# First report

## Effects of VigRX on Sexual Behavior, Erect Penile Size, Intracavernous Pressure, Testosterone Level, Sperm Density, and Some Organs in Male Sprague Dawley Rats and Its Long-Term Effects


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**Abstract**

Two dosages of VigRX (based on the human dosage used) i.e. 15 and 30 mg/kg/D were investigated by orally fed to the adult male Sprague Dawley rats for 14 consecutive days compared with the control group receiving 1 ml. of distilled water / D.

The results were found that the body weight gained of VigRX - treated groups did not significantly differ from the control group. VigRX especially at the dosage of 30 mg/kg/D could significantly increase sex drive , erect penile size , intracavernous pressure (ICP) and sperm density in the rats. Blood testosterone level of the VigRX treated groups and control group are not statistically different. The body weight and various organs weight i.e. penis , testes , epididymis , seminal vesicle , prostate gland , adrenal gland , spleen and pituitary gland of the VigRX - treated groups did not significantly differ from the control group except the liver and kidney weight of the VigRX - treated group significantly less than the control group.

It was concluded that the VigRX at the dosage of 30 mg/kg/D could induce the high sex drive, the penile size , the penile erection and could increase the sperm density in the rat whereas VigRX had no effect on the various organs weight but had some effects on liver and kidney by decreasing the weight of both organs significantly.

## Introduction

Some adult men wish to maintain or enhance erectile function, size of erect penis size, and level of sex drive. It is reported that some medicinal plants improve sex performance, such as *Butea superba* (Anusarnsunthorn, 1931). Research on *Butea superba* extract has revealed that it can induce erect penis size and erection in the rat by increasing intracavernous pressure (Smitasiri et al, 2003; Tocharus et al, 2002), and significantly increase the sperm density in the rat (Smitasiri et al, 2003), without affecting prostate gland size or weight (Kheowmung et al, 1992).

The rat is a suitable model for the study of erect penis size erection (Quinlan et al, 1989), and can be used to study the effects of medicinal plants on sexual behavior (Ang and Ngai, 2001; Carro – Juarez et al , 2004 ; Gauthaman and Adaikan, 2005 ; Islam et al, 1991; Ramachandran et al, 2004 ; Tajuddin et all, 2004 ).

There are many kinds of medicinal plant which have been reported for their aphrodisiac effects such as Yohimbe bark, *Muira puama* root , Catuaba bark(Antunes et al, 2001), Damiana leaf , *Tribulus terrestris* (Gauthaman and Adaikan, 2005) , *Syzygium aromaticum*(Tajuddin et all, 2004), *Montanoa tomentosa* (Carro – Juarez et al , 2004 ) , *Butea frondosa* (Ramachandran et al, 2004) , *Passiflora incarnata* ( Dhawan et al , 2003) and *Eurycoma longifloria* (Ang and Ngai, 2001). Some medicinal plants are also noted for abilities to enhance mental alertness and stimulate circulation, such as Ginkgo leaf; improve blood flow, such as Hawthorn berry; achieve anti-oxidant effect, such as Ginkgo leaf and Panax Ginseng root; stimulate the immune system, such as Panax Ginseng root; and have tonic effect, such as Saw Palmetto berry, Panax Ginseng root, and Catuaba bark.

This study is to investigate whether and to what extent an herbal product that contains not just one but various botanical ingredients can improve erectile function,

erect penile size, and sex drive. VigRX is a product that contains Asian ginseng root, Saw palmetto berry, *Gingko biloba* leaf, Hawthorn berry, Muira pauma bark extract, Catuaba bark extract, Cuscuta seed extract, and Epimedium leaf extract. It is claimed that this product may induce these effects.

**Aim of the project**

The aim of this project firstly is to investigate the effects of VigRX on the sexual behavior, erect penile size, intracavernous pressure, testosterone level, sperm density, and related reproductive and other organ changes, in adult male Sprague-Dawley rats;

**Materials and Methods**

1.Preparation of VigRX for testing in rats.

1.1 VigRX bottles from U.S. are sent to our laboratory.

1.2 VigRX will be mixed with distilled water and divided into 2 dosages - i.e. 15 mg , 30 mg. VigRX/kg /rat/day (Calculate from the recommended dose of VigRX for human: 2 and 4 capsules/70kg/day).

