Academic Journal of Cancer Research 16 (1): 18-30, 2023
ISSN 1995-8943

DOI: 10.5829/idosi.ajcr.2023.18.30

Antioxidant and Prostatic Protective Effect of Aqueous Extract of *Moringa oleifera* Seeds in Rats with Testosterone Induction of Benign Prostatic Hyperplasia

[1]*Jorge Luis Arroyo-Acevedo,* [1]*Yovani Martín Condorhuamán-Figueroa,*
[1]*Hugo Justil-Guerrero,* [1]*Rosa Elizabeth Carrera-Palao,* [2]*Claudio Jaime Flores-Flores,*
[2,3]*Lester Raymundo Domínguez-Huarcaya and* [4]*Jorge Alejandro Arroyo-Sandoval*

[1]Universidad Nacional Mayor de San Marcos, Lima, Perú
[2]Syneos Health del Peru
[3]Facultad de Farmacia y Bioquímica, Universidad Nacional Mayor de San Marcos, Lima, Perú
[4]Graduate in Statistics, Master in Biostatistics, San Juan Bautista Private University, Lima, Peru

Abstract: Objective: To demonstrate the antioxidant and prostatic protective effect of aqueous extract of *Moringa oleifera* (Moringa) seeds in rats with testosterone induction of benign prostatic hyperplasia (BPH). Methods: Qualitative phytochemical screening of the extract for secondary metabolites was performed. The experimental design considered 42 albino rats, randomly divided into six treatment groups (G1: saline solution, G2: enanthate 25mg/kg (ET), G3: ET + dutasteride 0.5 mg/kg, G4-G6: ET + Moringa (at 50, 250 and 500ug/mL). Treatments were administered orally for 28 days and ET intramuscularly on days 8, 14 and 21. Malondialdehyde (MDA, ug/mL), C-reactive protein (CRP, mg/dL), prostate specific antigen (PSA, ng/mL), prostate growth inhibition index and cellularity were determined. Average values were compared by analysis of variance considering a significance level of 5%. Results: Flavonoids, terpenoid saponins, etc. were observed. The CRP was 59.07% for dutasteride, in moringa from 39.07 to 53.33% (P<0.0001). MDA decreased by 51.15% with dutasteride, moringa from 46.95 to 56.87 (P<0.0001). Total PSA declined by 76.15% for animals treated with dutasteride and by 62.78 to 67.49% for those treated with moringa (P<0.0001). Percentage inhibition of prostate growth reached 66.27% for dutasteride group and 50.36 to 67.46 for moringa groups (P<0.0001). The treatments decreased cellularity compared to the ET group. Conclusion: Antioxidant and prostatic protective effect of aqueous extract of *Moringa oleifera* (Moringa) seeds has been demonstrated in rats with BPH.

Key words: Benign Prostatic Hyperplasia · Testosterone · *Moringa oleifera* Seeds · Albino Rats (Source: Mesh NLM)

INTRODUCTION

The worldwide prevalence of benign prostatic hyperplasia (BPH) reaches 40% in men older than 50 years and 90% for 90 years of age [1]. The prostate is in the posterior-inferior area of the pubic bone and has a volume of approximately 20 cc, but in patients with BPH, the prostate enlarges to more than 30 cc, making urination difficult, which would be associated with urinary retention infection [2].

Aging and androgens are established factors that lead to the occurrence of BPH, in addition, metabolic syndrome, genetics and lifestyle can also predispose to BPH, an increased risk of BPH has been found in those men with a history of prostatitis. Although there are controversies, epidemiological studies have revealed significant associations between prostatitis, BPH and prostate cancer risk. Lower urinary tract symptoms (LUTS) would occur and according to the International Incontinence Society (ICS), LUTS are divided into three groups: a) filling symptoms, b) emptying symptoms and c) postvoiding symptoms. In turn, one of the lower urinary tract symptoms that most frequently occurs in BPH and affects the patient's quality of life is [3, 4].

The medical treatment of BPH according to European and non-European guidelines includes pharmacotherapy,

Corresponding Author: Lester Raymundo Domínguez-Huarcaya, Syneos Health del Peru, Facultad de Farmacia y Bioquímica, Universidad Nacional Mayor de San Marcos, Lima, Perú.

lifestyle changes, surgery and phytotherapy, there are three groups of drugs for the treatment of BPH: alpha blockers (among them, alfuzosin, doxazosin, tamsulosin and silodosin), 5-alpha-reductase inhibitors (including finasteride and dutasteride [5].

Adverse drug reactions are of concern when prescribing to patients and specifically, those related to the pharmacological treatment of BPH, such as tamsulosin (α1 adrenergic antagonist that promotes relaxation of prostate smooth muscle). it has been characterized as an effective treatment for BPH [6, 7]. However, some studies show that epithelial cells remain proliferative even after drug administration and may be involved in the continued growth of the gland. In addition, tamsulosin can directly affect the ejaculation process, cognitive functions and mental health in men [8].

Mirabegron β3 adrenoceptor agonist is used to relieve the symptoms of overactive bladder (OAB) due to BPH, it induces blurred vision, dry mouth and constipation; and, in some patients, risk of increasing post-void residual urine volume [9]. The alpha-blockers doxazosin, tamsulosin, alfuzosin, terazosin and silodosin present: asthenia, dizziness and orthostatic hypotension, ejaculatory dysfunction (retrograde ejaculation, reduction of ejaculated seminal volume). Antimuscarinics can reduce detrusor muscle contractility such as oxybutynin, tolterodine, darifenacin and solifenacin, which have side effects such as dry mouth, constipation, urinary difficulties, nasopharyngitis, dizziness, confusion and restlessness. The 5-alpha-reductase inhibitors represented: finasteride and dutasteride; finasteride is a selective inhibitor of the type II isoenzyme and dutasteride, a non-selective inhibitor of type I and II isoenzymes, have relevant adverse effects such as decreased libido, erectile dysfunction and ejaculation disorders, gynecomastia. Combined alpha-blocker therapy with 5-alpha reductase inhibitors is an effective method, care must be taken in treatment lasting less than a year due to suspicion of intravesical obstruction and high residual urine volume [10].

In the pathophysiological evolution of BPH it occurs with processes of an inflammatory or infectious nature that affect the prostate gland; inflammation can independently affect the development of BPH [11], in different countries of the world and in Peru, there are medicinal plants that contain flavonoids, tannins, which give it an anti-inflammatory and antioxidant effect; This is the case of *Moringa oleifera*, whose leaves and roots have shown a greater antioxidant and anti-inflammatory effect than the seeds [12, 13].

The phyto-therapeutic treatment consists of the use of natural compounds (plant extracts) that contain flavonoids, tannins that confer antioxidant and anti-inflammatory properties with minimal side effects. In this study, the antioxidant and prostatic protective effect of *Moringa oleifera* seeds on BPH induced by testosterone in rats was evaluated.

MATERIALS AND METHODS

The Ethics Guide for experimental animals was considered [20]. For this, the present work is based on the 3R's: refinement, reduction, and replacement [21]. During the experimental process, international ethical principles for research with laboratory animals were respected. The rats were euthanized by intraperitoneal administration of pentobarbital (100 mg/kg), to achieve a quick and peaceful death. The research was submitted for evaluation by the Ethics Committee of the San Juan Bautista Private University, endorsing its approval with CERTIFICATE No. 1030-2021-CIEI-UPSJB, likewise, it had the approval of the Ethics Committee of the Faculty of Pharmacy and Biochemistry of the Universidad Nacional Mayor de San Marcos, giving its approval with Registry No. 008-CE-UDI-FFB-2020.

Preparation of Aqueous Extract of *Moringa oleifera* Seeds: The *Moringa oleifera* seeds came from crops in the Andean valley of the province of Huamanga, department of Ayacucho, having the certification and botanical identification by the Botanical Consultant CBP 3796 José Ricardo Campos de la Cruz. The seeds were subjected to a process of infusion, filtration and evaporation of water at 40°C, until a dry residue of constant weight was obtained, stored in a wide-mouthed, amber-colored glass bottle and kept in the freezer until later use.

