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# The Comparative Effects between Silver Nanoparticle, *Spirulina* Extract and Bicalutamide (Casodex®) On Experimentally Induced Prostatic Cancer in Rats

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**Abstract**

Over the past years, drug development direction has focused on the investigation of molecules that selectively target one protein. Executing rational combinations of targeted molecules will selectively regulate several pathways simultaneously. We aimed in this study to clear the relative therapeutic action between the outstanding agents silver nanoparticles (AgNPs), *Spirulina* (Sp), and a combination of them against bicalutamide (Casodex®) for the management of prostate cancer (PCa). PCa in rats was induced using bicalutamide and testosterone, followed by (7,12-dimethylbenz[a]anthracene). Rats were divided into six groups with 12 rats in each group. Group I was assigned as the control (co), group II as the PCa model, group III treated with AgNPs, group IV treated with Spirulina extract, group V treated with a combination of AgNPs plus Spirulina, and group VI treated with bicalutamide. Compared with bicalutamide treatment, AgNP treatment reduced the serum prostatic acid phosphatase (PAP) activity, improving PSA ratio, normalizing the IL-6 level, overcoming hormonal disturbance induced in PCa rats, and up-regulating P53, but it couldn't cure the pathological changes. *Spirulina* was significant up-regulated P53 and Caspase-3, in addition to regression of the histological pattern of high-grade prostatic intraepithelial neoplasia although the IL-6 level was still significantly high. Combination treatment decreased the PAP activity and up-regulating the expression of P53 and BAX, improving the pathological changes, increasing the E2 level and up-regulating the expression of BCL2 and TNF-α. Each of treatments has benefits and disadvantages over bicalutamide which need more experiments to discover the best combination, concentration, particle size and duration.

**Keywords:** Silver Nanoparticles, Bicalutamide, Spirulina extract, apoptosis, Prostatic Cancer

## Introduction

In men, the prostate gland consists of two cell types; secretory epithelial and stromal elements under androgenic influence. Many factors have been established to clarify the remarkable changes in cancer initiation and development that are connected to a history of this disease in the family, ageing and frequent genetic mutations. Dietary factors and lifestyle perform key functions in androgen hormone production which are regarded as mediators between molecular sites and external mediators related to the formation and spread of prostate cancer (**Sung *et al.*, 2020)**.

Silver nanoparticles are acting as potential anticancer materials. Unique physical, chemical, and biological features are present in nano-silver. It offers a new view for tumor prevention and treatment **(Firdhouse and Lalitha, 2015)**.

*Spirulina* (Sp) are filamentous, multicellular, and blue-green microalgae **(Kiran *et al.*, 2015).** Both in vivo and in a lab experiments have demonstrated that it helps to manage cancer. Some experiments used *Spirulina* itself, but others used its ingredients. Genetic damage protective effects of *Spirulina* were found overcoming the action of carcinogenic agents **(Chamorro *et al.*, 2002)**.

## Materials and Methods

### 1. Animals

In total, 72 male albino rats were involved in this experiment, weighing an average of 268 g (250 - 330 g). Animals were housed in separate metal cages supplied with fresh and clean tap water ad libitum. They were kept under constant nutritional and environmental conditions throughout the experimental period. The animals were housed for 15 days for acclimatization before starting the experiment. All animals were fed a frequently basal ration throughout the experiment as a standard pellet diet.

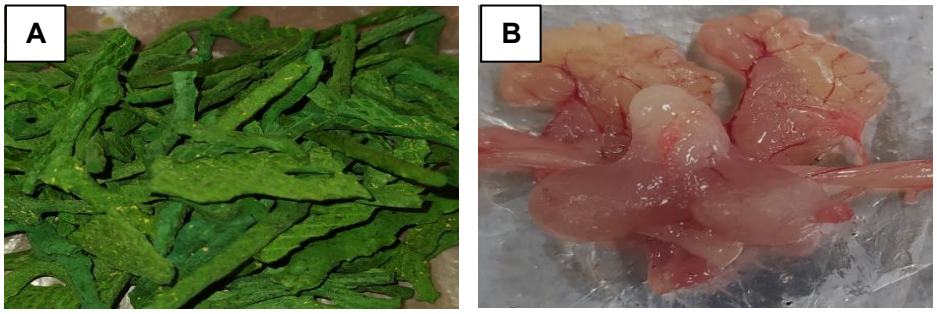


**Figure 1** A: photograph of rod shape of *Spirulina* algae, B: photograph of urogenital tract after dissection from PCa rat.

**2. Prostate cancer induction technique**

This experiment was required an initial 3 week treatment with bicalutamide (Casodex ®) tablet (15 mg/kg body weight) followed by treatment applying an androgen analogue, Testosterone propionate (Testonon) was given subcutaneously for three days at a dose of 100 mg/kg body weight. The following day after the third injection of testosterone, during the vegetative growth stage, DMBA was administered intraperitoneally at a dose of 30 mg/kg body weight. After that, testosterone hormone was injected subcutaneously at a dose of 2 mg per kg. The period extended for approximately three months **(Bosland *et al*. 1990; Quintar *et al*. 2017)**.

**3. Characterization of AgNPs**

Using a voltage of 200 kV and a magnification ratio of 25X, the transmission electronic microscopy (TEM) was applied in the microscopic class (EM-2100 High-Resolution-Japan model). Additionally to an electronic scanning microscope (SEM) (Japanese model: Jol 2000) **(Jannathul and Lalitha 2015)**.

**4. Design of the experiment**

72 rats in total were randomly assigned to six major groups, 12 rats per group. Group I control group (co), group II was PCa model, group III (AgNPs) AgNPs were administered orally to rats at a dose of 0.3 mg per kg b.w. group IV (Sp) spirulina extract was administered to rats at a dose of 4 gm per kg b.w. daily., group V (AgNPs/Sp) rats were given a combination of AgNPs + Spirulina extract, and group VI (a.a) rats were treated with bicalutamide (Casodex®) daily at a dose of 15 mg per kg b.w, summary are shown in table (1).