2. The laboratory rats

Adult male and female rats

The adult male and female rats of Sprague Dawley strain will be used in this study. The body weight of adult male and female rats are 250-280 grams and 200-240 grams, respectively, to assay sexual behavior, erect penile size, intracavernous pressure, testosterone level, sperm density and some organs in rats treated with VigRX.

**Note:** All of the rats will be supplied by the National Laboratory Animal Center, Mahidol University and transferred to Mae Fah Luang University by air.

3. Rearing of the laboratory rats

All of the laboratory rats are reared in the Animal House of Mae Fah Luang University at the ratio of one rat per cage (stainless steel cage, 8x8x12 inches), in a temperature-controlled room approximately 24±1 degrees Centigrade in a room with proper ventilation and a daily light cycle of 12 h light and 12 h dark. All of the rats are fed with rodent feed #082 (Pokkapan Animal Feed Co. Ltd.). During the whole period of the study, sufficient water is provided to these rats *ad libitum*.

4. Rat groupings

Adult male and female rats

All rats are allowed to recover in the Animal House for 6 days, then each rat is weighed and the data recorded.

The rats are divided into 3 groups (10 rats/group).

**Group 1** receives 1 ml of distilled water administered orally/day for 14 consecutive days.

**Group 2-3** Repeat the same as the control group but receive VigRX at the dosage of 15 and 30 mg/ kg/rat/day respectively for 14 consecutive days.

Each rat will be weighed every 3 days until the test is completed on Day 14. On Day 14, between 7.00 – 9.00 p.m., the sexual behavior of each adult male rat is observed by using the in-heat female rats (also resident in our facility since the start of the study on sexual behavior) which are induced into heat by injecting subcutaneously with 600 mcg./ 0.2 ml of estradiol benzoate mixed with olive oil at 72 hours before observation. Then at 6 hours before observation, 600 mcg./ 0.2 ml of progesterone mixed with olive oil is subcutaneously injected to the female rats. This is the Islam et al (1991) method to induce female rats into heat.

Each of the adult male rats in each group is placed into a 12x18x15 inch glass cupboard and a dim light is turned on for 5 minutes prior to the start of observation. After that, the female rats are placed into the glass cupboard using the ratio of male:female (1:1). The observation on the sexual behavior of the male rat in each cupboard is recorded by VDO recorder to record the male rat behavior for 20 minutes. After that, the VDO is played and the latency and frequency of mount , intromission and ejaculation of the male rats are recorded. The mean standard deviation (±S.D.) and statistical analysis will be calculated by ANOVA and LSD.

When the observation of the male rat's behavior is finished, the adult male rat is moved back into the same cage. The next morning (Day 15), after weighing the rat, penile size is measured while erect using the method of Pinmongkholgul (2001). Then the rat is anaesthetised with nembutal injected intraperitoneally. When the rat is completely anaesthetised, the intracavernosal pressure is recorded by the Maclab instrument using the method described by Tocharus et al (2002). After recording of the intracavernosal pressure is completed, the blood is collected via cardiac puncture for measuring the testosterone level by the electrochemical luminescence(ECL) method. The testes, epididymis, prostate gland, seminal vesicle, pituitary gland, adrenal gland, liver, kidney and spleen will also be removed and weighed using 4-digital electronic balance (Mettler: Toledo AB 204-S).

According to the epididymis, and after weighing, the cauda epididymis is removed and sperm density is counted using the method as described by Pinmongkholgul (2001). All of the collected data will be statistically analyzed using ANOVA and LSD.

**Results**

There was no significant difference between the body weight gained of the VigRX- treated groups and the control group (Fig.1)

The male rats fed with VigRX especially at the dosage of 30 mg/kg/D for 14 consecutive days could significantly have the intromission latency and the ejaculation latency less than the control group (Table 1) whereas the mounting frequency , the intromission frequency and the ejaculation frequency more than the control group significantly (Table 1 , Fig. 2-4) .

Investigation on the effects of both dosages of VigRX on the erect penile size of the rats revealed that VigRX 30 mg /kg/D could significantly increase the width and length of the erect penis when compared with the control group whereas the VigRX 15 mg/kg/D could significantly increase only the length of the erect penis (Tabel 2 , Fig. 5-7).