Phytochemical Study of the Aqueous Extract of *Moringa oleifera* Seeds: For the detection of secondary metabolites, it was carried out using chemical characterization tests of the Draguendorf reaction, Shinoda reaction, foam test, ferric trichloride reaction, gelatin reaction, Lieberman-Burchard reaction, Ninhydrin reaction and reaction from Molish [14].

Experimental Design Study Population: Experimental study in animal model. The level of research is analytical or explanatory of a comparative type. The study sample consisted of 42 male albino rats, with a body weight of 205 ± 5 grams of body weight.

Experimental Groups: Forty-two male albino rats were used, acquired from the Universidad Agraria La Molina Bioterio, they were kept in an environment for seven days, in the Bioterio of the Faculty of Medicine of the UNMSM, with food and water at liberty, light/dark period. of 12 hours. They were randomly classified into seven each group (G): G1 Physiological saline solution (SSF) 2 mL/Kg; G2 Testosterone enanthate 25 mg/Kg (ET); G3 ET + Dutasteride (DU) 0.5 mg/Kg; G4 ET + moringa 50 mg/Kg; G5 ET + moringa 250 mg/Kg; G6 ET + moringa 500 mg/Kg.

Administration of Treatment and Induction of Prostatic Hyperplasia [15, 16]: Testosterone administration increases serum testosterone and PSA levels with subsequent enlargement of the prostate gland, confirming the induction of BPH.

Prior to the setting period, the animals were weighed and randomized into six groups of seven animals each. The Moringa solution was administered to groups 4, 5 and 6, every day at 9 am (except Sunday) for 28 days. In the same way the animals that received dutasteride. Groups 2-6 underwent BPH induction, administering testosterone enanthate (ET) intramuscularly at a dose of 25mg/Kg, on Days 8, 14 and 21 at 10 am, one hour after administering the treatments. The animals were observed daily, to determine any abnormal changes. On the last day of the experiment the rats were weighed; They were induced to anesthesia with ethyl ether, to obtain blood samples to later separate the serum and quantify MDA, PSA and PCR. They were then euthanized with pentobarbital 100 mg/Kg, as established by the protocol. Their prostates were removed. Each prostate was weighed; and the percentage of inhibition of prostate growth in rats treated for benign prostatic hyperplasia with testosterone was determined, considering the prostate weight data, the formula was considered: % Inhibition of prostate growth (%) = 100 - [(Treated group - Normal group) x 100) / (BPH group control - Normal group)].

Evaluation of the Antioxidant Effect of the Aqueous Extract of *Moringa oleifera* Seeds: The antioxidant effect of the aqueous extract of *Moringa oleifera* seeds was evaluated by quantifying malondialdehyde (MDA) and C-reactive protein (PCR), performed on blood samples taken from rats (treated and untreated) during the treatment period. last day of the administration of the treatments and this was prior to the sacrifice of the animals. The blood was centrifuged, the serum was separated and the MDA and PCR were determined.

Determination of Malondialdehyde (MDA), by the Buege and Aust Method [17]: Malondialdehyde (MDA) reacts with thiobarbituric acid (TBA) in an acid medium (trichloroacetic acid) forming a pink colored compound (and the coloration is related to the detection of oxidizing substances), where the absorbance is determined with a UV-VIS Genesys spectrophotometer at 535nm. 0.3 mL of serum was taken, mixed with 0.6 mL of 20% trichloroacetic acid, then the tube was capped and mixed, placed in a boiling water bath for 10 min, cooled and added 0.9 mL of 0.67 thiobarbituric acid. % in 0.25 N hydrochloric acid, mixed, immediately placed in a boiling water bath for 30 minutes, cooled with ice water, separating the precipitate by centrifugation at 3500 rpm for 10 minutes, the supernatant was collected with a Pasteur pipette, determined the absorbance at 535 nm against a blank containing all the reagents except the serum, the malondialdehyde concentration of the sample was calculated using the extinction coefficient of 1.56 x 105 M-1 cm^{-1}, multiplied by 6, the result was expressed in mol/L.

Determination of PCR, Method: Quantitative Immunochromatography [18]: Immunochromatography allows checking a complex of antigens that migrates on nitrocellulose paper to react specifically with the antibodies immobilized on the paper, giving bands colored by the accumulation of a conjugate. Nitrocellulose paper allows capillary migration of fluids and solutes while keeping fixed antibodies active. An easy reader computer has been used. Aliquots of serum from treated and untreated rats were obtained, for amplification a DNA Polymerase enzyme was used, the dNTPs, water for DNase and RNAse free PCR, Magnesium Chloride (MgCl2), the reaction tubes were subjected to various temperatures: DNA denaturation at 95°C for 30 seconds, hybridization of the first ones at 55°C for 30 seconds and an extension at 72°C for 30 seconds, these 3 temperatures were repeated 35 times and the final extension at 72°C for 5 minutes, the hybridization temperature of the first ones was standardized, MgCl2 concentration curves and DNA curves were performed to demonstrate the sensitivity of the technique, the amplification products were visualized in 12% polyacrylamide gels, stained with bromide. ethidium and photographed, the C-Reactive protein or PCR level was expressed in mg/dL.

Microscopic Analysis of the Prostate: For histological analysis, the prostates were removed and fixed in 10% formalin, processed and embedded in paraffin blocks. Sections were cut to 5 µm and will be stained with H&E

(hematoxylin and eosin). The sheets were examined under a light microscope (Olympus BX51), it was classified as: (-): normal cellularity; (+): increase of few cells; (++): regular increase of cells; (+++): increase in many cells [19].

Design of Data Collection: The treatment groups, as well as the data related to the antioxidant effect (MDA and PCR), prostate weight, prostate-specific antigen and the results of the microscopic analysis were recorded in the Excel sheet, which were later exported to the software. SPSS for processing and analysis.

Data Processing and Analysis: Statistical analyzes included exploratory evaluations, descriptive tabulations and data analysis. Prior to the data analysis, exploratory evaluations of the data were carried out to identify the shape of the data normal distribution, the presence of odd data and apply some transformation method to normalize the shape of the data distribution and correct the atypical data. Comparisons of study parameters between treatment groups were made by analysis of variance (ANOVA) using the F test, followed by multiple comparisons using Tukey's method. The assumption of homogeneity of variances was evaluated using Levene's test. All evaluation was performed at a significance level of 5%. The data were processed and analyzed using the SPSS version 21 program.

Ethical Aspects: The Ethics Guide for experimental animals was considered [20]. For this, the present work is based on the 3R's: refinement, reduction and replacement [21]. During the experimental process, international ethical principles for research with laboratory animals were respected. The rats were euthanized by intraperitoneal administration of pentobarbital (100 mg/kg), to achieve a quick and peaceful death. The research was submitted for evaluation by the Ethics Committee of the San Juan Bautista Private University, endorsing its approval with CERTIFICATE No. 1030-2021-CIEI-UPSJB, likewise, it had the approval of the Ethics Committee of the Faculty of Pharmacy and Biochemistry of the Universidad Nacional Mayor de San Marcos, giving its approval with Registry No. 008-CE-UDI-FFB-2020.

RESULTS AND DISCUSSION

Qualitative Phytochemical Study: The phytochemical study of the aqueous extract of *Moringa oleifera* seeds suggests the presence of secondary metabolites such as flavonoids, terpenoid saponins, among others (Table 1).