**Table 1**. summary of experiment design

| Phase | Shrinking | Regrowth | Initiation | Promotion | Treatments |
|---|---|---|---|---|---|
| **Time (days)** | 1-21 | 22-24 | 25 | 26-116 | 117-145 |
| **Control** | Non | pure corn oil Inj. Sc. | physiological saline i.p. inj. | pure corn oil Inj. Sc. | No treatment |
| **PCa** | Casodex 15 mg/kg | androgen analogue/pure corn oil - 100 mg/kg b.w. Inj. Sc. | DMBA/ physiological saline-30 mg/kg b.w. i.p. inj. | androgen analogue/pure corn oil - 2 mg/kg b.w. Inj. Sc. | No treatment |
| **AgNPs** | Casodex 15 mg/kg | androgen analogue/pure corn oil - 100 mg/kg b.w. Inj. Sc. | DMBA/ physiological saline-30 mg/kg b.w. i.p. inj. | androgen analogue/pure corn oil - 2 mg/kg b.w. Inj. Sc. | AgNPs 0.3 mg/kg b.w |
| **Spirulina** | Casodex 15 mg/kg | androgen analogue/pure corn oil - 100 mg/kg b.w. Inj. Sc. | DMBA/ physiological saline-30 mg/kg b.w. i.p. inj. | androgen analogue/pure corn oil - 2 mg/kg b.w. Inj. Sc. | Spirulina 4 g/kg b.w |
| **Combination** | Casodex 15 mg/kg | androgen analogue/pure corn oil - 100 mg/kg b.w. Inj. Sc. | DMBA/ physiological saline-30 mg/kg b.w. i.p. inj. | androgen analogue/pure corn oil - 2 mg/kg b.w. Inj. Sc. | AgNPs / spirulina |
| **Bicalutamide** | Casodex 15 mg/kg | androgen analogue/pure corn oil - 100 mg/kg b.w. Inj. Sc. | DMBA/ physiological saline-30 mg/kg b.w. i.p. inj. | androgen analogue/pure corn oil - 2 mg/kg b.w. Inj. Sc. | Casodex 15 mg/kg |

All rats were brutally murdered after 28 days on the day after the last treatment was administered. Blood specimens were obtained from the rat's eye's medial canthus was punctured by an ocular vein at early morning after overnight fasting. . The prostate gland was removed and divided into two pieces; the first piece was deeply frozen until genetic expression was estimated, and the second piece was fixed in 10% neutral buffered formalin before further histopathological investigation.

**5. Determination of Prostate acid phosphatase activity (PAP)**

The PAP activity was identified according to **(Seiler *et al.*, 1983)**.

**6. Prostatic Specific Antigen (PSA) estimation**

Serum total and free PSA levels were assessed by registered STAIA-PACK free PSA and STAIA-PACK PSAII test kit (TOSOH India Private Limited) to the manufacturer's specifications utilizing an automated immunoassay analyzer (TOSOH)

**7. Steroid Hormones Measurements**

The measurements of hormones used commercial ELISA kit to measure (trade: TSOH India Private Limited): STAIA-PACK-Testosterone, STAIA-PACK-iE2, STAIA-PACK-FSH, and STAIA-PACK-LH II, to the manufacturer's specifications and analyzer for automated immunoassays (TOSOH).

**8. Analysis of mRNA expression of prostatic tissues**

Total ribonucleic acid was extracted from the frozen prostate using RNeasy® Mini kit (Qiagen) according to the manufacturer's protocol **(Zhang and Gurunathan, 2016)**. The amount and calibre of ribonucleic acid were assessed using Spectrophotar Nano Drop. Using High Capacity complementary deoxyribonucleic acid Reverse Transcription Kit and the manufacturer's instructions, 1000 ng of total ribonucleic acid was converted into single stranded complementary deoxyribonucleic acid (Applied Bio systems). Real-time PCR was used to evaluate genes, and the sense and anti-sense primers are listed in table (2). **(Yuan and Gurunathan, 2017)**. Each individual sample underwent PCR reactions for each gene. An Applied Biosystem 7500 Fast Real time PCR Detection system was implemented to evaluate the reactions. Changes in gene expression were calculated by comparing the obtained cycle threshold (Ct) values to a reference (housekeeping) gene (GAPDH) **(Gasparino *et al.* 2014)**.

**Table 2** Gene sense and anti-sense primers for real time-PCR

| Gene name | Primer sequence |
|---|---|
| P53 | F: CTACTAAGGTCGTGAGACGCTGCC<br>R: TCAGCATACAGGTTTCCTTCCACC |
| BAX | F: CCAGGACGCATCCACCAAGAAGC<br>R: TGCCACACGGAAGAAGACCTCTCG |
| Caspase 3 | F: GTGGAACTGACGATGATATGGC<br>R: CGCAAAGTGACTGGATGAACC |
| TNF-α | F: AAATGGGCTCCCTCTCATCAGTTC<br>R: TCCGCTTGGTGGTTTGCTACGAC |
| BCL2 | F: TATATGGCCCCAGCATGCGA<br>R: GGGCAGGTTTGTCGACCTCA |
| GAPDH | F: TGCACCACCAACTGCTTAGC<br>R: GGCATGGACTGTGGTCATGAG |

**9. Analysis of IL-6 cytokine in serum**

The level of IL-6 was quantified using the ELISA technique (ELISA, Quantikine®) biomarkers for immunoassays. The test was carried out in guidance manufacturer's guidelines and Microplate reader Ryto, 2100c.

