except for one and half hour prior to melatonin containing meal. Dietary melatonin was provided as a single daily dose, 2 hours prior to sunset.

Animals were divided into 6 groups, each consisting of 8 rats. Group I was the control: rats were provided with the same type of drug containing meal, but no drug was added (placebo), though, they were also deprived from food one and half hour prior to the time of treatment as other groups. Group II, III, IV, V and VI were given dietary melatonin as a daily dose of 125, 250, 500, 750 and 1000 µg/kg body weight, in sequence, for 30 successive days. The dietary melatonin was supplied to rats by mixing it with their food, in only one meal, 2 hours prior to sun set. After the last day of treatment, all animals were killed by dissection under effect of diethyl ether. The whole prostate was removed, separated from the surrounding connective tissues under a dissecting microscope, weighed by an electric sensitive balance and used for paraffin section, using Bouin's solution for fixation and (Haematoxylin & Eosin) for staining [(5)], then 5 serial sections of 5 µm thickness from the left lobe were studied.

Histological study was done both as descriptive and morphometric by light microscope. The morphometric data were estimated by using Zeiss Integrating Micrometer – disk Turret I of 25 point system (which measures the relative surface area by counting the points superimposed through a disk put on the microscopic eye piece during slide examination, so the number of these points positively related with the relative measurement of the surface area), the total points falling on the fibromuscular stroma were calculated. From each section 5 fields were taken randomly examined at 150X magnification. Also by using objective micrometer used on a light microscope; by which a distance of 10µm could be calculated, the average height of the glandular cells, as well as the diameter of their nuclei, were estimated. All the values were taken as mean ± SD of 8 rats. The significance of difference between each of treated groups and its control was evaluated by student – t – test [(6)].

Results

Descriptive and morphometric studies for all groups were done, as follows:

Prostate weight was affected significantly in all groups; it was enlarged with the increasing doses of melatonin till the dose 500µg/kg, and then regressed gradually with increasing doses (table 1).

In all of the treated groups; the cells kept their arrangement to form the prostatic mucosal glands, showing the general architecture of a typical prostatic gland with its papillary pattern of ingrowths (Fig.2). The glandular epithelial lining was appeared to get taller with the increment of melatonin dose (Fig.2, 3 & 4); till the dose of 500µg/kg, after which, it seemed to be regressed (Table2). The nuclei of the glandular epithelial cells; gradually got lighter in color and larger, with the increase in melatonin doses till the dose of 500µg/kg, afterwards it gradually regressed and became darker, in the same manner as their cells did (Table2).

The smooth muscular and fibrocollagenous stroma was increasingly enlarged with the increase in the dose of melatonin (because the number of counted points seen through the eye piece- disk was increased) as shown in table 3. There was an increase in the vascularity in all of the treated groups, proportionate positively with the given doses of melatonin.

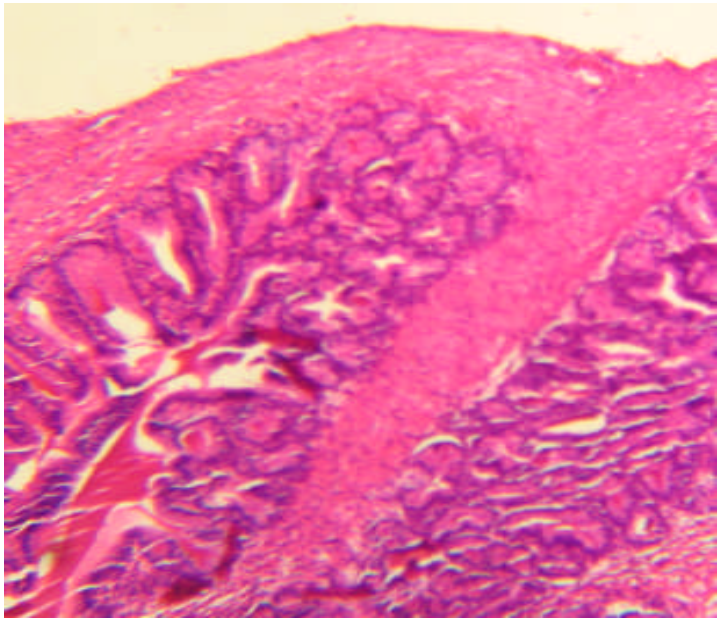

Figure 1: Prostate of control adult rat, X 200, H & E.