Evaluation of the Antioxidant: The mean of standard error was 308.6 (± 24.1) in the control group and in those who received dutasteride it was 126.3 (± 17.5) and in moringa at 50, 250 and 500mg/kg it was 143.1 (± 42.0), 188.0 (±19.7) and 144.0 (±17.3) ug/mL, respectively; which present a significant difference ($p < 0.001$); in which, the greatest reduction was 59% in the group that received dutasteride followed by the groups that received moringa 50 and 500 mg/kg that had a reduction greater than 53%, although at moringa 250mg/kg it was less than 40mg/kg.

On the other hand, the mean MDA was 3.7(±0.7) in the control group and in those who received dutasteride it was 1.8(±0.5) and in moringa at 50, 250 and 500mg/kg it was 1.8(±0.3), 2.0(±0.6) and 1.6 (±0.4) ug/mL, respectively; which present a significant difference ($p < 0.001$); in which, the MDA decreased in the treatments compared to the animals with BPH without treatment, reducing by 51.1% with dutasteride followed by moringa from 51.1, 46.9, to 56.8ug/mL at doses of 50, 250 and 500 mg/kg, respectively (Table 2-3, Figure 1-2 and Annex 1-3).

The Prostatic Protective Effect of the Aqueous Extract of *Moringa oleifera* (Moringa) Seeds: The mean PSA was 3.7(±0.4) in the control group and in those who received dutasteride it was 0.9(±0.1) and in moringa at 50, 250 and 500mg/kg it was 1.4(± 0.2), 1.2(±0.2) and 1.2(±0.2) ug/mL, respectively, which present a significant difference ($p < 0.001$). PSA reduction was 76.1% for dutasteride followed by 62.8 to 67.5% for moringa (Table 4, Figure 3, Annex 1-3).

Table 1: Identification of secondary metabolites in the extract in the aqueous extract of Moringa oleifera seeds.

Secondary metabolites	Identification reagent	Results
Alkaloids	Mayer	white precipitate (++)
	Wagner	brown precipitate (++)
	Dragendorff	(+++)
	Reineckato	Pink color (+)
Flavonoid	Shinoda	Red Color (++)
Anthraquinones	Borntrager's reaction	Red Color (+)
saponins	foam test	(+)
Terpenoids	copper acetate test	Green Color (+)

Note: (-) Coloration or precipitate is not evident; (+) Coloration or precipitate is little evident; (++) Coloration or precipitate is moderately evident; (+++) Coloration or precipitate is abundantly evident.

Table 2: Descriptive statistics of PCR and MDA in peripheral blood of rats treated for benign prostatic hyperplasia induced by testosterone.

				Moringa mg/kg			
	SSF 2 mL/kg	Testosterone 25 mg/kg	Dutasteride 0.5 mg/kg	50	250	500	
PCR (ug/mL)	Mean±SEM	308.57±	126.29	143.14±	188.00±	144.00±	P-value
MDA (ug/mL)	±	±	±	±	±	±	
CRP							
PSA							

Table 3: Assessment of Parameters of PCR, MDA.

					Confidence Interval 95%			
			Reduction*	Standar			Lower	Maximum
Indicator	Treatment	Lower value	(%)	Deviation	L. Lower	L. upper	Value	Value
PCR (ug/mL)	SSF 2 mL/kg	100.57	-	14.35	87.30	113.84	80	126
	Testosterone 25 mg/kg	308.57	-	24.10	286.28	330.86	270	340
	Dutasteride 0.5 mg/kg	126.29	59.07	17.46	110.14	142.43	100	150
	Moringa 50 mg/kg	143.14	53.61	42.04	104.26	182.02	100	228
	Moringa 250 mg/kg	188.00	39.07	19.71	169.77	206.23	150	213
	Moringa 500 mg/kg	144.00	53.33	17.28	128.02	159.98	120	170
MDA (ug/mL)	SSF 2 mL/kg	0.44	0.00	0.15	0.30	0.58	0.2	0.6
	Testosterone 25 mg/kg	3.74	0.00	0.71	3.08	4.40	2.9	5.0
	Dutasteride 0.5 mg/kg	1.83	51.15	0.46	1.40	2.26	1.0	2.5
	Moringa 50 mg/kg	1.80	51.91	0.34	1.49	2.11	1.4	2.3
	Moringa 250 mg/kg	1.99	46.95	0.60	1.43	2.54	1.0	3.0
	Moringa 500 mg/kg	1.61	56.87	0.43	1.22	2.01	1.0	2.3

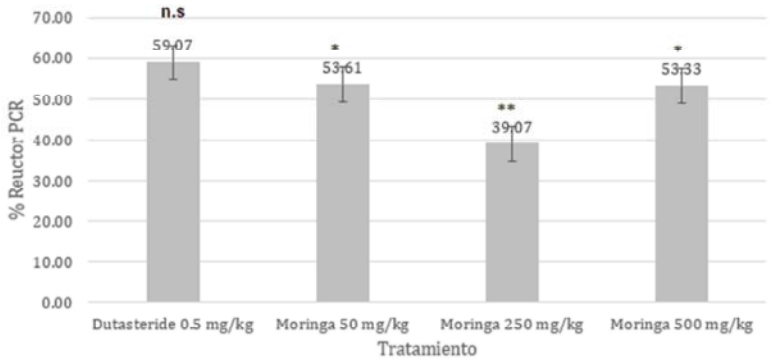


Fig. 1: Percentage reduction of the CRP level in peripheral blood of rats treated for benign prostatic hyperplasia induced by testosterone. Where: % PCR reduction= ((Control - Treatment) / (Control)*100) ns: Not significant; *=*P*< 0.0001

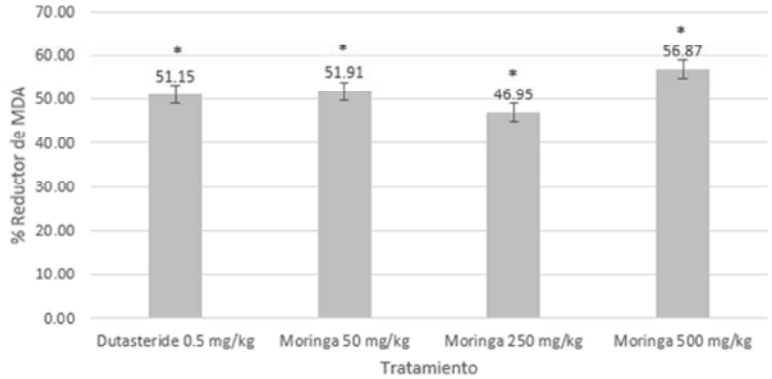


Fig. 2: Percentage reduction in the level of MDA in peripheral blood of rats treated for benign prostatic hyperplasia induced by testosterone. Where: % reduction of MDA=((Control-Treatment) / (Control*100). *=*P*<0.0001

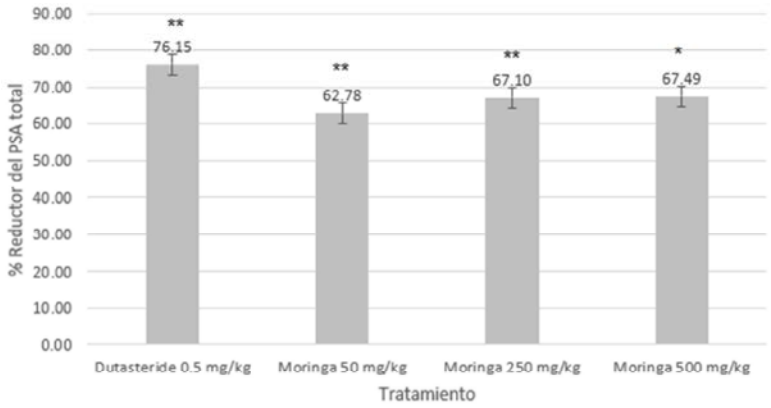


Fig. 3: Percentage reduction of the total PSA level in peripheral blood of rats with treatment for benign prostatic hyperplasia induced by testosterone *= $P< 0.001$; **= $P< 0.0001$

