### **CHAPTER IV**

### RESULTS

Experiment 1; Effects of *Mucuna collettii* on hormone-related ovarian functions and reproductive organs in cyclic female rats.

# 1.1 Effects of Mucuna collettii on hormone-related ovarian functions

### 1.1.1 Serum estradiol levels

When compared to the pre-treatment levels  $(D_1)$ , there were no significant differences of serum  $E_2$  levels in rats treated with DW, Mc-1 and Mc-10 throughout the study periods (Figure 7). In Mc-100 and TP groups, serum  $E_2$  levels were significantly increased only at  $D_{31}$  or 15 days after the administration of Mc-100 and TP.

Comparison to the DW group, serum  $E_2$  levels of Mc-1 group were significantly higher than that of the DW group at  $D_1$ ,  $D_{46}$  and  $D_{61}$ . There were no significant differences from the DW group throughout the study periods in Mc-10 group. Serum  $E_2$  levels of Mc-100 group at  $D_{16}$  and  $D_{31}$ , and TP group at  $D_{31}$  were significantly higher than those of DW group.

Comparison to the TP group, serum  $E_2$  levels of Mc-1 group showed the similarity of significant difference as it was to the DW group, the serum  $E_2$  levels were significantly higher than that of TP group at  $D_1$ ,  $D_{46}$  and  $D_{61}$ . Serum  $E_2$  levels of Mc-

10 and Mc-100 groups did not significant differences from the TP group throughout the study periods, except for D<sub>46</sub> of Mc-100 group, the level was significantly higher.

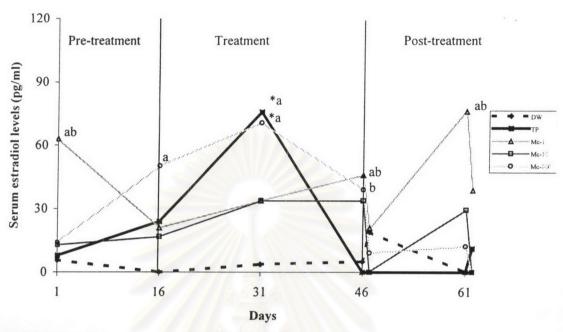
### 1.1.2 Serum LH levels

When compared to the pre-treatment levels (D<sub>1</sub>), serum LH levels of DW and Mc-100 groups did not significant differences throughout the study periods (Figure 8). Serum LH levels of TP group were significantly decreased during D<sub>31</sub>-D<sub>62</sub>. Serum LH levels of Mc-1 and Mc-10 groups were significantly increased at D<sub>62</sub> and D<sub>61</sub>, respectively.

Comparison to the DW group, serum LH levels of Mc-1 and Mc-10 groups were no significant differences throughout the study periods. Serum LH levels of Mc-100 group were significantly higher than the DW group at  $D_1$ ,  $D_{16}$ ,  $D_{46}$  and  $D_{47}$ . Serum LH levels of TP group were significantly lower than the DW group at only  $D_{47}$ .

Comparison to the TP group, serum LH levels of Mc-1 group at  $D_{47}$  and  $D_{62}$  were significantly higher than the TP group. Serum LH levels in Mc-100 group were also higher than that of TP group at  $D_1$ ,  $D_{16}$  and  $D_{46}$ - $D_{61}$ . Serum LH levels of Mc-10 group did not significantly different throughout the study periods.

Figure 7 Changes of serum estradiol levels in cyclic female rats treated with distilled water, testosterone propionate and *M. colettii*.

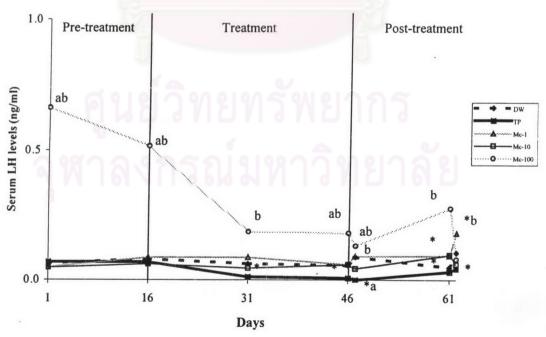


\* Significantly different compared to D<sub>1</sub> levels.

<sup>a</sup> Significantly different of TP and Mc groups compared to DW group.

<sup>b</sup> Significantly different of Mc groups compared to TP group.

Figure 8 Changes of serum LH levels in cyclic female rats treated with distilled water, testosterone propionate and M. colettii.



\* Significantly different compared to D<sub>1</sub> levels.

<sup>a</sup> Significantly different of TP and Mc groups compared to DW group.

<sup>b</sup> Significantly different of Mc groups compared to TP group.

# 1.1.3 Serum FSH levels

When compared to the pre-treatment levels ( $D_1$ ), serum FSH levels of Mc-1 group at  $D_{47}$  and Mc-10 group at  $D_{16}$  and  $D_{62}$  were significantly increased (Figure 9). In Mc-100 and TP groups, serum FSH levels did not changes throughout the study periods.

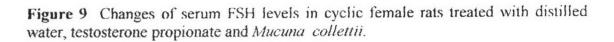
Comparison to the DW group, serum FSH levels of Mc treated groups were not significant differences throughout the study periods, except serum FSH levels of Mc-1 group were significantly higher at  $D_{47}$ . In contrast, serum FSH levels of TP group were significantly lower at  $D_{62}$ .

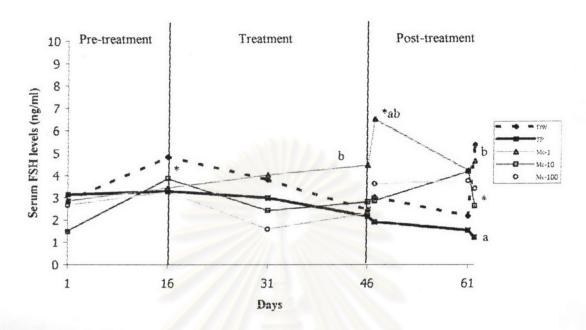
Comparison to the TP group, serum FSH levels of Mc treated groups were no significant differences throughout the study periods, except serum FSH levels of Mc-1 group were significantly higher at  $D_{46}$ ,  $D_{47}$  and  $D_{62}$ .

# 1.2 Effects of Mucuna collettii on ovaries and uteri.

Comparison the weight and absolute weight of ovaries and uteri between the end of treatment to the end of post-treatment period, there were no significant differences in DW and all Mc groups (Table 1). The TP group showed the increase of weights and absolute weights of ovaries and uteri at the end of post-treatment period.