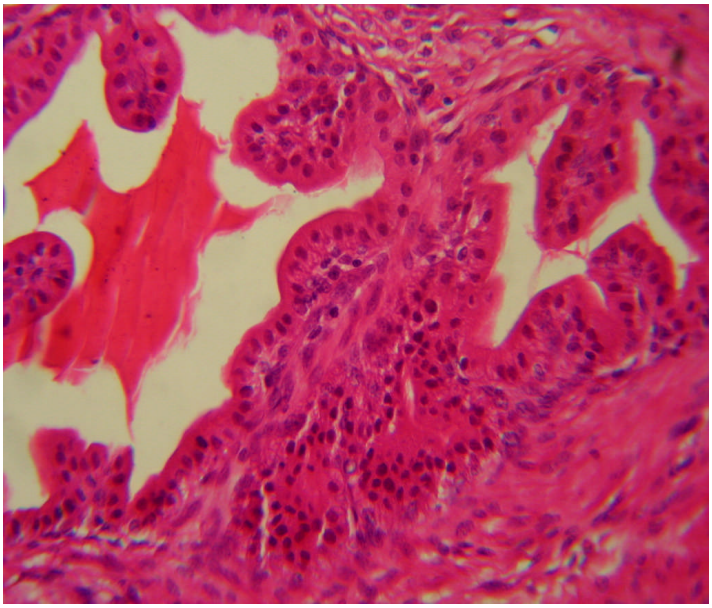

Figure 2: Prostate of adult rat treated with 500µg/kg dose, X 200, H & E.

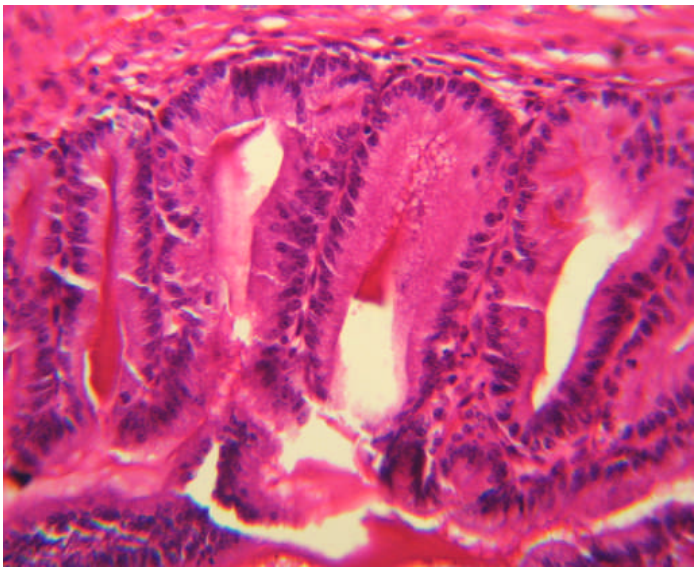

Figure 3: Prostate of adult rat treated with 500µg/kg dose, X 200, H & E.

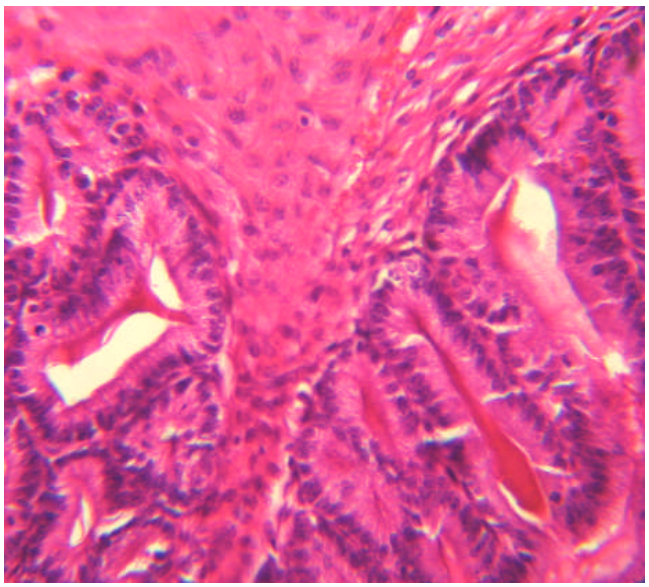

Figure 4: Prostate of adult rat treated with 1000µg/kg dose, X 200, H & E.

Table1: the effect of melatonin on prostatic weight of adult male rats.

Daily dose of melatonin in µg/kg body weight	***Prostatic weight at autopsy(mg)***
Control	74.2±3.3
125	78.9±2.1*
250	85.2±2.2**
500	91.9±3.3**
750	69.6±1.7**
1000	61.2±1.3**

-Results were expressed in mean± SD of 8 rats.
-The difference of each dose-group was statistically significant when compared with its control:
(* P<0.0002; ** P<0.0001).