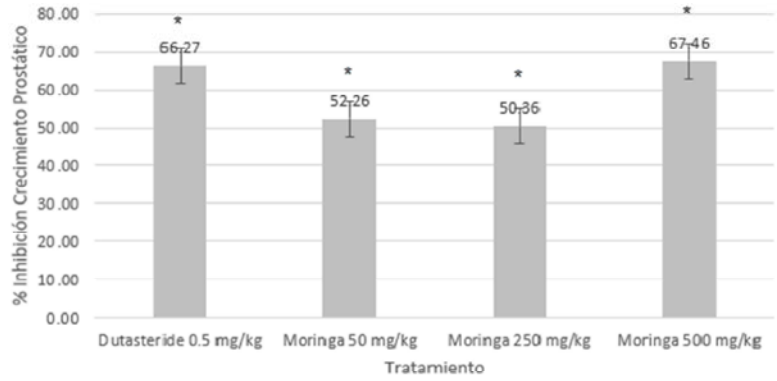


Fig. 4: Percentage inhibition of prostate growth in rats treated for benign prostatic hyperplasia with testosterone. Considering the prostate weight data from Table 4, the % inhibition of prostate growth (%) = (100 - ((Treated group -Normal group) x100) / (BPH group control - Normal group)). *= $P< 0.0001$

Table 4: Prostate weight and total PSA quantified in peripheral blood of rats treated for benign prostatic hyperplasia induced by testosterone.

					Confidence Interval 95%			
		Lower	Reduction*	Standar	----------		Lower	Maximum
Indicator	Treatment	value	(%)	Deviation	L. Lower	L. upper	Value	Value
Peso próstata	SSF 2 mL/kg	0.35	0.00	0.03	0.32	0.38	0.3	0.38
final (g)	Testosterona 25 mg/kg	0.95	0.00	0.05	0.90	1.00	0.88	1
	Dutasteride 0.5 mg/kg	0.55	36.95	0.05	0.51	0.59	0.5	0.6
	Moringa 50 mg/kg	0.64	28.08	0.05	0.59	0.68	0.58	0.7
	Moringa 250 mg/kg	0.65	26.88	0.05	0.60	0.70	0.56	0.71
	Moringa 500 mg/kg	0.54	37.71	0.05	0.50	0.59	0.5	0.6
PSA total	SSF 2 mL/kg	1.03	0.00	0.21	0.83	1.23	0.9	1.5
(ng/mL)	Testosterona 25 mg/kg	3.70	0.00	0.39	3.34	4.05	3	4.1
	Dutasteride 0.5 mg/kg	0.88	76.15	0.13	0.77	1.00	0.7	1
	Moringa 50 mg/kg	1.38	62.78	0.19	1.20	1.55	1.1	1.51
	Moringa 250 mg/kg	1.22	67.10	0.23	1.01	1.42	0.99	1.5
	Moringa 500 mg/kg	1.20	67.49	0.24	0.98	1.43	0.91	1.5

*: % reduction in prostate weight and total PSA = ((Control - Treatment) / (Control)*100).

On the other hand, the percentage of inhibition of prostate growth was 66.3% for dutasteride, while for moringa it was 50.4 to 67.5% (Table 4, Figure 4, and Annex 1-3).

Pathological Changes of the Prostate: When evaluating the anatomopathological changes of the prostate in rats with induction of benign prostatic hyperplasia by testosterone that received aqueous extract of *Moringa*

The arrows of the pathology must be typed on the picture
Please use numbering to each photo like A, B, C,
The comments under the image according to it number not beside it

I advise to magnify the images and save the images as PDF file and add from it your arros in different colors and the number of each inage
And collect all the comments underneath them

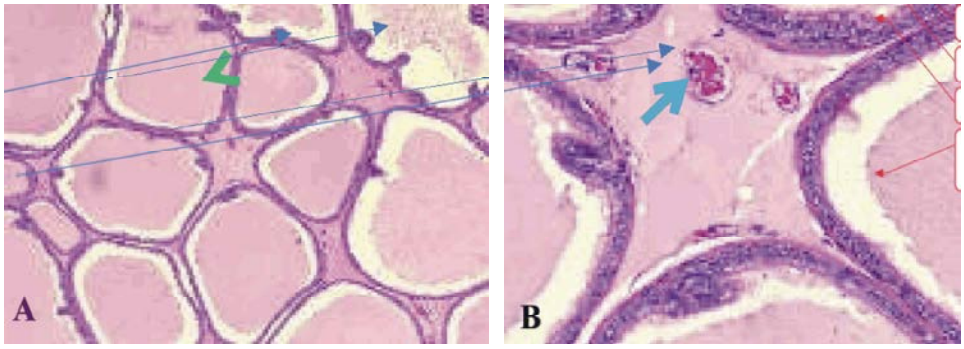
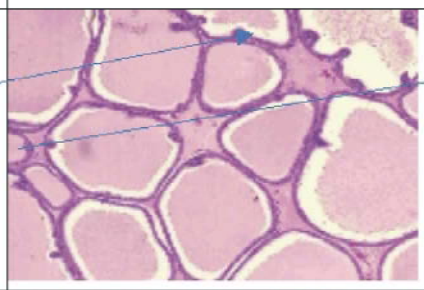
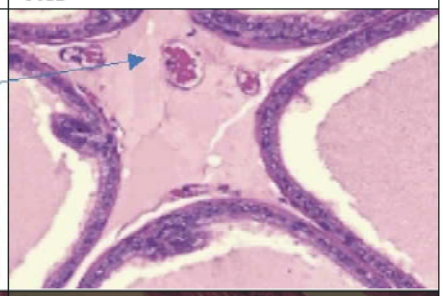
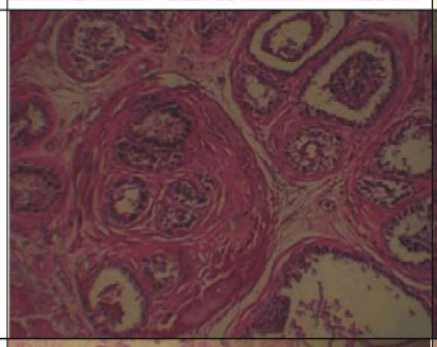
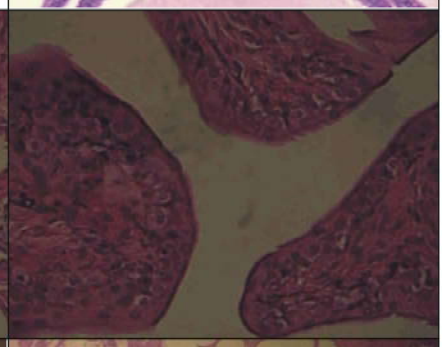
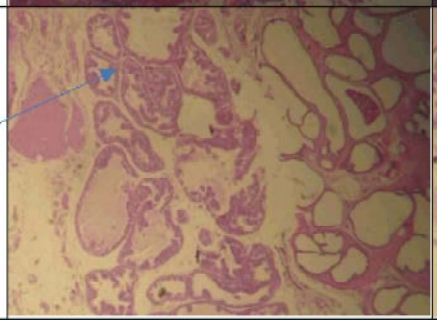
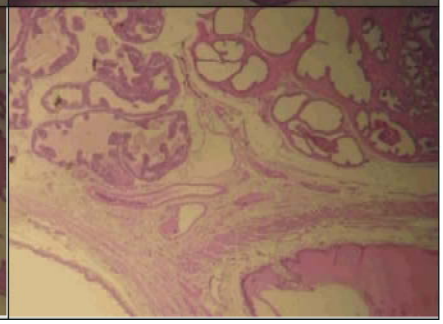


Fig. X: Shows normal prostate of SSF: Physiological saline solution 2 mL/Kg (A with 20x, Bwith 40x)