Comparison to the DW group, the uterine weight of Mc-10 group at the end of treatment period was significantly higher, while the absolute ovarian weight of Mc-





\* Significant difference compared to D<sub>1</sub> levels.

<sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

# ศูนย์วิทยทรัพยากร จุฬาลงกรณ์มหาวิทยาลัย

Table 1 Changes of the weights and absolute weights of ovaries and uteri at the end of treatment and at the end of post-treatment periods of cyclic female rats treated with distilled water, testosterone propionate and Mucuna collettii.

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	A MANAGE CONTINUES OF STREET,		Uteri	Post-	treatment	1 54+0 15	1.06+0.09*	1.64+0.19 <sup>b</sup>	1.46+0.19	$1.32\pm0.15$
the state of the s	weight	grams)	Ü	Treatment		1.49+1.50	$2.55+0.09^{a}$	1.33+0.08 <sup>b</sup>	1.21+0.14 <sup>b</sup>	$1.24\pm0.07^{b}$
The state of summer two depths is stated only and way depths and place	Absolute weight	(x 10 grams)	res	Post-	treatment	0.57+0.03	0.30+0.02*8	0.55±0.02 <sup>b</sup>	0.58±0.03b	0.47±0.02ab
			Ovaries	Treatment		0.55+0.02	$0.41\pm0.02^{a}$	$0.54 \pm 0.03^{b}$	0.64+0.03ab	$0.53\pm0.02^{b}$
The second secon		ind	211	Post-	neamment	$0.41 \pm 0.03$	0.32+0.03*	0.45±0.05°	0.36+0.04	0.37±0.04
	weignt ms)	Itori	5	Ireatment		0.42±0.05	0.68±0.02ª	0.34±0.02°	0.31+0.04	0.33+0.02
Organo	(gran	Ovaries		Fost- treatment		0.015±0.01	0.09±0.01	0.15±0.01°	0.14+0.01	10.0±0.01
		Ova	Trootmont	Heatillell	0 16 0 01	0.10-0.01	0.11-0.01	0.14±0.01	0.14+0.01	10.01
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Ę.		4			DW	TP	Mc-1	Mc-10	Mc-100	

\* Significant difference compared to at the end of treatment period.

<sup>&</sup>lt;sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

100 group was significantly lower at the end of post-treatment period. In TP group, the weight and absolute weight of ovaries were significantly lower than the DW group in both periods. The uterine weight of TP group were significantly higher at the end of treatment period and recovered during the end of post-treatment period, but the absolute uterine weight was lower than that of DW group at the end of post-treatment period.

Comparison to the TP group, the weight and absolute weight of ovaries in Mc treated groups were significantly higher in both periods. Uterine weight and its absolute weight in all Mc treated groups were significantly lower at the end of treatment period and only in Mc-1 group were significantly higher at the end of post-treatment period.

# 1.3 Effects of Mucuna collettii on histological changes of ovaries and uteri

### 1.3.1 Histological changes of ovaries

The ovarian tissue of DW and all Mc treated groups at the end of treatment period presented normal ongoing folliculogenesis and corpus luteum formation (Figure 10A-D). There were many corpus lutea in all Mc treated groups than DW group. In contrast, the ovaries treated with TP at the end of treatment period showed some large follicles atresia. In addition, some large corpora lutea were occurred in this group (Figure 10E-F).

As regards to ovary of DW and all Mc treated groups at the end of post-treatment period, it demonstrated normal ongoing folliculogenesis and corpus luteum formation. There were some atretic follicles with some large corpora lutea in TP treated group (Figure 10G).

When the ovarian histology at the end of treatment period was compared to ovary at the end of post-treatment period, it was found that there were no differences in all groups.

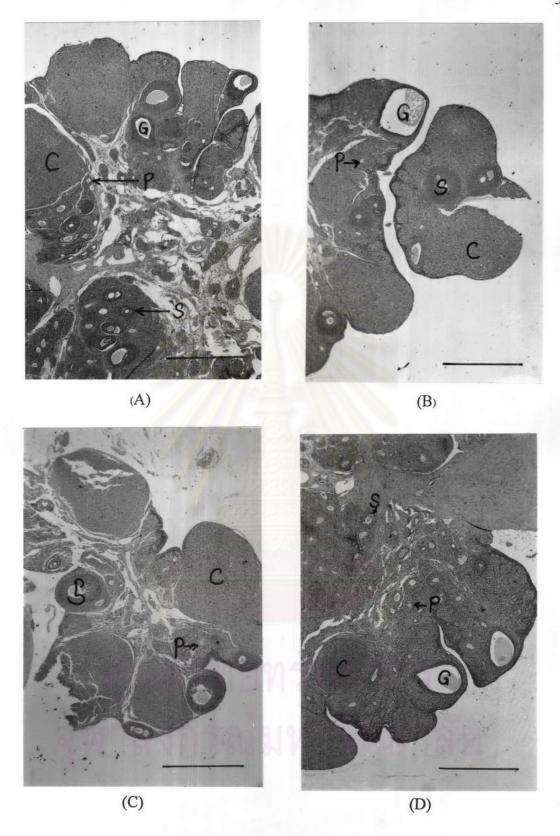


Figure 10 Ovarian tissue of cyclic female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E,F) and TP at the end of post-treatment periods (G) with H & E staining for primary follicles (P), secondary follicles (S), Graafian follicles (G) and corpus lutea (C). Bar scale = 200  $\mu$ m

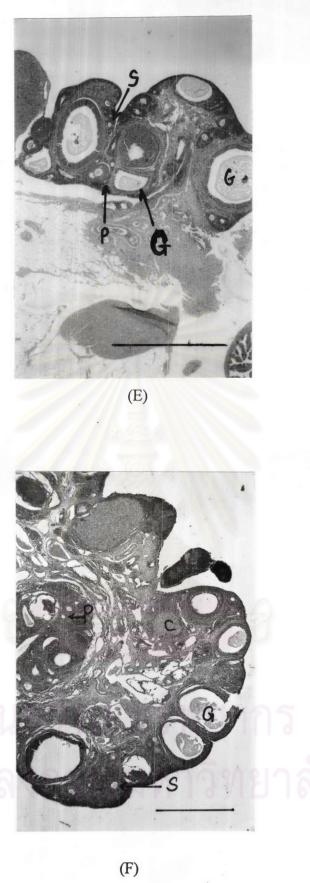


Figure 10 (continued) Ovarian tissue of cyclic female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E,F) and TP at the end of post-treatment periods (G) with H & E staining for primary follicles (P), secondary follicles (S), Graafian follicles (G) and corpus lutea (C). Bar scale = 200 μm

### 1.3.2 Histological changes of uteri

The uterine histological study of DW and all Mc treated groups at the end of treatment period showed the normal proliferation and no differences between all 4 groups. The mucosal epithelial cell lining was a layer of cuboidal cells (Figure 11A-D). In contrast, the uteri treated with TP revealed the marked increased in endometrial thickness, number and diameter of endometrial glands when compared to DW group. The mucosal epithelial cell lining was a layer of columnar cells (Figure 11E-F).