The electrical stimulation of cavernous nerve of the control group could induce penile erection with the intracavernosal pressure of 49.70 $\pm$ 2.49 mmHg but both dosages of VigRX could significantly increase the intracavernosal pressure more than the control group i.e. 70.68 $\pm$ 3.66 mmHg and 83.56 $\pm$ 5.33 mmHg respectively (Table 3, Fig. 8).

Investigation on the effects of VigRX on sperm density and testosterone level in the rats revealed that only VigRX 30 mg/kg/D could significantly increase the sperm density . There was no significant difference between the testosterone level of both dosages of VigRX and the control group (Table 4)

The body weight and various organs weight i.e. penis , testes , epididymis , seminal vesicle , prostate gland , adrenal gland , spleen , pituitary gland of the both dosages of VigRX - treated groups did not significantly differ from the control group (Table 5) but the liver weight of both dosages of VigRX could significantly

different from the control group by decreasing the weight (Table 5) whereas the kidney weight of only VigRX 15 mg/kg/D could significantly different from the control group by decreasing the weight also (Table 5).

Table 1 Comparison between the mounting , intromission & ejaculation latency and frequency of the VigRX - treated groups and the control group for 30 minutes on Day 14

| Group | No. of rats | Latency (minutes) | | | Frequency (Times) | | |
|---|---|---|---|---|---|---|---|
| | | Mount | Intromission | Ejaculation | Mounting | Intromission | Ejaculation |
| Control | 10 | 1.13 ± 0.61 | 5.76 ± 2.66 | 15.90 ± 5.35 | 34.80 ± 8.47 | 19.00 ± 8.12 | 1.25 ± 0.50 |
| VigRX 15 mg/kg/D | 10 | 0.80 ± 0.43 | 3.82 ± 1.57 | 12.28 ± 2.82 | 43.00 ± 10.63 | 28.20 ± 6.69 | 2.75 ± 1.50 |
| VigRX 30 mg/kg/D | 10 | 0.66 ± 0.37 | 1.82 ± 0.54** | 8.99 ± 2.37* | 50.57 ± 10.28* | 35.71 ± 10.13** | 3.14 ± 1.21* |

* $P < 0.05$ ** $P < 0.01$

Table 2 Compare the length and width of the erect penile size between the rats treated with VigRX and control group on Day 15

| Group | No. of rats | Erect penile size (mm.) | |
|---|---|---|---|
| | | Length | Width |
| Control | 10 | 10.31 ± 0.80 | 5.60 ± 0.44 |
| VigRX 15 mg/kg/D | 10 | 11.38 ± 1.16* | 5.80 ± 0.45 |
| VigRX 30 mg/kg/D | 10 | 12.57 ± 1.12** | 6.03 ± 0.21* |

* P < 0.05 ** P < 0.01

Table 3 The intracavernosal pressure (ICP) of the VigRX - treated groups compare with the control group on Day 15

| Group | No. of rats | ICP (mm Hg) |
|---|---|---|
| Control | 10 | 49.70 ± 2.49 |
| VigRX 15 mg/kg/D | 10 | 70.68 ± 3.66*** |
| VigRX 30 mg/kg/D | 10 | 83.56 ± 5.33*** |

*** P < 0.001

Table 4 Compare the sperm density and testosterone level of the rats treated with VigRX and the control group on Day 15

| Group | No. of rats | Sperm density ($x10^7$ / ml) | Testosterone (ng/ml) |
|---|---|---|---|
| Control | 10 | 21.00 ± 2.95 | 5.79 ± 2.15 |
| VigRX 15 mg/kg/D | 10 | 22.67 ± 1.64 | 4.69 ± 2.37 |
| VigRX 30 mg/kg/D | 10 | 24.05 ± 3,76* | 5.51 ± 2.18 |