Treatment	20X	40X
SSF: Physiological saline solution 2 mL/Kg		
Enantato Testosterone 25 mg/Kg (ET) VIM. **Description**: adenomere hyperplasia papillary peripheral. Atypical papillary ductal urethral hyperplasia **Qualification**: HBP = +++		
ET + Dutasteride (DU) 0.5 mg/Kg Description: hyperplasia Papillary adenomeres partial Qualification: HBP = ++		

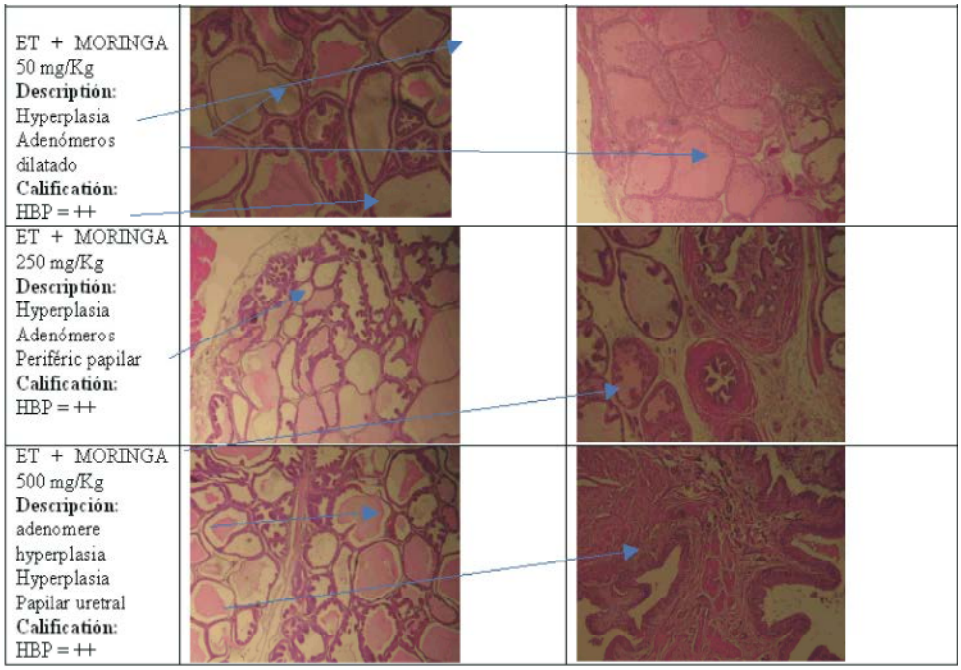


Fig. 5: Histopathological result in the study of BPH with treatment. Where: (-) = normal cellularity; (+) = increase in small number of cells; (++) = increase in regular number of cells; (+++) = increase in large number of cells.

oleifera (Moringa) seeds, it was observed: that in animals with physiological saline solution (SSF) cellularity was normal; the animals with Testosterone Enanthate 25 mg/Kg (ET) showed the presence of hyperplasia in adenomeres, peripheral papillary, atypical papillary ductal urethral hyperplasia, with an increase in a large amount of cellularity; those who received ET and dutasteride presented partial papillary adenomere hyperplasia, with an increase in the regular amount of cellularity; while, those who were administered ET and Moringa oleifera at the three dose levels, there was adenomere hyperplasia, urethral papillary hyperplasia, with a regular increase in the amount of cellularity.

This research aimed to demonstrate the antioxidant and prostatic protective effect of the aqueous extract of Moringa oleifera (Moringa) seeds in rats with induction of benign prostatic hyperplasia by testosterone, using the scientific method and the corresponding methodology, the main evidence has been achieved. secondary metabolites and the effects, the explanations of the findings are presented below; thus, as the contrasts found to the present are exposed.

Phytochemical screening was used, which is a technique used to detect secondary metabolites present in plant species, from the qualitative point of view, based on the application of chemical reactions with different reagents, where the appearance of a certain color or precipitate, colored or not, it is indicative of the presence of a certain metabolite [20-22].

When investigating through the qualitative phytochemical study the presence of the main secondary metabolites in the aqueous extract of Moringa oleifera (Moringa) seeds, the presence of Alkaloids compound followed by flavonoids, saponins, etc. has been observed. (Table 2). The presence of isothiocyanates, phenolic glycosides, flavonoids, fats, proteins and carbohydrates [23] is reported; GABA has been found in seeds and sprouts [24], niazirin, marumoside A and sitosterol have been isolated and identified. 3-O-b-D-glucoside [23] contains bioactive molecules that include lectins, proteins of non-immune origin; likewise, it contains non-hemolytic saponins [25]. Mass spectrometry has characterized phenolic compounds and glucosinolates, phenolics and glucosinolates [26]. In the aqueous extract of moringa seeds, the presence of alkaloids, anthraquinones, flavonoids, cardiac glycosides, terpenes (steroids), saponins [27] is reported.

Testosterone-induced benign prostatic hyperplasia (BPH) is related to the androgen receptor signaling pathway, to promote epithelial cell proliferation; testosterone by the action of 5α reductase is converted into dihydrotestosterone (DHT), this is a more potent androgen than testosterone to stabilize and activate the transcriptional activity of the androgen receptor (AR),

thus significantly increasing tissue BPH compared to the normal prostate, also increase PSA [28].

In Figure 1 and Table 3, the PCR reducing percentage was 59.07% for dutasteride followed by moringa from 39.07 to 53.33%. The MDA decreased in the treatments compared to the animals with BPH without treatment, reducing the MDA by 51.15% with dutasteride followed by moringa from 46.95 to 56.87 (Figure 2 and Table 3). When comparing the findings of MDA and PCR in peripheral blood of rats with induction of benign prostatic hyperplasia by testosterone and when administering the hydroalcoholic extract to 70% of the *Moringa oleifera* leaves, a 34.35% reduction in malondialdehyde was found, showing an increase in enzymes. antioxidant superoxide dismutase, catalase and reduced glutathione [29]. On the other hand, when evaluating the level of sexual hormones and biomarkers of oxidative stress in male rats treated with fractions of *Moringa oleifera* seeds, a reduction in the level of MDA was evidenced from 91.64% to 93.96% in doses of 50 to 100 mg/kg. [30, 31]. Also, that *Moringa oleifera* (MO) seeds improved oxidative and nitrosative vascular stress in spontaneously hypertensive rats (SHR); likewise, it positively regulated SODE, reduced circulating nitrites and C-reactive protein, associated with decreased expressions of iNOS and NF-κB proteins after treatment with MO[32]. Although it is true that in the present study the MDA and PCR are decreased, the difference observed with those of other authors is possibly due to the number of secondary metabolites, the species of experimental animal used and the environment.

In the present investigation it is shown that the aqueous extract of Moringa oleifera contains flavonoids and terpenoids in (Table 2), which would have contributed to the reduction of MDA. It is known that oxidative stress allows the circulation of inflammatory cytokines and the activation of the corresponding proinflammatory gene expression as cytokines; Also, the serum levels of TNF-, IL-6 and IL-1, leading to the presence of markers of oxidative stress such as malondialdehyde or MDA and flavonoids by inhibiting the secretion of cytokines, would be acting as anti-inflammatory agents and reducers of MDA [33]. It is also known that substances that induce oxidative stress do so by mitochondrial dysfunction and increased expression of mRNA, caspase-3 and Bax and by increasing MDA levels; and terpenoids as essential oil would be regulating mitochondrial alteration, leading to a decrease in malondialdehyde [34].

Genistein was found to inhibit the contractions of human prostate tissues caused by neurogenic, 1-adrenergic, endothelin-1 and U46619 stimuli [35, 36-43] effects of 1-adrenoceptor antagonists on EFS-induced and 1-adrenergic contractions of human prostate tissues were identical to what inhibitions with 50 M genistein found. The effects were still noticeable, but less strongly, at 10 M. Genistein also suppressed contractions brought on by U46619 and endothelin-1 to an extent comparable to that of 1-adrenergic contractions. Although they are resistant to 1-adrenergic antagonists, these non-adrenergic contractions have the potential to cause a full, maximal prostate smooth muscle tone [44-45].