According to the uterine morphology of DW and all Mc treated groups at the end of post-treatment period, it was found that these groups revealed normal proliferation and no differences between all 4 groups. The mucosal epithelial cell lining was a layer of cuboidal cells. In case of TP group, the endometrial layer, number and diameter of endometrial glands were increased when compared to DW group. In addition, the epithelial cell lining was a layer of columnar cells (Figure 11G-H).

The histology of uteri at the end of treatment period was compared to the end of post-treatment period, there were no changes of uterine histology in all groups, except for TP group, endometrial thickness, number and diameter of endometrial glands at the end of treatment period were higher than at the end of post-treatment period.

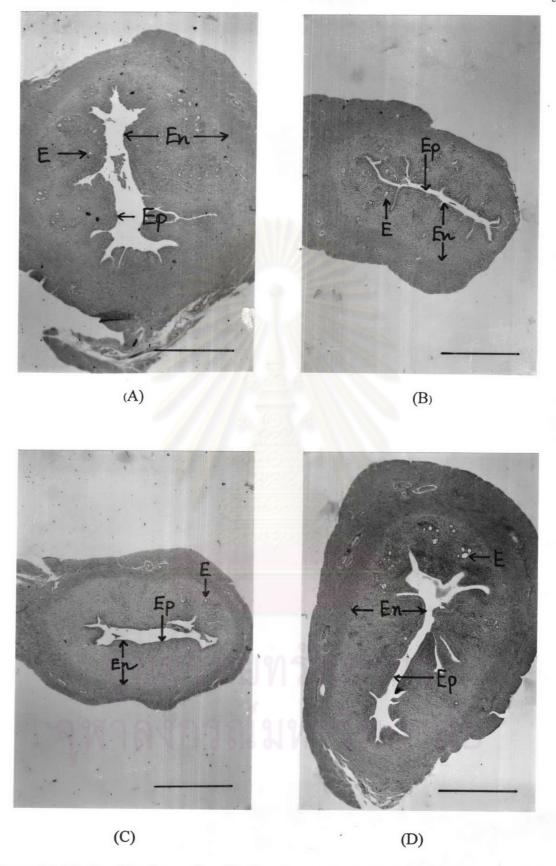


Figure 11 Uterine histology of cyclic female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment period (E,F) with H & E staining for endometrium (En), epithelial cells (Ep) and endometrial gland (E). Bar scale =  $200 \, \mu m$ 

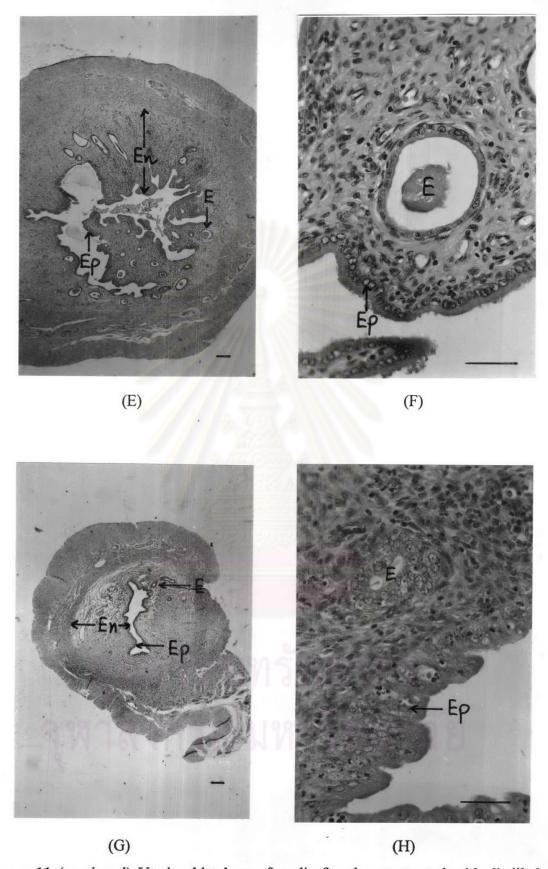


Figure 11 (continued) Uterine histology of cyclic female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E,F) and TP at the end of post-treatment periods (G,H) with H & E staining for endometrium (En), epithelial cells (Ep) and endometrial gland (E). Bar scale =  $50 \mu m$ 

### 1.4 Effects of Mucuna collettii on estrous cycles

The estrous cycles in DW and all Mc treated groups were regular approximately 4-5 days throughout the study periods (Table 2,4-6), except for the Mc-100 group, the estrous cycles tended to be prolonged during treatment period (estrous cycle length = 5.40±0.11 days, p=0.058). In TP group, all rats showed leukocytes after TP treatment for approximately 1 estrous cycle until the last day of study periods (Table 3).

Estrous cycle length were no significant differences between each period between all Mc treated groups (Table 7), except for Mc-100 group, estrous cycle length in treatment period tended to be longer (p=0.058) when compared to DW group (Table 7).

Experiment 2; Efects of *Mucuna collettii* on hormone-related ovarian functions and reproductive organs in ovariectomized female rats.

# 2.1 Effects of Mucuna collettii on hormone-related ovarian functions

# 2.1.1 Serum estradiol levels

Basal serum  $E_2$  levels ( $D_1$ ) in femal rats after ovariectomy were significantly lower than the cyclic female rats ( $0\pm0$  and  $19.56\pm7.04$  pg/ml in ovariectomized rats and cyclic rats, respectively). Serum  $E_2$  levels were kept in the low levels throughout the study periods (Figure 12). No significant differences were found between that 3

periods in any treated groups, and also between that 4 groups in any treatment periods.

### 2.1.2 Serum LH levels

Basal serum LH levels (D<sub>1</sub>) in female rats after ovariectomy were significantly higher than the normal cyclic female rats in Experiment 1 (12.67±1.08 and 0.18±0.12 ng/ml in OVX rats and normal cyclic female rats, respectively) (Figure 13). In DW and all Mc treated groups, serum LH levels were significantly increased since D<sub>1</sub> until the end of study period. Serum LH levels of TP group were significantly increased at D<sub>1</sub>-D<sub>16</sub>, and then decreased to the D<sub>-14</sub> levels after 2 weeks of TP injection until 2 weeks of TP withdrawal.