**10. Statistical analysis**

One-way ANOVA system was applied to determine the statistical significance of differences between groups. The Duncan's test was used as a post hoc test, microsoft SPSS (Version 25, SPSS Inc., Chicago, IL, USA). All values are displayed as the average ± standard error of the mean. The value of the significant difference was limited to probability of =< 0.05.

**RESULTS**

**1. Interpretations of the Scanning Electronic Microscope (SEM) and Transmission Electronic Microscope (TEM) images.**

Data in Fig 2 shows three dimensions SEM and two dimensions TEM images for AgNPs. The SEM photos show how AgNPs have a spherical shape, with uniform size and shape (fig 2 A). Also, the (fig 2B) illustrated a TEM image that shows spherical or subspherical particle. However, the prevailing one is spherical having about 65 nm size.

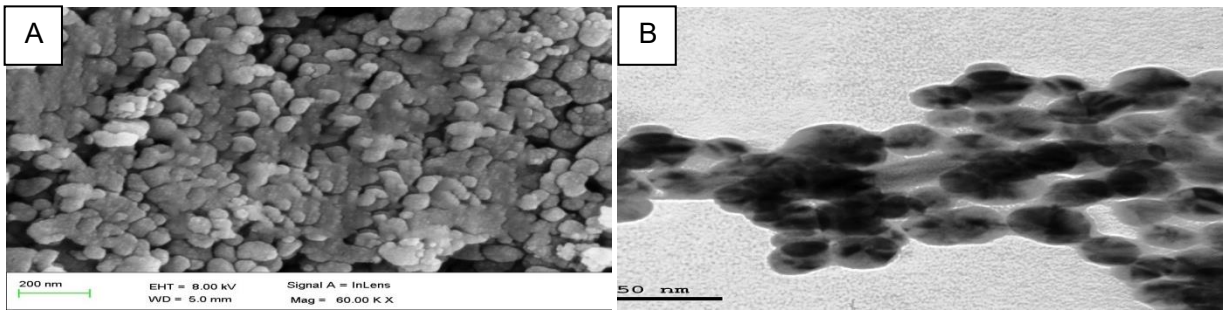


**Figure 2** AgNPs characterisation images, (A) SEM image, (B) TEM image

**2. The relative effects of silver nano-particles compared with bicalutamide treatment.**

The findings on serum biochemical parameters after treatment with AgNPs are described in Fig 3. Treatment with AgNPs didn't alter the total PSA and free PSA levels, however it still high compared with the bicalutamide group. AgNPs group showed significant decrease in RPSA and PAP compared with the bicalutamide group. The alterations in LH did not show any noticeable differences between AgNPs group and remaining experimental groups. AgNP significantly reduced the estradiol level relative to the bicalutamide group, which is on equal level with the PCa group's level. With PCa group, testosterone levels diminished. However, AgNPs might reverse this reduction and restore it to its normal level. The IL-6 level was restored to that of the control group by AgNP, however it was still noticeably elevated compared with bicalutamide group. All rats except control group underwent significant decrease in the gene expression of all studied genes. Compared with bicalutamide group, AgNPs could significantly up-regulated the expression of P53, Bax, and TNF-α genes.

**3. The relative effects of *Spirulina* compared with bicalutamide treatment.**

All of the Spirulina and bicalutamide groups had similar levels of free PSA, total PSA, RPSA, and PAP activity. The best effect of treatment on total and free PSA was achieved among Spirulina, combination and bicalutamide groups, while RSPA was achieved among Spirulina and bicalutamide groups. Spirulina or bicalutamide treatment normalised the E2 level in comparison to the control group and brought FSH levels back to normal levels. (Fig 4).

**4. The relative effects of AgNPs/Sp combination compared with bicalutamide treatment.**

The total and free PSA levels in the combination group are identical to those in the bicalutamide group, whereas RPSA and PAP activity significantly decreased. The level of FSH returned to normal after treatment with combination or bicalutamide, just like in the control group. Comparative to the bicalutamide group, the combination therapy considerably raised the E2 level. AgNPs/SP combination could significantly up-regulated the expression P53, BAX, BCL2, and TNF-α relative to bicalutamide group (fig 5).

**5. Histopathological findings**

The prostatic acini in the control group are normal as shown by the histological study in figure (6). Rats' prostate cancer has been demonstrated by photos showing the cross-section slide under a microscope of the PCa group in Fig (7). Fig 8 photos showing the cross-section slides under a microscope from the 4 treatment groups showed that high-grade prostatic intraepithelial neoplasia had regressed, and that normal prostatic acini had taken its place in varying proportions for each group. For the AgNPs, Spirulina, combination, and bicalutamide groups, respectively, the percentages are roughly 10, 75, 25, and 50%.

**Discussion**

When compared to bicalutamide treatment, silver nanoparticle therapy reduced PAP levels while having no effect on the PSA tumour marker. These outcomes coincide with those of (Sheida et al. 2021). On the other hand (Shahin and Mohamed 2017) found that Titanium dioxide NPs (TiO2 NPs) could significantly increase serum (PAP) activity. The effect of AgNP treatment on serum (PAP) activity is unique against all other treatments.

The findings demonstrated that AgNPs and bicalutamide both achieved the lowest FSH levels, demonstrating that their effects on FSH levels are equivalent. They all eliminated the pathologic elevation that was present along with a malignant condition. These outcomes supported **(Olugbodi *et al*., 2020)**. Others, however, observed non-significant differences between the treatment groups and the control group during the course of the experiment **(Garcia *et al*., 2014 and Mathias *et al*., 2015)**.