Along with earlier research from epithelial cells, the present findings could, assuming appropriate amounts are present in the prostate, genistein could inhibit prostate development in BPH in vivo. In fact, genistein decreased ex vivo growth in prostate tissues from BPH patients as well as rat models of testosterone-induced BPH and high-fat-induced prostate growth [39-43, 46, 47].

Table 4 and Figure 3 show a total PSA reduction percentage of 76.15% for dutasteride followed by moringa from 62.78 to 67.49%. Observing a percentage of inhibition of prostate growth in 66.27% for dutasteride, while it was 50.36 to 67.46 in moringa (Figure 4 and Table 4). The inhibition of prostate growth reported by Ishola *et al*. [35], with 70% hydroalcoholic extract of Moringa oleífera leaves, reports an inhibition of 86.42 and 98.77 with doses of 50 and 100 mg/Kg, while the percentage of PSA reduction was 92.31 to 97.44% for the same doses.

As previously stated by Thongphichai *et al.* [42, 44]. Many of these compounds have no biological activity and do not reflect the efficacy of the plant extract. As a result, we wanted to see if the major alkaloid components have antiproliferative activity. Major alkaloid components significantly reduced the proliferation of TGF-treated WPMY-1 cells in the antiproliferative assay. The presence of major alkaloid components and their ratio, according to the findings, may improve the effectiveness of C. latifolium leaf extract in inhibiting TGF-induced WPMY-1 proliferation in BPH.

Androgens affect gene expression in various tissue and cell types by binding to androgen receptors (AR), dihydrotestosterone (DHT) has a higher affinity for AR than testosterone; in the prostate, the interaction between DHT and AR induces the synthesis of proteins, such as prostate-specific antigen (PSA). PSA, which is a glycoprotein in humans, is encoded by the kallikrein-related peptidase 3 (KLK3) gene and is secreted by prostatic epithelial cells; when PSA rises in the blood, it is used as a clinical marker for the prognosis of the disease. The hyperplasia of the stromal cells and the

prostatic epithelium would have as a mechanism an imbalance between cell proliferation and death, favoring cell proliferation and inhibiting apoptosis [36]. Sinomenine hydrochloride (SIN) is a sinomenine hydrochloride. Sinomenium acutum Rehderett Wilson is the main bioactive alkaloid isolated from the root of the traditional Chinese medicinal plant. In BPH-1 cells, SIN therapy significantly decreased Bcl-2 protein expression, significantly increased Bax protein expression and significantly decreased PCNA protein expression. These results indicate that SIN therapy inhibited the proliferation of BPH-1 cells through the apoptotic pathway. SIN therapy significantly reduced protein expression of Bcl-2 and PCNA in the PG tissues of mice with TP-induced BPH in vivo, indicating that SIN treatment alleviated BPH via the apoptotic pathway, which was consistent with in vitro data. These findings suggest that SIN treatment may improve BPH via the apoptotic pathway [43, 44].

Although there are controversies, epidemiological studies have revealed significant associations between prostatitis, BPH and prostate cancer risk. Also, there are studies expressing that BPH is related to chronic inflammation, overexpression of the COX-2 protein, an increase in the expression of iNOS that generates NO, would be situations that contribute to generating an inflammatory process, with a subsequent increase in inflammation markers such as malondialdehyde (MDA); there are reports that the BCL-2 and BCL-XL proteins, involved in regulating cell apoptosis, inhibit cell cycle progression; They are reduced in the epithelial cells of benign prostatic hyperplasia, all of which would also be involved in prostate enlargement, increased PSA. Finasteride does not act through BCL-2 family proteins [9, 10].

It is known that prostatitis can contribute to an increase in PSA, although there is disagreement about the effect of histological inflammation and the PSA level, but it is suggested that subclinical inflammation could affect PSA values [38]. Flavonoids are anti-inflammatory or antioxidant; they inhibit cell proliferation, migration and invasion; by avoiding or reducing oxidative stress, with fewer reactive oxygen species (ROS); the elimination of free radicals, inhibits the initiation of growth and prostate cancer; it may increase activation of the PI3K/Akt or RAF/MEK/ERK system and androgen receptor-mediated growth [39]. Youn *et al.* [47] presented their research on Chrysophanic acid (CA), a member of the anthraquinone family. CA had a significant suppressive effect on 5AR in the current study, implying that CA could be a 5ARI. CA has also decreased the levels of AR and its coactivating protein SRC1. Because 5AR is the initial cause of prostatic hyperplasia and AR is the main receptor involved, these findings suggest that CA may have a pharmaceutical potential in the treatment of BPH. In addition to downregulating 5AR and AR, CA treatment suppressed the level of ER more than the BPH group, suggesting that CA has another potential action mechanism besides the 5AR-AR axis. RWPE-1 cells are epithelial cells derived from the kidney.

The prostate is an immunocompetent organ equipped with an immune system that prevents autoimmune and inflammatory diseases. From the twelfth week of fetal life, the gland is populated with T lymphocytes, which develop the lymphoid tissue associated with the prostate. Within the prostate, >90% of the cells are T lymphocytes, particularly the CD8+ subtype, located at the prostatic epithelial and stromal level; there are also small numbers of B lymphocytes, macrophages and mast cells. In the peri glandular area of the prostate are cytotoxic T lymphocytes (CD8 +) and in the fibromuscular stroma there are B lymphocytes and T lymphocytes, mainly CD4 +. Epithelial and stromal cells express receptors for interleukins IL-1α, IL-1β, IL-6 and IL-18, which are positively regulated by inflammatory stimuli. The IL-6 cytokine is associated with the acute phase of inflammation and the transition from the acute phase to the chronic stage of inflammation; associated with a variety of chronic diseases, including inflammatory prostatitis, BPH and prostate cancer [39].

The dutasteride used in this investigation, as a standard drug, is a 5α-reductase inhibitor and thus reduces the level of DHT. Another therapeutic agent for BPH, α1-adrenoceptor antagonists (such as α-blockers) primarily target the smooth muscles of the bladder neck and prostate, resulting in relaxation of the smooth muscles and subsequent symptom relief [40, 41].

The histopathological study has shown a decrease in cellularity or a decrease in cell proliferation in the prostate gland, evidenced with dutasteride and treatment with the aqueous extract of *Moringa oleifera* seeds (Figure 5). Possibly these findings are explained because the function of the 5-alpha reductase enzyme and oxidative stress play an important role in the development of benign prostatic hyperplasia; oxidative stress is caused by a disruption in the balance between antioxidant capacity and reactive oxygen species (ROS); ROS increase active radicals and decrease the functionality of the immune system in the body; since testosterone increases the level of malondialdehyde (MDA), protein carbonyls

(PCO, a biomarker of oxidative stress); and decreased glutathione levels [30, 33].

The research exposed on the antioxidant and prostatic protective effect of *Moringa oleifera* seeds on benign prostatic hyperplasia induced by testosterone in rats, corroborates what has been reported by other researchers for this medicinal plant in its antioxidant and organ-protective properties such as the brain [30, 35].

CONCLUSION

The qualitative phytochemical study of the aqueous extract of *Moringa oleifera* (Moringa) seeds has revealed the presence of the main secondary metabolites such as flavonoids, terpenoids and saponins. In rats with testosterone-induced benign prostatic hyperplasia, an antioxidant effect has been determined by orally administering the aqueous extract of *Moringa oleifera* (Moringa) seeds. A prostatic protective effect of the aqueous extract of *Moringa oleifera* (Moringa) seeds has been demonstrated in rats with testosterone induction of benign prostatic hyperplasia. The prostatic protective effect of the aqueous extract of the seeds of *Moringa oleifera* (Moringa) has been confirmed with the anatomopathological evaluation of the prostate in rats with testosterone induction of benign prostatic hyperplasia. An antioxidant and prostatic protective effect of the aqueous extract of *Moringa oleifera* (Moringa) seeds has been demonstrated in rats with testosterone-induced benign prostatic hyperplasia.