* $P < 0.05$

Table 5 Body weight and various organs weight of VigRX - treated groups compare with the control group on Day 15

| Parameters | Control | VigRX | |
|---|---|---|---|
| | | 15 mg/kg/D | 30 mg/kg/D |
| No. of rats | 10 | 10 | 10 |
| Body weight (g) | 398.20 ± 19.59 | 393.40 ± 16.90 | 392.30 ± 11.53 |
| Penis weight (mg%) | 71.00 ± 11.81 | 75.82 ± 9.86 | 71.22 ± 5.79 |
| Testes weight (mg%) | 928.29 ± 60.25 | 934.52 ± 51.29 | 929.01 ± 18.13 |
| Epididymis weight (mg%) | 256.99 ± 14.53 | 265.30 ± 16.84 | 250.50 ± 10.91 |
| Seminal vesicle weight (mg%) | 313.30 ± 47.27 | 340.56 ± 45.77 | 332.02 ± 40.94 |
| Prostate gland weight (mg%) | 96.13 ± 18.68 | 106.30 ± 14.66 | 110.37 ± 25.25 |
| Liver weight (mg%) | 4010.33 ± 498.80 | 3512.82 ± 197.19** | 3552 .43 ± 220.40** |
| Kidney weight (mg%) | 710.38 ± 38.34 | 664.68 ± 24.80** | 685.69 ± 27.64 |
| Adrenal grand weight (mg%) | 16.08 ± 1.77 | 15.53 ± 1.47 | 14.98 ± 1.42 |
| Spleen weight (mg%) | 232.80 ± 22.06 | 233.01 ± 11.42 | 247.74 ± 25.64 |
| Pituitary gland weight (mg%) | 2.65 ± 0.45 | 2.65 ± 0.49 | 2.79 ± 0.48 |

** $P < 0.01$

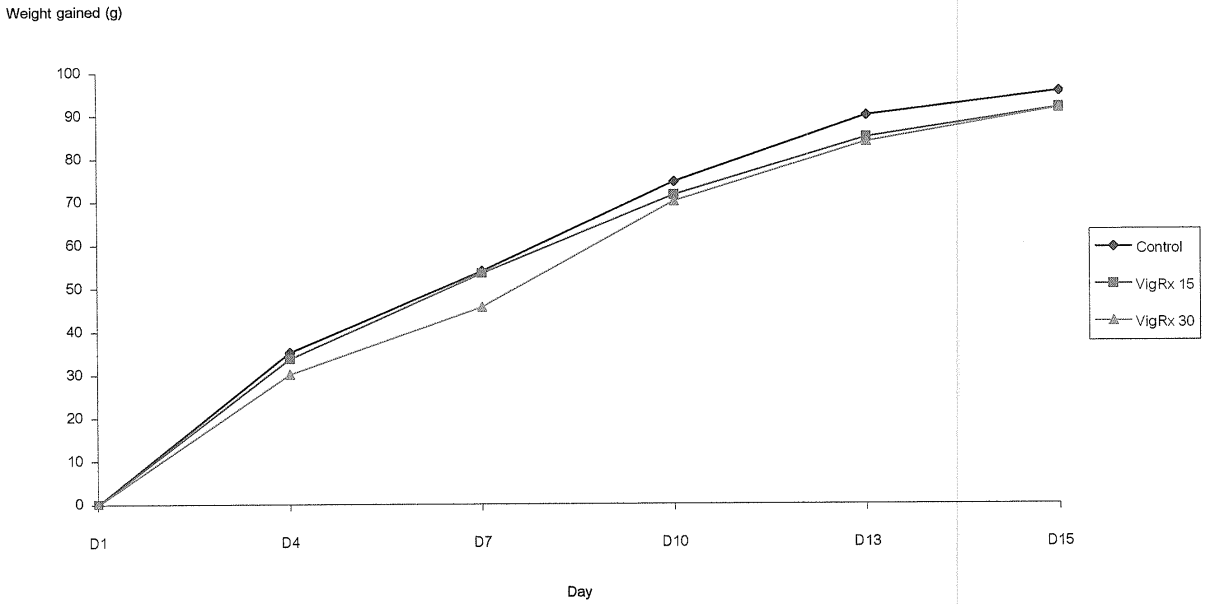


Fig. 1 Compare the body weight gained of the male rats between the VigRX treated groups and the control group