Changes in LH in the current investigation did not show any appreciable distinctions between the AgNPs group and the experimental bicalutamide group. This may be due to the fact that it is more challenging to ascertain their roles in the prostate

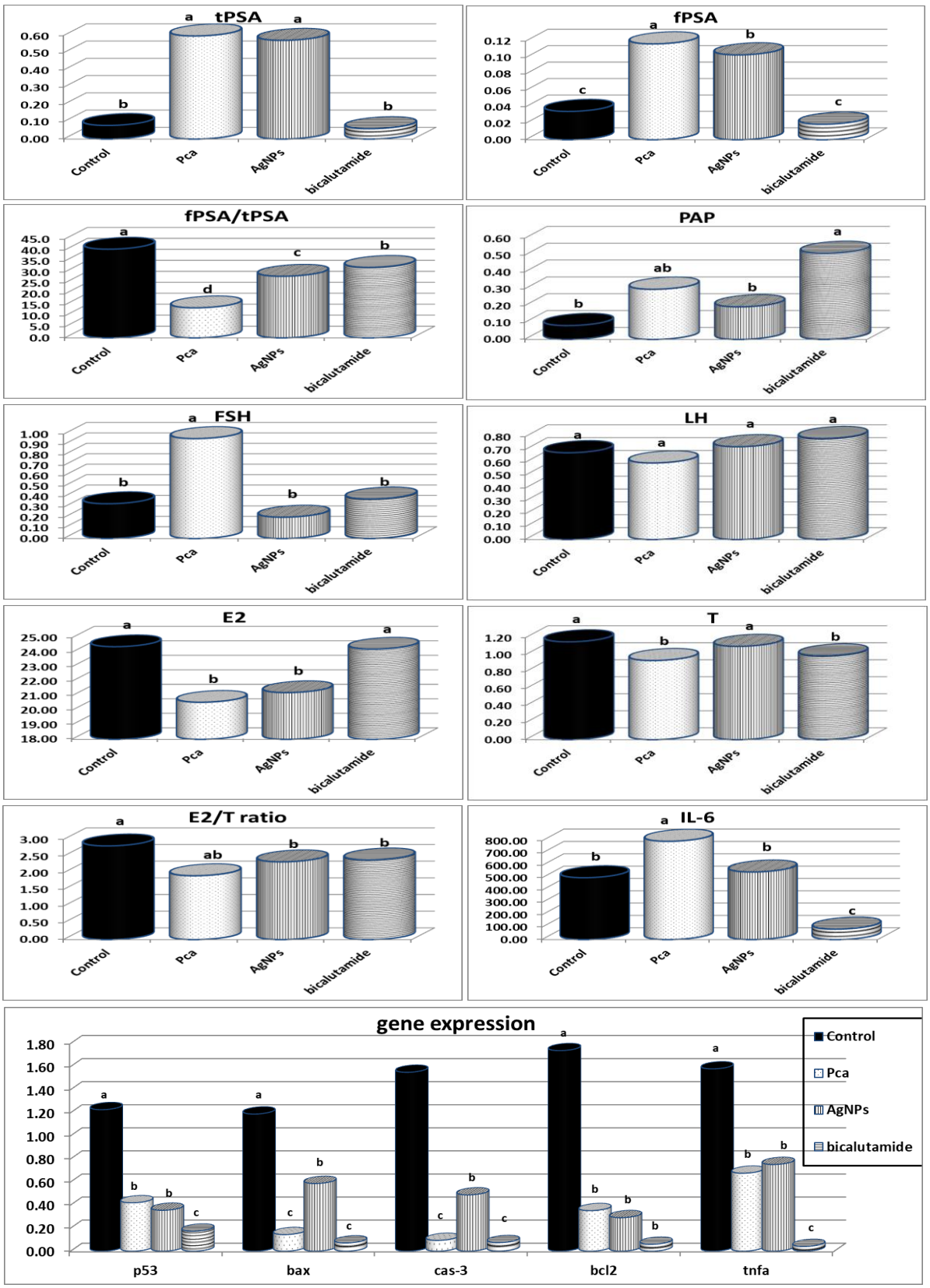


**Fig 3** The relative effects of silver nano-particles compared with bicalutamide treatment**,** PCa: prostate cancer, AgNPs: silver nano-particles, FSH: follicular stimulating hormone, LH: luteinizing hormone, E2: estradiol, T: testosterone tPSA: total prostatic specific antigen, fPSA: free prostatic specific antigen, PAP: prostatic acid phosphatase, IL-6: interleukine-6.