It is recommended: Continue with a phytochemical study to identify and quantify secondary metabolites using spectrophotometric methods and mass chromatography with the aqueous extract of *Moringa oleifera* (Moringa) seeds. Evaluate the antioxidant effect using other biomarkers such as protein carbonyls (PCO), an indicator of oxidative stress; Enzyme levels such as glutathione, superoxide dismutase, in vivo with the aqueous extract of *Moringa oleifera* (Moringa) seeds. Carry out studies of the prostatic protective effect with the aqueous extract of *Moringa oleifera* (Moringa) seeds and their fractions: *In vivo* with other species, *in vitro* with the enigma prostatic hyperplastic epithelial cell line, or in silico. Carry out histopathological studies at the epithelial, stromal and muscular level; as well as immunohistochemical staining for IL10 and HSP-90, using the aqueous extract of *Moringa oleifera* (Moringa) seeds. Seek the antioxidant and prostatic protective effect using other types of extracts from the seeds and other parts of the *Moringa oleifera* (Moringa) plant.

REFERENCES

1. Bhat, S.A., S.A. Rather and N. Islam, 2022. An overview of benign prostatic hyperplasia and its appreciation in Greco-Arab (Unani) system of medicine. Asian Journal Urology, 9(2): 109-18.
2. Lerner, L.B., K.T. McVary, M.J. Barry, B.R. Bixler, P. Dahm and A.K. Das, 2021. Management of Lower Urinary Tract Symptoms Attributed to Benign Prostatic Hyperplasia: AUA GUIDELINE PART II-Surgical Evaluation and Treatment. Journal Urology, 206(4): 818-26.
3. Mobley, D., A. Feibus and N. Baum, 2015. Benign prostatic hyperplasia and urinary symptoms: Evaluation and treatment. Postgraduate Medicine, 127(3): 301-7.
4. Sasidharan, S., K.P. Srinivasakumar, S. Poddar, A. Bhaumik, S.K. Das and H.N. J, 2022. Benign Prostatic Hyperplasia (BPH): A Comprehensive Analysis of the Malaise and Summarizing Possible Management Options through Phytotherapeutic Agents. Kesmas J Kesehat Masy Nas., 17(2): 81-8.
5. Csikós, E., A. Horvαth, K. Ács, N. Papp, V.L. Balázs and M.S. Dolenc, 2021. Treatment of benign prostatic hyperplasia by natural drugs. Molecules., 26(23): 1-32.
6. Tamalunas, A., A. Wendt, F. Springer, A. Ciotkowska, B. Rutz and R. Wang, 2022. Inhibition of Human Prostate and Bladder Smooth Muscle Contraction, Vasoconstriction of Porcine Renal and Coronary Arteries and Growth-Related Functions of Prostate Stromal Cells by Presumed Small Molecule Gαq/11 Inhibitor, YM-254890. Frontier Physiology, 13(May): 1-20.
7. Huang, R., Y. Liu, A. Ciotkowska, A. Tamalunas, R. Waidelich and F. Strittmatter, 2021. Concentration-dependent alpha1-Adrenoceptor Antagonism and Inhibition of Neurogenic Smooth Muscle Contraction by Mirabegron in the Human Prostate. Frontier Pharmacology, 12(June): 1-20.
8. Shindel, A.W., S.E. Althof, S. Carrier, R. Chou, C.G. McMahon and J.P. Mulhall, 2022. Disorders of Ejaculation: An AUA/SMSNA Guideline. Journal Urology, 207(3): 504-12.
9. Ito, H., T. Matsuo, K. Mitsunari, K. Ohba and Y. Miyata, 2022. Impact of Mirabegron Administration on the Blood Pressure and Pulse Rate in Patients with Overactive Bladder. Medicine, 58(6).
10. Elterman, D., M. Aubé-Peterkin, H. Evans, H. Elmansy, M. Meskawi and K.C. Zorn, 2022 Canadian

Urological Association guideline on male lower urinary tract symptoms/benign prostatic hyperplasia (MLUTS/BPH). Canadaian Urology Associate Journal, 16(8).

11. Xie, J., Y.Y. Qian, Y. Yang, L.J. Peng, J.Y. Mao and M.R. Yang, 2022. Isothiocyanate From Moringa oleifera Seeds Inhibits the Growth and Migration of Renal Cancer Cells by Regulating the PTP1B-dependent Src/Ras/Raf/ERK Signaling Pathway. Frontier Cell Develovpent Biology, 9(January): 1-16.
12. Jaja-Chimedza, A., B.L. Graf, C. Simmler, Y. Kim, P. Kuhn and G.F. Pauli, 2017. Biochemical characterization and anti-inflammatory properties of an isothiocyanate-enriched moringa (*Moringa oleifera*) seed extract. PLoS One, 12(8): 1-21.
13. Yong-Bing, X., C. Gui-Lin and G. Ming-Quan, 2019. Antioxidant and anti-inflammatory activities of the crude extracts of moringa oleifera from kenya and their correlations with flavonoids. Antioxidants, 8(8).
14. Sudha, R., X.C. Philip and K. Suriyakumari, 2021. Phytochemical Constituents of Leaves of Moringa oleifera Grow in Cuddalore District, Tamil Nadu, India. Clinical Applied Health Science, 3(4): 164-7.
15. Nishino, T., T. Wedel, O. Schmitt, K. Bühlmeyer, M. Schönfelder and C. Hirtreiter, 2004. Androgen-dependent morphology of prostates and seminal vesicles in the Hershberger assay: Evaluation of immunohistochemical and morphometric parameters. Annal Anatomy, 186(3): 247-53.
16. Gonzales, C., J. Leiva-Revilla, J. Rubio, M. Gasco and G.F. Gonzales, 2012. Effect of red maca (Lepidium meyenii) on prostate zinc levels in rats with testosterone-induced prostatic hyperplasia. Andrologia, 44(SUPPL.1): 362-9.
17. Buege, J.A. and S.D. Aust, 1978. Biomembranes - Part C: Biological Oxidations. Methods Enzymol, 52: 302.
18. St. Sauver, J.L., A.V. Sarma, D.J. Jacobson, M.E. McGree, M.M. Lieber and C.J. Girman, 2009. Associations between C-reactive protein and benign prostatic hyperplasia/lower urinary tract symptom outcomes in a population-based cohort. American Journal Epidemioly, 169(11): 1281-90.
19. Li, J., Y. Tian, S. Guo, H. Gu, Q. Yuan and X. Xie, 2018. Testosterone-induced benign prostatic hyperplasia rat and dog as facile models to assess drugs targeting lower urinary tract symptoms. PLoS One, 13(1): 1-13.
20. MacArthur Clark, J.A. and D. Sun, 2020. Guidelines for the ethical review of laboratory animal welfare People's Republic of China National Standard GB/T 35892. Animal Model Experimental Medicine, 3(1): 103-13.
21. Lee, K.H., D.W. Lee and B.C. Kang, 2020. The 'R' principles in laboratory animal experiments. Laboratory Animal Respiratory, 36(1): 45-7.
22. Usman, A., M.U. Kawu, M. Shittu, A. Saleh and G. Bilbonga, 2022. Comparative effects of Moringa oleifera and ascorbic acid on biochemical changes induced by subchronic lead toxicity in male Wistar rats. Pharmacol Respiratiry - Module Chinese Medicine, 4(July): 100140.
23. Suganandam, K., A. Jeevalatha, C. Kandeepan, N. Kavitha, N. Senthilkumar and S. Sutha, 2022. Profile of Phytochemicals and GCMS Analysis of Bioactive Compounds in Natural Dried-Seed Removed Ripened Pods Methanolic Extracts of Moringa oleifera. Journal of Drug Delivery and Therapeutics, 12: 133-41.
24. Coello, K.E., J. Frias, C. Martínez-Villaluenga, M.E. Cartea, R. Abilleira and E. Peñas, 2020. Potential of germination in selected conditions to improve the nutritional and bioactive properties of moringa (*Moringa oleifera* L.). Foods, 9(11): 1-15.
25. Argungu, L., F. Umar, H. Jibrin and A. Hashim, 2022. Nutritional, phytochemical and biochemical composition of (*Moringa oleifera*) raw seed, seed cake and leaf meal for Aquaculture feeds. International Journal Aquac Fish Science, 8(2): 037-44.
26. Maldini, M., S.A. Maksoud, F. Natella, P. Montoro, G.L. Petretto and M. Foddai, 2014. Moringa oleifera: Study of phenolics and glucosinolates by mass spectrometry. Journal Mass Spectrometry, 49(9): 900-10.
27. Lugo-Rubio, J.Y., E.S. López-Álvarez, N. Vázquez-Montoya, R. Escamilla-Montes, J.A. Félix-Ortiz and E. Lugo-Medina, 2022. Extracts of Moringa oleifera and Croton californicus against infections of Vibrio parahaemolyticus (IPNGS16) in juvenile Pacific white shrimp (*Penaeus vannamei*). American Journal Aquat Respiratoy, 50(4): 541-52.
28. Kusuma Duarsa, G.W., Y.A. Sari, A.A. Gde Oka, K.B. Santosa, I.W. Yudiana and P.M. Wisnu Tirtayasa, 2021. Serum testosterone and prostate-specific antigen levels are major risk factors for prostatic volume increase among benign prostatic hyperplasia patients. Asian Journal Urology, 8(3): 289-97.
29. Ceci, R., M. Maldini, M.E. Olson, D. Crognale, K. Horner and I. Dimauro, 2022. Moringa oleifera Leaf Extract Protects C2C12 Myotubes against H2O2-Induced Oxidative Stress. Antioxidants, 11(8).