Fig.2 shows the mounting behavior of the male rat

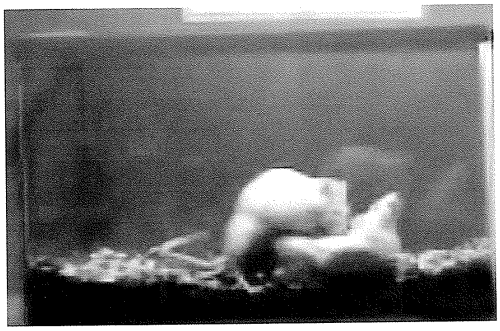

Fig.3 shows the intromission behavior of the male rat

Fig.4 shows the ejaculation behavior of the male rat

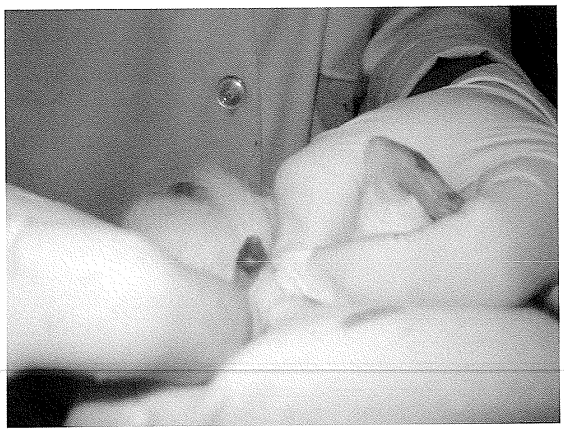

Fig.5 shows the erect penile size of the control male rat at Day 15

Fig.6 shows the erect penile size of the VigRX (15 mg/kg/D) - treated male rat at Day 15

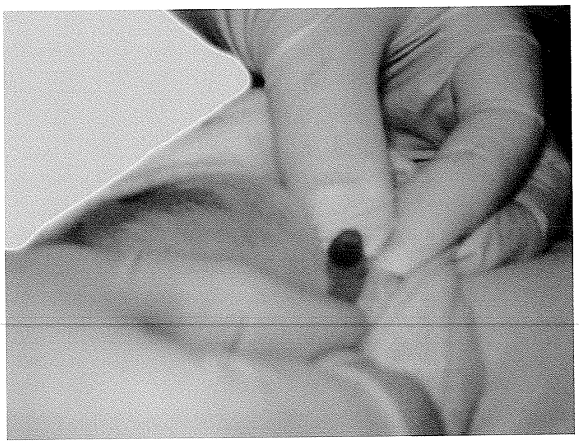

Fig.7 shows the erect penile size of the VigRX (30mg/kg/D) - treated male rat at Day 15