When compared to the pre-treatment levels  $(D_1)$ , serum LH levels of DW and Mc-1 groups during  $D_{31}$ - $D_{62}$  were significantly increased. However, the LH levels of Mc-10 and Mc-100 groups were not significantly different from the  $D_1$  levels at  $D_{31}$  and  $D_{16}$ - $D_{31}$ , respectively. Serum LH levels of TP group were significantly higher than the  $D_1$  only at  $D_{16}$ .

Comparison to the DW group, serum LH levels of Mc-1 group were significantly higher than the DW group at D<sub>61</sub>. There were not significant differences in serum LH levels of Mc-10 and Mc-100 groups throughout the study periods except at D<sub>31</sub> of Mc-10 LH levels was lower than the DW group. In TP group, serum LH levels were significantly lower than the DW group since 2 weeks of TP injection until 2 weeks of TP withdrawal.

Table 2 Changes of vaginal cytology in cyclic female rats treated with distilled water. | and mindicate fully and partial cornification, respectively.

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Table 3 Changes of vaginal cytology in cyclic female rats treated with testosterone

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\* indicated the sacrifice day of the rats

Table 4 Changes of vaginal cytology in cyclic female rats treated with 1 mg/kgBW of Mucuna collettii. and mindicate fully and partial cornification, respectively 3/4 • ü \* -34 \* . × . -Days 51 52 Days . . m 37 38 . % 8.1 No 8.1 

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Table 5 Changes of vaginal cytology in cyclic female rats treated with 10 mg/kgBW of Mucuna collectii. 📓 and windicate fully and partial cornification, respectively.

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Table 6 Changes of vaginal cytology in cyclic female rats treated with 100 mg/kgBW of Mucuna collectif. 📓 and windicate fully and partial cornification, respectively.

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**Table 7** Changes of estrous cycle length in each study period in cyclic female rats treated with distilled water, testosterone propionate and *Mucuna collettii*.

Group	Est	rous cycle length	(days)
	Pre-treatment	Treatment	Post-treatment
DW	4.76+0.18	4.44+0.16	4.75+0.25
TP	4.36+0.15	-	-
Mc-1	4.54+0.19	4.71+0.17	4.22+0.14
Mc-10	4.03±0.32	4.48+0.15	4.27+0.19
Mc-100	4.87±0.24	5.40+0.11	4.42+0.16



Figure 12 Changes of serum estradiol levels in ovariectomized female rats treated with distilled water, testosterone propionate and *Mucuna collettii*.

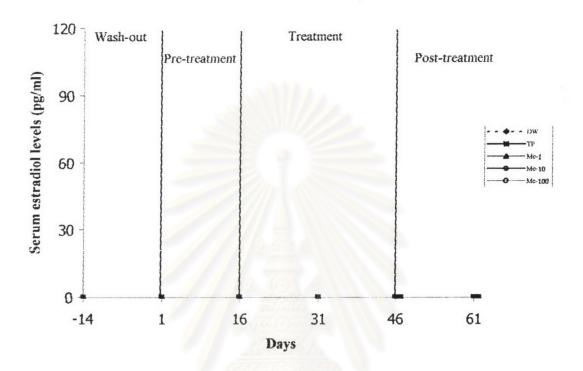
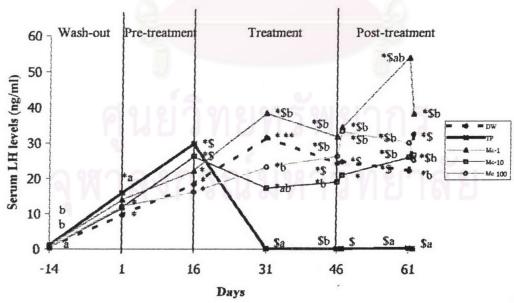


Figure 13 Changes of serum LH levels in ovariectomized female rats treated with distilled water, testosterone propionate and Mucuna collettii.



<sup>\*</sup> Significant difference compared to D-14 levels.

<sup>\$</sup> Significant difference compared to D<sub>1</sub> levels.

<sup>&</sup>lt;sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>&</sup>lt;sup>b</sup> Significant difference of Mc groups compared to TP group.

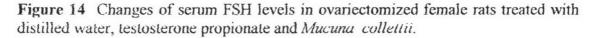
### 2.1.3 Serum FSH levels

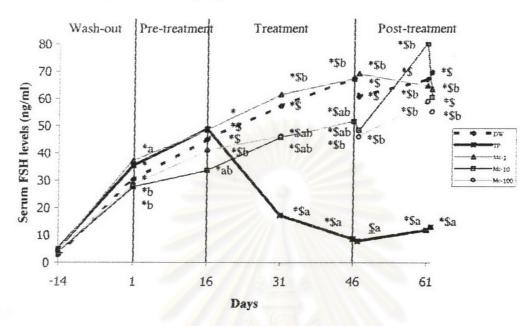
Basal serum FSH levels (D<sub>1</sub>) in female rats after ovariectomy were significantly higher than the normal cyclic female rats in Experiment 1 (31.75±1.93 and 2.60±0.29 ng/ml in OVX rats and normal female rats, respectively) and the levels at D<sub>-14</sub> as shown in Figure 14. Serum FSH levels were kept in the high levels throughout the study periods in DW and all Mc treated groups. Except for the TP group, serum FSH levels at D<sub>47</sub> were not significantly differences from D<sub>-14</sub>.

When compared to the pre-treatment levels ( $D_1$ ), serum FSH levels of DW and all Mc treated groups were significantly increased during  $D_{16}$ - $D_{62}$ , except for  $D_{16}$  of the Mc-1 and Mc-10 groups. In TP group, serum FSH levels were significantly increased higher than  $D_1$  at only  $D_{16}$  while the levels were significantly decreased at  $D_{31}$ - $D_{62}$ .

Comparison to the DW group, there were no different of serum FSH levels between Mc-1 and DW group, except at  $D_1$ . The serum FSH levels of Mc-10 group during  $D_{16}$ - $D_{46}$  and Mc-100 group during  $D_{31}$ - $D_{46}$  were significantly lower and recovered to the DW levels thereafter. On the other hand, serum FSH levels in Mc-10, Mc-100 and TP groups were significantly lower than that of DW group at  $D_{16}$ - $D_{46}$ ,  $D_{31}$ - $D_{46}$  and  $D_{31}$ - $D_{62}$ , respectively.