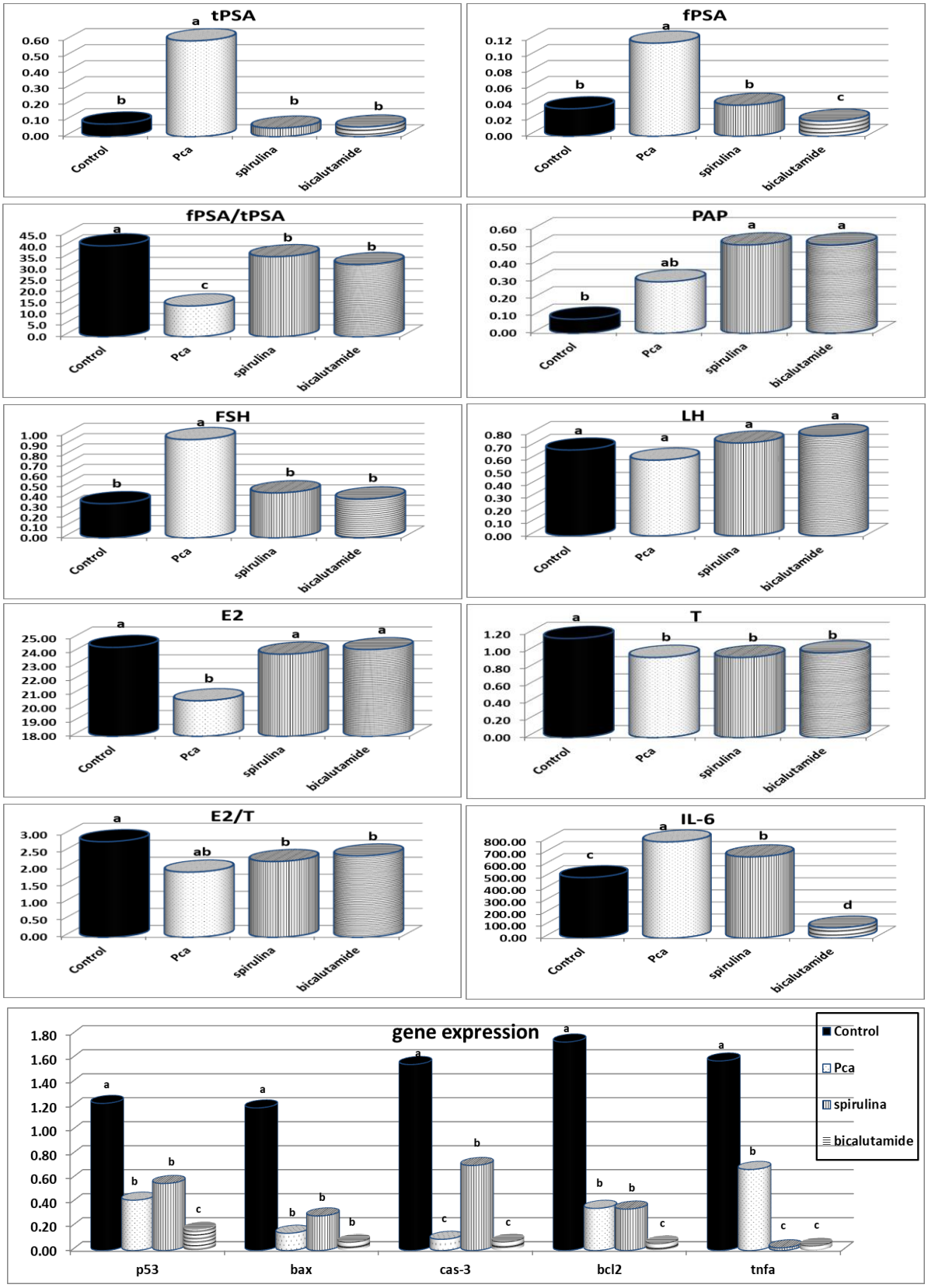


**Fig 4** The relative effects of spirulina compared with bicalutamide treatment**,** PCa: prostate cancer, Sp: *Spirulina*, FSH: follicular stimulating hormone, LH: luteinizing hormone, E2: estradiol, T: testosterone tPSA: total prostatic specific antigen, fPSA: free prostatic specific antigen, PAP: prostatic acid phosphatase, IL-6: interleukine-6.