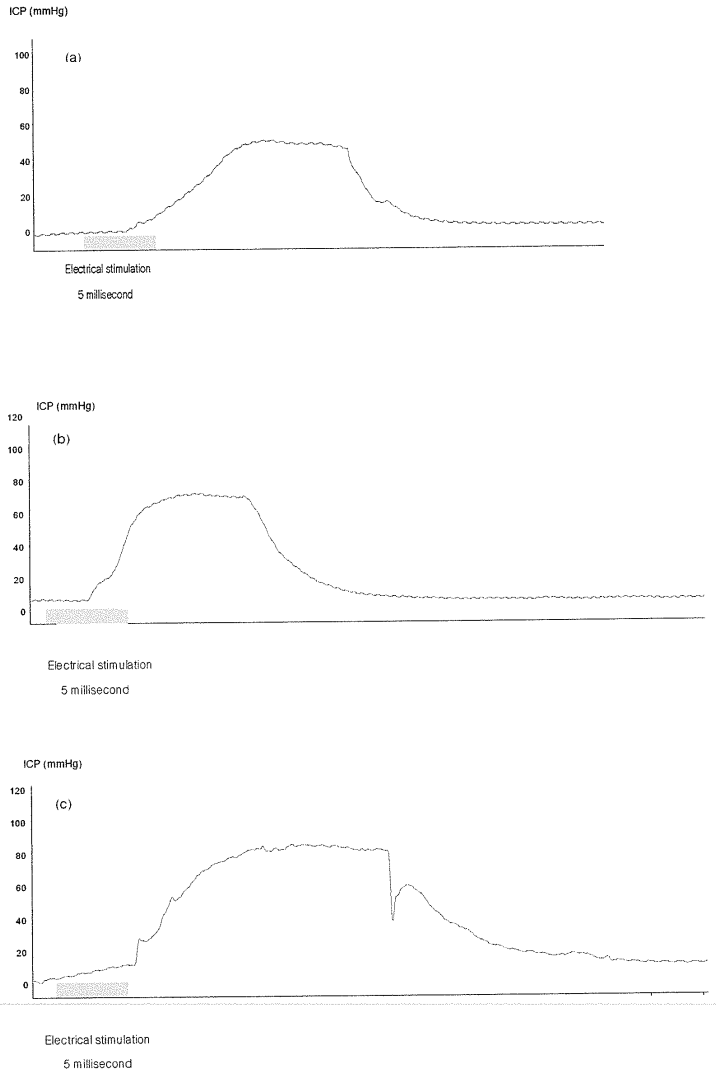


Figure 8 Representative changes in the ICP recorded from (a) a control, (b) 15 mg/kg /D and (c) 30 mg/kg/D of the VigRX-treated rats. Cavernous nerve stimulus parameters were 5 Volts, frequency of 20 Hertz and duration of 5 msec.

Discussion

VigRX had no effect on the body weight gained but VigRX 30 mg/kg/D seemed to have some effect on the body weight gained by decreasing the body weight gained although the statistical analysis did not significantly different.

Studies on the sexual behavior of the rats treated with 2 dosages of VigRX compared with the control group revealed that the rats fed with VigRX especially at the dosage of 30 mg/kg/D showed very high sex drive to the induced heat female rats as reflected by the high frequency of every parameters i.e. mounting , intromission and ejaculation frequencies and short latency of intromission and ejaculation latency during 30 - minute of sexual behavior observation. The male rats of this group frequently mount the female rats. The results showed that VigRX could induce high sex drive. These behaviors may be due to some ingredients in VigRX which can pronounce the aphrodisiac effects i.e. Saw Palmetto berry , Muira pauma bark extract and Catuaba bank extract.

The results of VigRX on increasing of the sex drive is approved in this experiment . VigRX not only increase the sex drive but also increase the erect penile size , the intracavernosal pressure and the sperm density. These important evidences mean that VigRX could increase sex drive , penile size , erection and may treat the male sterility. These actions of VigRX like *Butea superba* in increasing the erect penile size , erection and sperm density ( Smitasiri et al , 2003). According to VigRX , the increasing of sperm density may involve with Cuscuta seed extract ( Peng et al , 1997).

The increasing of sperm density by VigRX may be due to the increasing of spermatogenesis but the increasing of spermatogenesis may not involve with testosterone . In spermatogenesis , the hormones involved are FSH and testosterone , then the mechanism of VigRX on spermatogenesis may be due to the increasing of

blood flow to the testes . This may involve with Hawthorn berry and Ginkgo leaf in VigRX because Hawthorn berry and Ginkgo leaf can increase blood flow.

VigRX had no effects on various organs weight i.e. penis , testes , epididymis , seminal vesicle , prostate gland , adrenal gland , spleen and pituitary gland but VigRX could decrease the liver and kidney weight of the rats.

The mechanisms by which the VigRX could decrease the liver and kidney weight are still unknown. But it is possibly that VigRX may decrease the glycogen content accumulate in the liver by glycogenolysis because the rats receiving VigRX need energy (from glucose which is break down from glycogenolysis) too much for sex drive , mounting behavior , blood flow etc. These effects may be due to some ingredients in VigRX especially Asian Ginseng root and Ginkgo leaf. To prove this hypothesis , histopathological studies of liver and kidney included with some blood chemistry measurement which reflected the liver and kidney function are needed to explore . The repeated experiment on the effects of VigRX on liver and kidney are needed to explain what happen to the liver and kidney .

Conclusion

VigRX at the dosage of 15 mg/kg/D and 30 mg / kg / D orally fed for 14 consecutive days to the adult male Sprague Dawley rats compared with the control group were investigated.

It was concluded that VigRX at the dosage of 30 mg / kg / D could induce the high sex drive, the erect penile size, the penile erection and could increase the sperm density in the rat whereas VigRX had no effect on the various organs weight but had some effects on the liver and kidney by decreasing the weight of both organs significantly.

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