Comparison to the TP group, serum FSH levels of Mc-10 and Mc-100 groups were significantly lower at  $D_1$ - $D_{16}$  while the FSH levels of all Mc treated groups were significantly higher during  $D_{31}$ - $D_{62}$ .





<sup>\*</sup> Significant difference compared to D-14 levels.

<sup>6</sup> Significant difference of Mc groups compared to TP group.

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<sup>\$</sup> Significant difference compared to D<sub>1</sub> levels.

<sup>&</sup>lt;sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

### 2.2 Effects of Mucuna collettii on uteri

There were no significant differences of uterine weight and its absolute weight in DW and all Mc treated groups at the end of post-treatment period when compared to that of the end of treatment period (Table 8). In TP group, uterine weight and its absolute weight at the end of post-treatment period were significantly decreased when compared to the treatment period.

Comparison to the DW group, uterine weight and its absolute weight of DW and all Mc treated groups were no significant differences between groups. In TP group, uterine weight and its absolute weight were significantly higher in both treatment and post-treatment periods.

Comparison to the TP group, uterine weight and its absolute weight of all Mc treated groups were significantly lower in both treatment and post-treatment periods.



**Table 8** Changes of the weight and absolute weight of uteri at the day of treatment and at the day of post-treatment periods in ovariectomized female rats treated with distilled water, testosterone propionate and *Mucuna collettii*.

Group		ne weight rams)	Absolute t (x 10	uterus weight <sup>3</sup> grams)
	Treatment	Post-treatment	Treatment	Post-treatment
DW TP 600 μg/100gBW/day M. collettii 1 mg/kg/day M. collettii 10 mg/kg/day M. collettii 100 mg/kg/day	0.13±0.01 0.50±0.01 <sup>b</sup> 0.12±0.01 <sup>c</sup> 0.11±0.01 <sup>c</sup> 0.12±0.01 <sup>c</sup>	0.12±0.00 0.29±0.03 <sup>ab</sup> 0.13±0.02 <sup>c</sup> 0.12±0.01 <sup>c</sup> 0.12±0.01 <sup>c</sup>	0.38±0.02 1.62±0.03 <sup>b</sup> 0.39±0.03 <sup>c</sup> 0.32±0.02 <sup>c</sup> 0.37±0.03 <sup>c</sup>	0.33±0.03 0.84±0.06 <sup>ab</sup> 0.39±0.06 <sup>c</sup> 0.34±0.02 <sup>c</sup> 0.37±0.03 <sup>c</sup>

<sup>&</sup>lt;sup>a</sup> Significant difference compared to the end of treatment period.

### 2.3 Effects of Mucuna collettii on histological changes in uteri

### Histological changes in uteri

The uterine morphology of DW and all Mc treated groups at the end of treatment period appeared to be the thin endometrial mucosa layers. The mucosa epithelial cell lining was a layer of low cuboidal cells. Few number and narrow lumen of endometrial glands that dispersed in some area of endometrium (Figure 15A-D). In controversy, the endometrium of rats in TP group was thick. The epithelial cell lining was a layer of columnar cells. Number and diameter of endometrial glands were increased when compared to DW group (Figure 15E).

<sup>&</sup>lt;sup>b</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>&</sup>lt;sup>c</sup> Significantl difference of Mc groups compared to TP group.

The histology of uteri of DW and Mc treated groups at the end of post-treatment period demonstrated the thin endometrium. The epithelial cell lining was a layer of low cuboidal cells. Few dispersal endometrial glands with narrow lumen were noticed. In contrast, the endometrial thickness, number and diameter of endometrial glands of TP group increased when compared to DW group. In addition, the epithelial cell lining was a layer of columnar cells (Figure 15F).

There were no different of histological structure of uteri at the end of treatment period and at the end of post-treatment period in all treatment groups, except endometrial layer thickness, number and diameter of endometrial glands of rats in TP group at the end of treatment period were higher than that of post-treatment period.

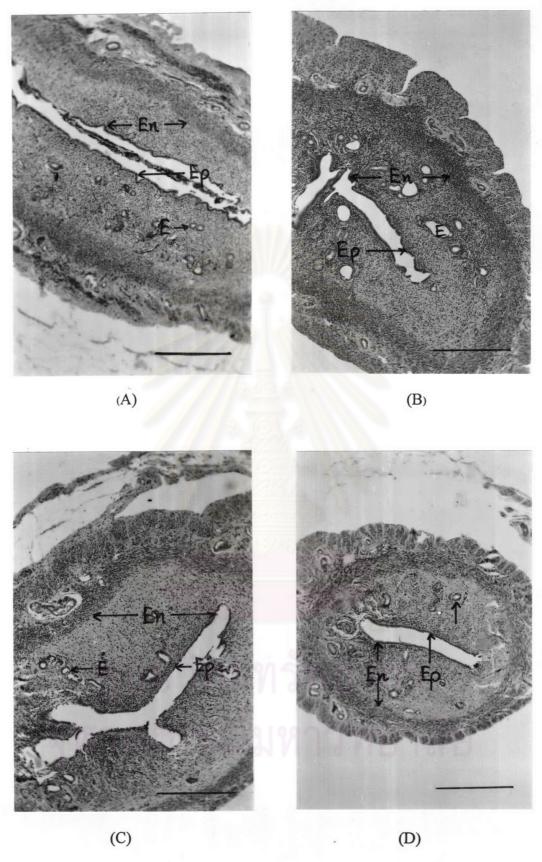


Figure 15 Uterine histology of ovariectomized female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining for endometrium (En), epithelial cells (Ep) and endometrial gland (E). Bar scale = 200  $\mu$ m

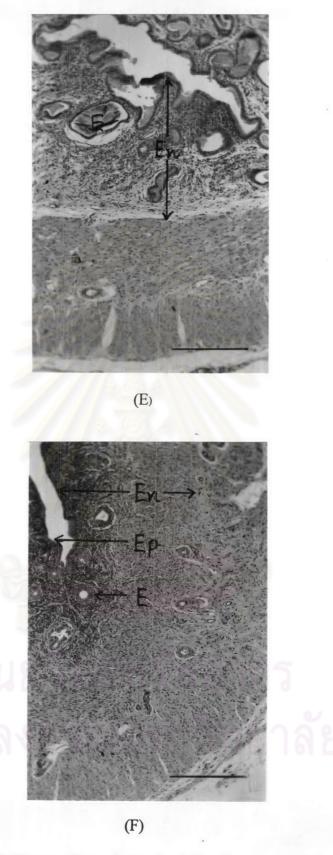


Figure 15 (continued) Uterus of ovariectomized female rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining for endometrium (En), epithelial cells (Ep) and endometrial gland (E). Bar scale =  $200 \mu m$ 

# 2.4 Effects of Mucuna collettii on estrous cycles

There were no estrous cycles in all of the experimental groups and vaginal smear was completely found leukocyte cells throughout the experiment (Table 9-13).