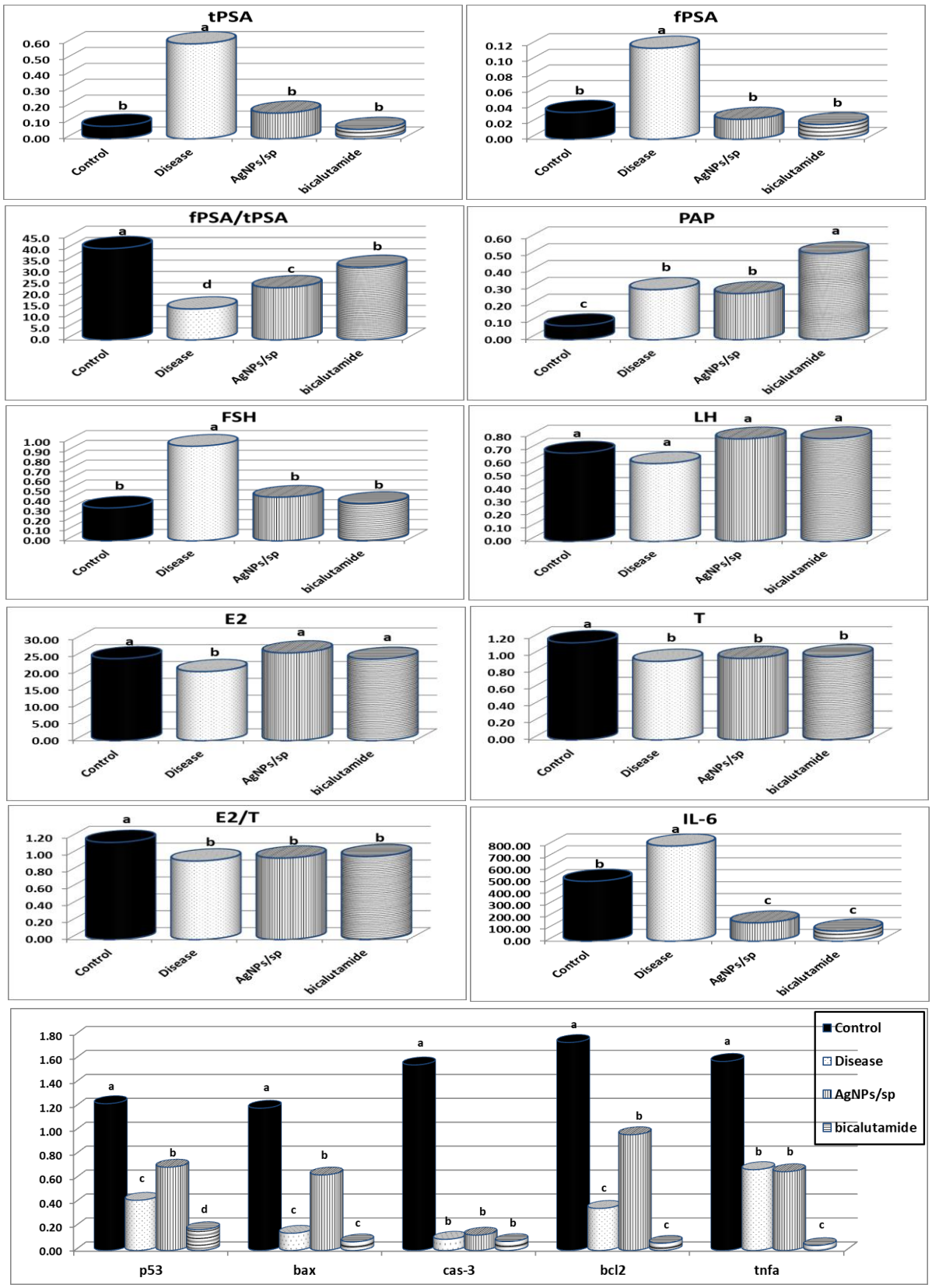


**Fig 5** The relative effects of AgNPs/spirulina combination compared with bicalutamide treatment**,** PCa: prostate cancer, AgNPs/Sp: Combining Silver Nanoparticles plus Spirulina (AgNPs/Sp), FSH: follicular stimulating hormone, LH: luteinizing hormone, E2: estradiol, T: testosterone tPSA: total prostatic specific antigen, fPSA: free prostatic specific antigen, PAP: prostatic acid phosphatase, IL-6: interleukine-6

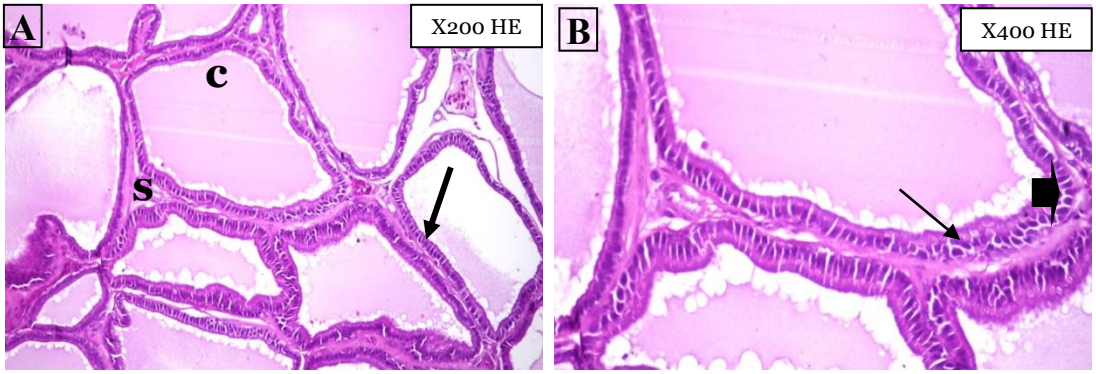


**Fig 6** prostate gland cross-section is photographed under a microscope for the negative control group. C letter: luminal prostate concretion, S letter: stand for thin smooth muscle interglandular fibre, (thick black arrow) for basal type of epithelial cells, (thin black arrow), for secretory type of normal epithelial cells, and (arrowhead) for thin intraglandular epithelia lining.

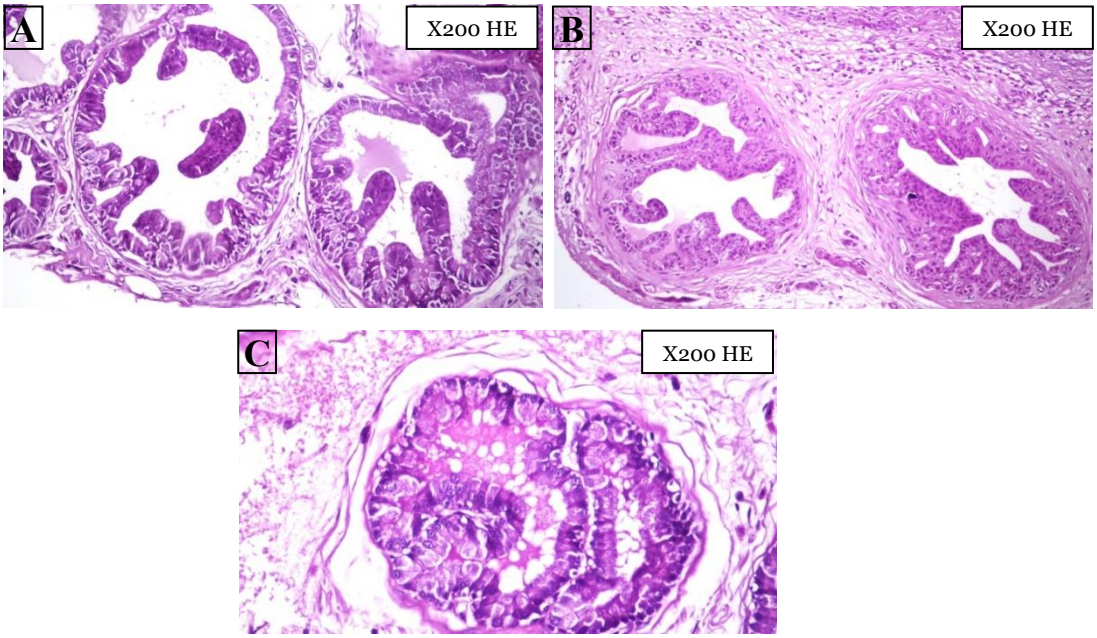


**Fig 7** The prostate gland cross-section photographed under a microscope for the prostate cancer group are shown in A slide as micropapillary, B slide as tufted, and C slide as cribriform, tufted (X200 HE)