30. Yang, M., L. Tao, X.R.Kang, L.F. Li, C.C. Zhao and Z.L. Wang, 2022. Recent developments in Moringa oleifera Lam. polysaccharides: A review of the relationship between extraction methods, structural characteristics and functional activities. Food Chem X, 14(February): 100322.
31. Abd-Elnaby,Y.A., I.E. ElSayed, M.A. AbdEldaim, E.A. Badr, M.M.Abdelhafez and I. Elmadbouh, 2022. Anti-inflammatory and antioxidant effect of Moringa oleifera against bisphenol-A-induced hepatotoxicity. Egypt Liver Journal, 12(1).
32. Sailaja, B.S., R. Aita, S. Maledatu, D. Ribnicky, M.P. Verzi and I. Raskin, 2021. Moringa isothiocyanate-1 regulates Nrf2 and NF-κB pathway in response to LPS-driven sepsis and inflammation. PLoS One, 16(4 April), pp: 1-18.
33. Al-Khayri, J.M., G.R. Sahana, P. Nagella, B.V. Joseph, F.M. Alessa and M.Q. Al-Mssallem, 2022. Flavonoids as Potential Anti-Inflammatory Molecules: A Review. Molecules, 27(9).
34. Masyita, A., R. Mustika Sari, A. Dwi Astuti, B. Yasir, N. Rahma Rumata and Emran T. Bin, 2022. Terpenes and terpenoids as main bioactive compounds of essential oils, their roles in human health and potential application as natural food preservatives. Food Chem., 13Oct2021:100217.
35. Ishola, I.O., K.O. Yemitan, O.O. Afolayan, C.C. Anunobi and T.E. Durojaiye, 2018. Potential of Moringa oleifera in the Treatment of Benign Prostate Hyperplasia: Role of Antioxidant Defence Systems. Medicine Principla Practice, 27(1): 15-22.
36. Pistritto, G., D. Trisciuoglio, C. Ceci, Alessia Garufi and G. D'Orazi, 2016. Apoptosis as anticancer mechanism: Function and dysfunction of its modulators and targeted therapeutic strategies. Aging, 8(4): 603-619.
37. Zhang, L., Y. Wang, Z. Qin, X. Gao, Q. Xing and R. Li, 2020. Correlation between prostatitis, benign prostatic hyperplasia and prostate cancer: A systematic review and meta-analysis. Journal Cancer, 11(1): 177-89.
38. Kim, S.H., W.A. Kwon and J.Y. Joung, 2020. Impact of benign prostatic hyperplasia and/or prostatitis on the risk of prostate cancer in Korean patients. World Journal Mens Health, 38(2): 358-65.
39. Akbari, F., M. Azadbakht, K. Megha, A. Dashti, L. Vahedi and A. Barzegar Nejad, 2021. Evaluation of *Juniperus communis* L. seed extract on benign prostatic hyperplasia induced in male Wistar rats. African Journal Urology, 27(1).
40. Zhou, Z., Y. Cui, J. Wu and H. Jin, 2020. Efficacy and safety of dutasteride compared with finasteride in treating males with benign prostatic hyperplasia: A meta-analysis of randomized controlled trials. Experimental Therapie Medicine, 20(2): 1566-74.
41. Shah, J.P. and Z. Gil, 2009. Current concepts in management of oral cancer - Surgery. Oral Oncol., 45(4-5): 394-401.
42. Thongphichai, W., T. Uttarawichien, P. Chanvorachote, S. Pitiporn, T. Charoen-Ame, P. Kwankhao, P. Towiwat and S. Sukrong, 2022. Standardization of the ethanolic extract of Crinum latifolium leaves by two bioactive markers with antiproliferative activity against TGF-β-promoted prostate stromal cells (WPMY-1). BMC Complement Medicine Therapeutic, 22(1): 139.
43. Fan, M.S., Y.F. Xia, R.H. Ye, Z.R. Sun, M.Y. Wang, M.F. An, S.S. Zhang, L.J. Zhang, Y.L. Zhao, Z.M. Xiang and J. Sheng, 2023. Sinomenine Hydrochloride Can Ameliorate Benign Prostatic Hyperplasia by Lowering the 5α-Reductase 2 Level and Regulating the Balance between the Proliferation and Apoptosis of Cells. Molecules, 28(2): 803.
44. Lee, H., S.J. Moon, Y.D. Yoo, E.J. Jeong and J.R. Rho, 2022. Voratins A-C: Pyridinium Alkaloids from the Marine Dinoflagellate Effrenium voratum with Inhibitory Effects on Biomarkers for Benign Prostatic Hyperplasia. J. Prodduct, 85(6): 1495-1502.
45. Huang, R., Y. Liu, S. Hu, A. Tamalunas, R. Waidelich, F. Strittmatter, C.G. Stief and M. Hennenberg, 2022. Inhibition of α1-Adrenergic, Non-Adrenergic and Neurogenic Human Prostate Smooth Muscle Contraction and of Stromal Cell Growth by the Isoflavones Genistein and Daidzein. Nutrients, 14(23): 4943.
46. Yang, C.H., Y.C. Ou, C.C. Lin, Y.S. Lin, M.C. Tung, C.C. Yu, J.T. Lin and C.Y. Wen, 2022. Phloretin in Benign Prostate Hyperplasia and Prostate Cancer: A Contemporary Systematic Review. Life (Basel), 12(7): 1029.
47. Youn, D.H., J. Park, H.L. Kim, Y. Jung, J. Kang, M.Y. Jeong, G. Sethi, K. Seok Ahn and J.Y. Um, 2017. Chrysophanic acid reduces testosterone-induced benign prostatic hyperplasia in rats by suppressing 5α-reductase and extracellular signal-regulated kinase. Oncotarget, 8(6): 9500-9512.