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Table 10 Changes of vaginal cytology in ovariectomized female rats treated with testosterone propionate. @ indicates vaginal leukocytes.

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Table 11 Changes of vaginal cytology in ovariectomized female rats treated with 1 mg/kgBW of Mucuna collettii. © indicates vaginal leukocytes.

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Table.12 Changes of vaginal cytology in ovariectomized female rats treated with 10 mg/kgBW of Mucuna collettii. © indicates vaginal leukocytes.

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Table 13 Changes of vaginal cytology in ovariectomized female rats treated with 100 mg/kgBW of Mucima collettii. © indicates vaginal leukocytes.

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Experiment 3; Effects of *Mucuna collettii* on hormone-related testicular functions and reproductive organs in normal male rats.

### 3.1 Effects of Mucuna collettii on hormone-related testicular functions

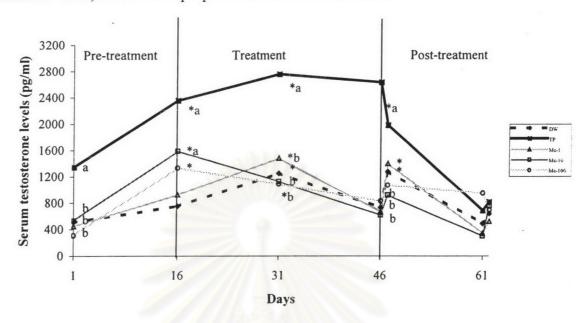
### 3.1.1 Serum testosterone levels

When compared to the pre-treatment levels (D<sub>1</sub>), serum testosterone levels of DW and Mc-1 groups were significantly increased only at D<sub>31</sub>-D<sub>47</sub> (Figure 16). In Mc-10 and Mc-100 groups, serum testosterone levels were significantly increase at D<sub>16</sub> and D<sub>16</sub>-D<sub>31</sub>, respectively. Serum testosterone levels of TP group were significantly increased during treatment period and recovered to the pre-treatment values after 2 weeks of TP withdrawal.

Comparison to the DW group, there were no significant differences in all of Mc treated groups throughout the study periods, except of Mc-10 group, serum testosterone levels were significantly higher at  $D_{16}$ . Serum testosterone levels of TP group were significantly higher than the DW group since  $D_1$  and kept higher than the DW group throughout the treatment and early post-treatment periods ( $D_{16}$ - $D_{47}$ ).

Comparison to the TP group, serum testosterone levels of all Mc treated groups were significantly lower since  $D_1$  of study period and the serum testosterone levels were kept in the lower levels than the TP group throughout the treatment and early post-treatment periods ( $D_{16}$ - $D_{47}$ ).

Figure 16 Changes of serum testosterone levels in normal male rats treated with distilled water, testosterone propionate and *Mucuna collettii*.

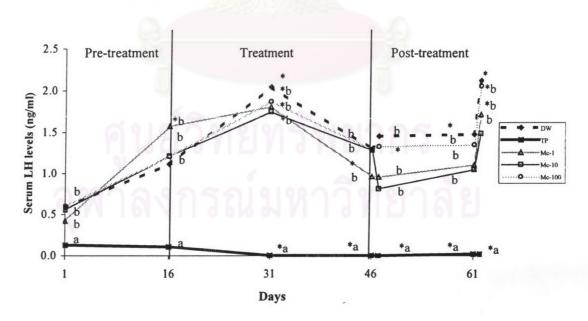


\* Significant difference compared to D<sub>1</sub> levels.

<sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

Figure 17 Changes of serum LH levels in normal male rats treated with distilled water, testosterone propionate and *Mucuna collettii*.



\* Significant difference compared to D<sub>1</sub> levels.

<sup>a</sup> Significant difference of TP and Mc groups compared to DW group.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

#### 3.1.2 Serum LH levels

When compared to the pre-treatment levels (D<sub>1</sub>), serum LH levels of DW group were significantly increased during D<sub>31</sub>-D<sub>62</sub> (Figure 17). Serum LH levels of Mc treated groups were significantly higher than the pre-treatment levels in some points, Mc-1 group at D<sub>16</sub>-D<sub>31</sub> and D<sub>62</sub>, Mc-10 group at D<sub>31</sub> and Mc-100 group at D<sub>31</sub> and D<sub>62</sub>. Serum LH levels of TP group were significantly decreased after 15 days of TP administration until the injection was stop for 2 weeks.

Comparison to the DW group, serum LH levels of all Mc treated groups were no significant differences throughout the study periods. In contrast, serum LH levels of TP group were significantly lower than the DW group since  $D_1$  to the end of study period  $(D_1-D_{62})$ .

Comparison to the TP group, serum LH levels of all Mc treated groups were significantly higher since  $D_1$  until the end of study period ( $D_1$ - $D_{62}$ ).

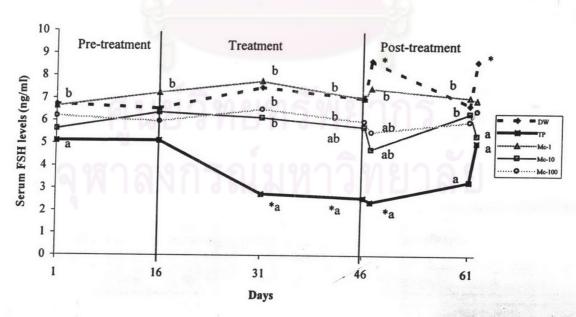
### 3.1.3 Serum FSH levels

When compared to the pre-treatment levels (D<sub>1</sub>), serum FSH levels in DW and all Mc groups were no significant differences throughout the study periods (Figure 18), except in the DW group, the levels were significantly increased at D<sub>47</sub> and D<sub>62</sub>, but serum FSH levels of TP group were significantly decreased during D<sub>31</sub>- D<sub>47</sub>.