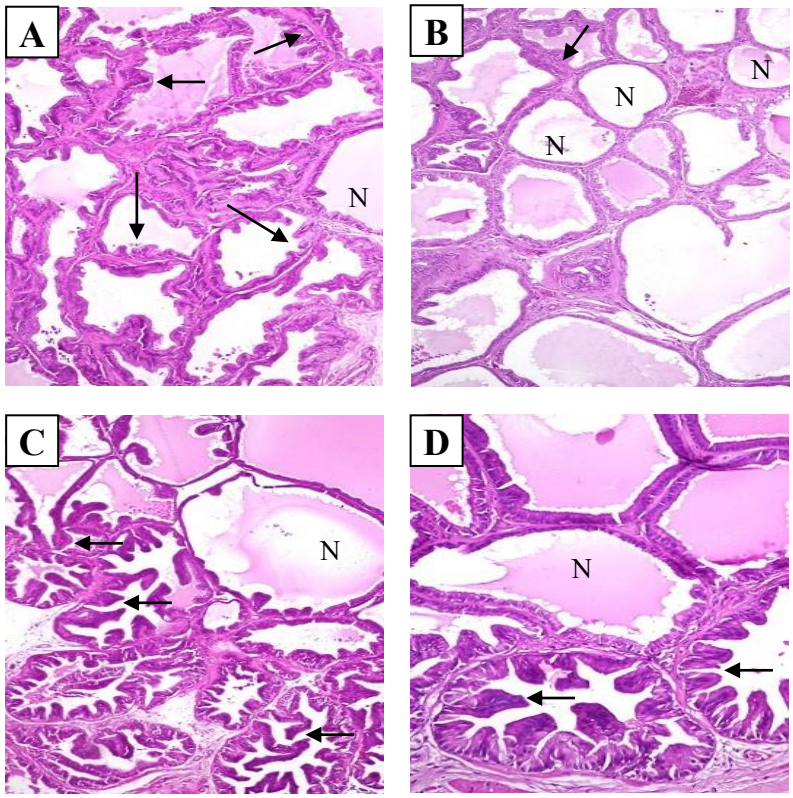


**Fig 8** Prostate gland cross-sections for those that received treatment, N normal prostatic acini, black arrows: high-grade prostate intraepithelial neoplasia. Slide (A) AgNPs group, Spirulina group slide (B), AgNPs/Sp group slide (C), and bicalutamide group slide (D) (X200 HE).

because of how their level is impacted by diurnal change and pulsatile secretion (Hsing and Comstock, 1993).

In contrast to the bicalutamide group, AgNPs considerably raise testosterone levels, and they ultimately equaled the control group's level, removing hormonal imbalance that was experimentally generated in these animals. When treating hormone-induced prostate enlargement with biogenic AgNPs, **(Ajayi *et al.* 2021)** achieved the same outcomes. Contrary, **(Mathias *et al.* 2015)** revealed that the testosterone profiles were not altered by AgNP treatment.

Comparing the AgNP regimen to the bicalutamide group—a group that was similar to the PCa group—the level of estradiol was dramatically reduced. Overall, AgNPs were more effective at correcting hormonal disruption brought on by PCa along the hypothalamic-pituitary-testis axis than bicalutamide, which has peripheral selectivity due to limited blood-brain barrier penetration **(Furr, 1996)**.

The IL-6 level was considerably higher compared to the bicalutamide group after AgNP treatment, however it had returned to the concentration seen in the negative control group. Similar findings were reported by **(Muniyappan and Nagarajan, 2014)**, who came to the conclusion that silver nanoparticles have significant anti-inflammatory and antioxidant action. By contrast, multiple pro-inflammatory gene mediators were discovered to rise in a dose-related manner by **(Ramadi *et al.*, 2016)**.

Compared with bicalutamide group, AgNPs could significantly up-regulated the expression of P53, Bax, and TNF-α genes. Merging these finding together, we can conclude that although silver nanoparticles could reduce the serum (PAP) activity, improving PSA ratio, regulating the hormonal disruption that PCa rats were exposed to, but it couldn't cure the pathological changes in PCa tissues relative to bicalutamide treatments.

The total PSA, free PSA, RPSA and PAP activity were the same in both the Spirulina and bicalutamide groups which is concurrent with (**McCarty *et al.*, 2014)**. It was observed that the PAP activity was increased over the PCa by both the Spirulina and the bicalutamide groups. Spirulina platensis, on the other hand, was found to significantly lower serum acid phosphatase activity in rats, according to **(Kumar *et al.*, 2009)**. Our work produced the opposite results because we utilized a dose that was ten times higher

(4 gm/kg) than what Kumar et al. used in their trial. Our results revealed that FSH, LH, testosterone, E2, and E2/T levels are non-significantly different between *Spirulina* and bicalutamide groups and it were restored to normal levels comparable to the bicalutamide group, which is approved by **(Ibrahim *et al.*, 2021)**. In comparison to results for AgNP treatment, the systemic hormonal imbalance persisted with Spirulina and bicalutamide regimens. The IL-6 concentration in the spirulina extract group was lower than that in the prostate cancer group, however it was still substantially greater than in the control and bicalutamide groups. In vivo experimental models support our findings **(Abdel-Daim *et al.*, 2015 and Romay *et al.*, 1999)**. However, several others reported moving in the opposite direction **(Tzachor *et al.*, 2021)**. This research showed that Spirulina had no impact on the release of IL-6 **(Chen *et al.*, 2014)**. The kind of model that was studied could be the root of these interactions. *Spirulina* was significant up-regulated P53 and Caspase-3 compared with bicalutamide group. The results gained for Caspase-3 concurrent with **(Ying *et al.*, 2015, Gantar *et al.*, 2017)**. Although each of *Spirulina* and bicalutamide treatments have a close biochemical results, there was a varieties in prostate tissues cure that is related to up-regulation of P53 and Caspase-3 genes and one can concluded that *Spirulina* alone has induced apoptosis by pathway far from inflammation cascade and involving genetic pathway.