Table2: Average cell height and nuclear diameter in µm, in the prostate of adult rats treated with dietary melatonin.

Daily dose of melatonin in µg/kg body wt	***Average cell height of prostatic epithelium (in µm)***	***Average nucleus diameter of prostatic epithelium (in µm)***
Control	39.2±1.4	21.4±1.3
125	41.2±1.5*	22.6±1.1†
250	42.8±0.9**	22.9±1.6††
500	44.1±1.6***	23.5±0.7‡
750	37.1±0.8**	20.2±1.1†
1000	32.3±1.4***	19.1±0.8‡

-Data were expressed as mean ± SD of 8 rats.
-When any dose-group was compared with its control, the difference was statistically significant:
(* P<0.003; **P<0.0003; *** P<0.0001;
† P<0.04; †† P<0.02; ‡ P<0.005).

Table3: Number of points overlying the fibromuscular stroma in prostate of adult rats treated with dietary melatonin (in unit area of 0.0025mm²).

Daily dose of melatonin in µg/kg body wt	***Points on prostatic stroma***
Control	15.1±1.3
125	15.6±0.9 **NS**
250	16.1±1.1 **NS**
500	17.2±1.2*
750	18.0±1.4**
1000	18.7±1.5**

-Data were expressed as mean ± SD of 8 rats.
-When any dose-group was compared with its control, the difference was statistically significant:
(* P<0.001; ** P<0.0009; NS= non significant).

Discussion

The prostatic weight was significantly affected by melatonin in the instant work. The explanation for this might be highlighted by the fact that prostatic weight principally follows its function status [7 & 8]. The physiological condition of the prostate determined basically by its glandular epithelial histological appearance; so it is considered to be actively functioning whenever its epithelial cells are tall columnar with pale large nuclei, whereas it is said to be insufficient in case its epithelia are atrophied, low cuboidal or squamous with dark relatively small nuclei [1, 7 & 8]. The glandular cells height and diameter of their nuclei showed a clear positive effect of melatonin on those parameters; i.e., they were steadily increased with the stepping up the doses up to the level of 500 µg/kg, then after decreased gradually. This could be due to the concept that melatonin is well designed to exert its physiologic action in a dose – dependent manner, being stimulating at normal therapeutic level and harmful at its overabundance [9 & 10]. The glandular cells significantly got taller with more and more larger nuclei observed in those groups treated with 125, 250 and 500 µg/kg dose, then regressed at 750 and 1000 µg/kg dose, these findings might indicate the inhancement in the function of epithelial glandular cells, as a consequence of exogenous melatonin on those cells, affecting them directly through melatonin receptors found in all tissues and cells [11], and/or indirectly through the hypothalamic-hypophysial-gonadal axis stimulating the secretion of FSH thereby promotes other sexual hormones secretion [12]. Nevertheless, there could be probably an induction of Sertoli cells by melatonin supplement to secret surplus amount of androgen binding

protein (Abp), which binds testosterone and hydroxytesteron produced out side the genital ducts, high concentration of these hormones are required within the genital epithelium and lumen for normal function [13 & 14]. The cell height and size of nuclei diminished with 750 µg/kg doses and a great regression noticed at 1000 µg/kg dose. The suggestion for those finding could be through suppression of hormone inhibin, which is secreted by Sertoil cells normally, inhibiting the secretion of FSH by the pituitary under control of hypothalamus and plays an important feed back role in controlling the secretion of sex hormones, which could be the cause of that regression consequently [1&14]. The significant effect on average diameter of nuclei in all of treated groups; may lead to the impression that melatonin could affect most of the cell activities, since the nucleus is the archive of the cell [13].

The buildup in prostatic weight might be the consequence of the raise in the cell height and nuclear bulk. The number of points overlying the stroma was increasing incrementally in all treated groups, which also could contribute to that enlargement of prostate. This large increment in septal thickness perhaps, due to the allowance in production of fibrocollagenous tissue; once there are any damaging events to any organ [7 & 14]. Moreover Melatonin hormone has special effect on fibroblasts, which are the active collagen – secreting cells and the basic forming cells of the connective tissues [1 & 15]. The other contributor to the prostate enlargement could be the dilated blood vessels, because melatonin has a well-known vasodilator action [16]. Those results could be explained by the fact that melatonin has damaging effects only when it is administered in excess [9 & 10].

The results of the instant work went with the concept that melatonin administration within a therapeutic dose might be helpful in the amelioration of the fertility state [3 & 17].

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