Comparison to the DW group, serum FSH levels of Mc-1 group were no significant differences throughout the study periods. Serum FSH levels of Mc-10 group at D<sub>46</sub>, D<sub>47</sub> and D<sub>62</sub> were significantly lower and serum FSH levels of Mc-100 group only D<sub>47</sub> were significantly lower. In TP group, serum FSH levels were significantly lower throughout the study period (D<sub>1</sub>-D<sub>62</sub>), except at D<sub>16</sub>, serum FSH levels were not significant differences.

Comparison to the TP group, serum FSH levels of Mc-1 group during  $D_1$ - $D_{61}$  were significantly higher. Serum FSH levels of Mc-10 group during  $D_{31}$ - $D_{61}$ , and Mc-100 group during  $D_{31}$ - $D_{47}$  were significantly higher and recovered to the TP levels at  $D_{62}$  and during  $D_{61}$ - $D_{62}$ , respectively.

Figure 18 Changes of serum FSH levels in normal male rats treated with distilled water, testosterone propionate and *Mucuna collettii*.



<sup>\*</sup> Significant difference compared to D<sub>1</sub> levels.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

a Significant difference of TP and Mc groups compared to DW group.

### 3.2 Effects of Mucuna collettii on testes, epididymis and seminal vesicle

When the weights and absolute weight of testes, epididymis and seminal vesicle in each group were compared between 2 periods of at the end of treatment and at the end of post-treatment, epididymal weights of DW group were significantly increased, but absolute testicular weight was significantly decreased (Table 14A and 14B). The weights and absolute weights of testes, epididymis and seminal vesicle of Mc treated groups were no significant differences within each group. Except for Mc-10 group, the absolute weight of epididymis was significantly decreased. In TP group, the weights and absolute weights of testes, epididymis and seminal vesicle were significantly decreased.

Comparison to the DW group, the weight and absolute weight of testes, epididymis and seminal vesicle of Mc treated groups were no significant differences. Except epididymal weight of Mc-1 group was significantly higher at the end of treatment period and decreased to the DW levels at the end of post-treatment period. Testicular and epididymal weights and absolute weight of epididymis of Mc-10 group were significantly higher at the end of treatment period and recovered to the DW levels at the end of post-treatment period. In TP group, testicular weight and its absolute weight were no significant differences in both at the end of treatment and at the end of post-treatment periods, except testicular weight at the end of post-treatment period were significantly lower. In addition, the weight and absolute weight of epididymis and seminal vesicle of TP group were significantly higher in both at the end of treatment and at the end of post-treatment periods. Except epididymal weight at the end of post-treatment period were significantly lower.

Comparison to the TP group, Testicular weight of Mc treated groups were significantly higher at the end of post-treatment period. The absolute weight of testes of Mc treated groups were no significant differences in both at the end of treatment and at the end of post-treatment periods. Epididymal weight and its absolute weight of all Mc treated groups were significantly lower at the end of treatment period and increased to the DW levels at the end of post-treatment period. Except epididymal weight of Mc-1 group was significantly higher at the end of post-treatment period. In addition, seminal vesicle weight and its absolute weight of Mc treated groups were significantly lower in both at the end of treatment period and at the end of post-treatment period. Except seminal vesicle of Mc-1 group were increased at the end of post-treatment period.

Table 14A Changes of the weights of testes, epididymis and seminal vesicle at the end of treatment and at the end of post-treatment periods in normal male rats treated with distilled water, testosterone propionate and Mucuna collettii.

	T		Г	-	Г							_
		Vesicle	Door teachers	rost-ucamient	1 30+0 13	1:07-0:13	1.84 + 0.07 * 8	20.00	1.03+0.12	147+0:10 <sup>b</sup>	1.32+0.07 <sup>b</sup>	1
	1-11-10-0	Seminal Vesicle	Treatment	Trannon	1 04+0 10	200	3.66+0.14	1 22±0 1¢b	1.22-0.10	1.24+0.13 <sup>b</sup>	1.31±0.16 <sup>b</sup>	
Organs weight (grams)	Fnididymis	are juins	Post-treatment		1.25+0.07*	8*000-100	1.0/+0.03*	1 26+0 05 <sup>b</sup>	00:01:1	1.13+0.05	1.24+0.07	
Orga	Fini	1	Treatment		0.95+0.06	1 67±0 058	1.07-10.03	1.16+0.07 <sup>ab</sup>	1 25 10 02 ab	1.25±0.03	1.07±0.07 <sup>b</sup>	
2 8	Testes		Post-treatment	20000000	3.30±0.08±	2 89+0 08*a	00:0:0:0	3.63+0.32°	3 87±0 12b	3.0/±0.13	3.85±0.13°	
	Te	Treatment	Heamieni	3 48±0 12	21.0-0.12	3.59+0.07	10000	3.08+0.0/	3 88+0 01*	10:00:0	3.69±0.12	
Group				DW		IF	Me 1	IAIC-I	Mc-10	16.100	MC-100	

Table 14B Changes of absolute weights of testes, epididymis and seminal vesicle at the end of treatment and at the end of post-treatment periods in normal male rats treated with distilled water, testosterone propionate and Mucuna collettii.

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		Seminal vesicle		Post-treatment	3 27+0 31	10:01/4:0	5.23+0.18*a	000 0 00 C	3.79+0.28	3.61+0.27 <sup>b</sup>	3.12±0.17b
		Sem	Teacher	Teament	3 14+0 22		10.40+0.44	3 20±0 23b	3.27 10.32	3.45+0.37 <sup>b</sup>	3.55±0.40b
Absolute weight	granis	Epididymis	Post-treatment	1 Ost-ucalifell	2.91+0.10	84000	3.03+0.09**	2 90+0 09	00:01:0	2.77+0.12*	$2.93\pm0.15$
Absol	(A IV	CD	Treatment		2.90+0.14	A 76±0 108	4.70±0.19	3.17+0.17	1000 p	3.4 /+0.09-	2.92±0.15°
W	estes	1,	Post-treatment	10000	9.18+0.29*	8 23+0 37*	0.0.0.0	8.33+0.68	0 64+0 40	7.04+0.49	9.08+0.35
			1 reatment	10.66.0.22	10.00±0.23	10.20+0.21	10 11 0 20	10.11±0.38	10 74+0 13	2001000	10.04±0.26
Group				DW		IF	Mc-1	I-OIA	Mc-10	Me 100	1910-100

Significant difference compared to at the end of treatment period.

<sup>&</sup>lt;sup>a</sup> Significant difference of TP and Mc groups compared to DW group.
<sup>b</sup> Significant difference of Mc groups compared to TP group.