While RPSA and PAP activity significantly decreased as compared to the bicalutamide treatment, the combination group has the same free and total PSA concentration as the bicalutamide group. Moreover none of treatments could normalize the PAP activity. The level of FSH returned to normal after treatment with combination or bicalutamide, just like in the control group. Comparative to the bicalutamide group, the combination treatment considerably raised the E2 level. The action of combination on tumor markers and sex hormones may due to the effect of silver nano-particles as described in details above. Combination group could decrease IL-6 level to level under all other groups except bicalutamide group which is lying at close level. Again, the action of combination treatment on IL-6 level may relate to the presence of silver nano-particles.

AgNPs/SP combination could significantly up-regulated the expression of P53, BAX, BCL2, and TNF-α relative to bicalutamide group. As AgNPs/SP combination could significantly up-regulated the expression of BCL2, and TNF-α, but bicalutamide treatment didn't, we can suppose that AgNPs/SP combination contains silver nano-particles which restore hormonal balance along the hypothalamic–pituitary–testis axis resulted in overcome the testosterone withdrawal action and up-regulated the expression of series of genes including BCL2, and TNF-α.

## Conclusion

Each of treatments have benefits and disadvantages over bicalutamide, AgNP treatment corrected the hormonal imbalance that PCa rats had developed, and up-regulating P53. Spirulina was significant up-regulated P53 and Caspase-3 in addition to regression of the high-grade prostatic intraepithelial neoplasia histological pattern. Combination treatment increased BAX and P53 expression while reducing PAP activity. So, we advise to use more than one modality of drug combinations that target different pathways (biochemical, genetic, and pathology) involved in PCa development and progression.

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## التأثيرات المقارنة بين جزيئات الفضة النانوية ومستخلص السبيرولينا ومادة البيكلوتاميد (كازودكس®) على سرطان البروستاتا المحدث تجريبيا فى الفئران

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ركز اتجاه تطوير العلاجات خلال السنوات الماضية على البحث عن الجزيئات التي تستهدف بروتينًا واحدًا بشكل انتقائي. ان اجراء استهداف مجموعات متعددة و مناسبة من الجزيئات الحيوية سيؤدى الى التحكم فى عدة مسارات بشكل انتقائي في وقت واحد. لقد هدفنا في هذه الدراسة إلى اكتشاف ومقارنة التأثير العلاجي النسبي للمواد المميزة كجزيئات الفضة النانوية و مستخلص طحالب السبيرولينا ، و معالجة مزدوجة منهما مقارنة مع مادة البيكالوتاميد (كازودكس®) للسيطرة على سرطان البروستاتا. تم احداث سرطان البروستاتا في الفئران باستخدام مادة بيكالوتاميد وهرمون تستوستيرون ، يليه حقن المادة المسرطنة (7،12–ثنائي ميثيل بنز [أ] أنثراسين). تم تقسيم التجربة إلى ست مجموعات ، 12 فأراً في كل مجموعة. تم تعيين المجموعة الأولى كعنصر ضابطة ، المجموعة الثانية هى المجموعة المريضة ، المجموعة الثالثة اعطيت جزيئات الفضة النانوية ، المجموعة الرابعة اعطيت مستخلص السبيرولينا ، المجموعة الخامسة اعطيت جزيئات الفضة النانوية بالإضافة إلى مستخلص السبيرولينا ، والمجموعة السادسة اعطيت مادة البيكالوتاميد. مقارنةً بالعلاج بمادة البيكالوتاميد فأن جزيئات الفضة النانوية قلل من نشاط انزيم فوسفاتاز الحمضى (PAP) وكذلك أدى إلى تحسين نسبة PSA ، واعادة مستوى IL–6 الى المستوى الطبيعى ، ولديه القدرة على التغلب على الاضطرابات الهرمونية التي حدثت في الفئران المصابة بسرطان البروستاتا ، وارتفاع التعبير الجينى P53 ، ولكنه لم يستطع علاج التغيرات المرضية الموجودة داخل انسجة البروستاتا. اما مستخلص السبيرولينا رفع التعبير الجينى لكلا من P53 و Caspase–3 بشكل كبير مقارنةً بالعلاج بمادة البيكالوتاميد ، بالإضافة إلى تقلص ظهور النمط النسيجي المرضى لسرطان البروستاتا على الرغم من أن مستوى IL–6 لا يزال مرتفعًا بشكل ملحوظ. بالنسبة للعلاج المزدوج من جزيئات الفضة النانوية و مستخلص طحالب السبيرولينا فأن نشاط انزيم فوسفاتاز الحمضى PAP انخفض. اما التعبير الجينى لكلا من P53 و BAX فقد ارتفع مقارنةً بالعلاج بمادة البيكالوتاميد ، مما أدى إلى تحسين التغيرات المرضية داخل انسجة البرؤستاتا. بالاضافة الى زيادة مستوى هرمون الاستروجين وارتفاع التعبير الجينى لكلا من BCL2 و TNF–α. اتضح من الدراسة ان كل من العلاجات المستخدمة لها نقاط قوة ونقاط ضعف مقارنةً بالعلاج بمادة البيكالوتاميد الامر الذى يتطلب اجراء مزيد من التجارب لاكتشاف أفضل تركيبة وانسب تركيز من كل مادة مع تحديد حجم الجسيمات النانوية المناسبة ومدة العلاج.

الكلمات الدالة: جزيئات الفضة النانوية, بيكالوتاميد, سبيرولينا, الموت المبرمج للخلايا, سرطان البروستاتا