3.3 Effects of *Mucuna collettii* on histological changes in testes, epididymis and seminal vesicle

### 3.3.1 Histological changes in testes

The testicular histology of all groups at the end of treatment period revealed the intact structure of seminiferous tubules, Leydig's cells, Sertoli cells and spermatogenic cells. The spermatogenesis and spermatogenic lineage were normally found in seminiferous tubules (Figure 19A-E). In addition, the testes treated with TP showed more abundant spermatozoa in the lumen of seminiferous tubules than DW group.

The microanatomy of testes of all groups at the end of post-treatment period presented the intact structure of seminiferous tubules, Leydig's cells, Sertoli cells and spermatogenic cells. The spermatogenesis and spermatogenic lineage were normally developed in seminiferous tubules. The density of spermatozoa was the same in all groups (Figure 19F).

When the testicular tissue study at the end of treatment period groups were compared to post-treatment period groups, it revealed no changes of testicular structure in all groups, except the spermatozoa of TP group at the end of treatment period was more abundant than that at the end of post-treatment period.

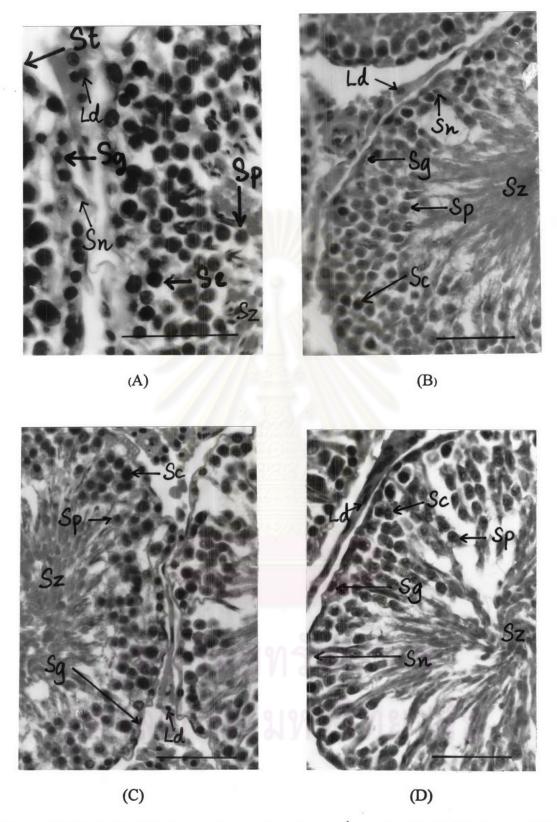


Figure 19 Testicular histology of normal male rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in seminiferous tubules (St), Leydig's cells (Ld), Sertoli cells (Sn), spermatogonia (Sg), spermatocytes (Sc), spermatids (Sp) and spermatozoa (Sz). Bar scale =  $50 \mu m$ 

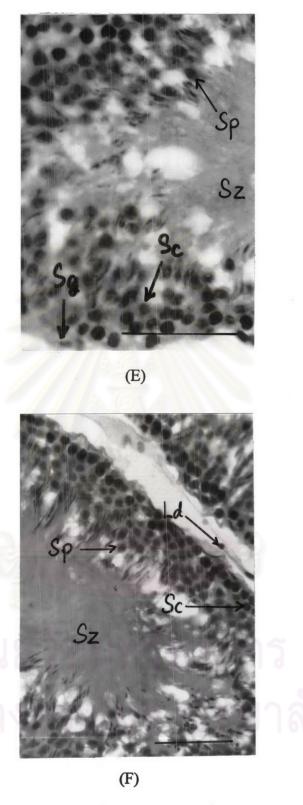


Figure 19 (continued) Testicular histology of normal male rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in seminiferous tubules (St), Leydig's cells (Ld), Sertoli cells (Sn), spermatogonia (Sg), spermatocytes (Sc), spermatids (Sp) and spermatozoa (Sz). Bar scale =  $50 \mu m$ 

### 3.3.2 Histological changes in epididymis

The structure of epididymis of all groups at the end of treatment period demonstrated the cuboidal shape of epididymal epithelial cell lining with stereocilia in head, body and tail regions of epididymis. The diameter of epididymal tubules tended to be wider in the latter regions and the height of epididymal epithelial cell lining tended to be lower. The spermatozoa tended to be abundant in the latter parts of the epididymal tubules. In addition, the numbers of spermatozoa in TP group tended to be higher than DW group (Figure 20A-E).

The epididymal histology of all groups at the end of post-treatment period presented the cuboidal shape of the epithelial cell lining with stereocilia in head, body and tail regions of epididymis and diameter of epididymal tubules tended to be wider in the latter region and the height of epididymal epithelial cell lining tended to be lower. The numbers of spermatozoa were no different between groups.

The epididymal tissue at the end of treatment period groups were compared to at the end of post-treatment period, it showed no differences of the histological structure in all groups, except the numbers of spermatozoa of TP group at the end of treatment period tended to be increased than that of at the end of post-treatment period.

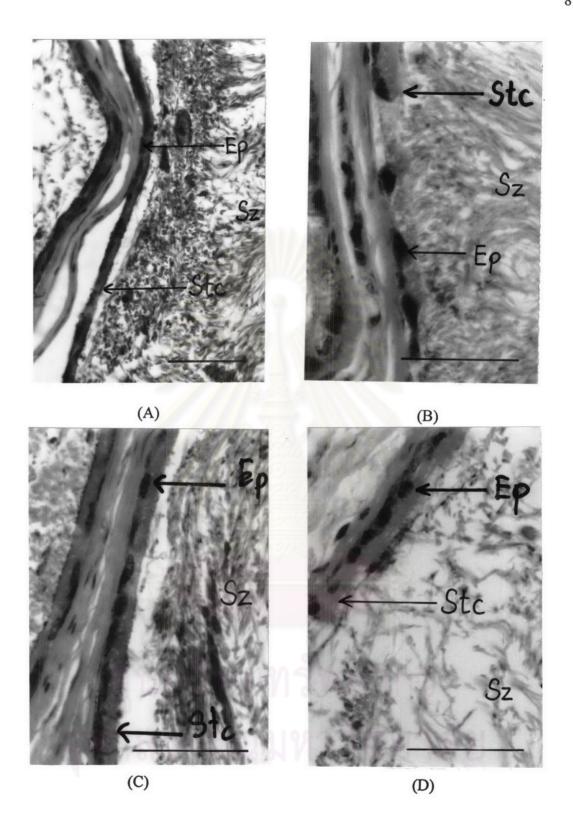


Figure 20 Tail region of epididymis of normal male rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep), spermatozoa (Sz) and stereocilia (Stc). Bar scale =  $50 \mu m$ 

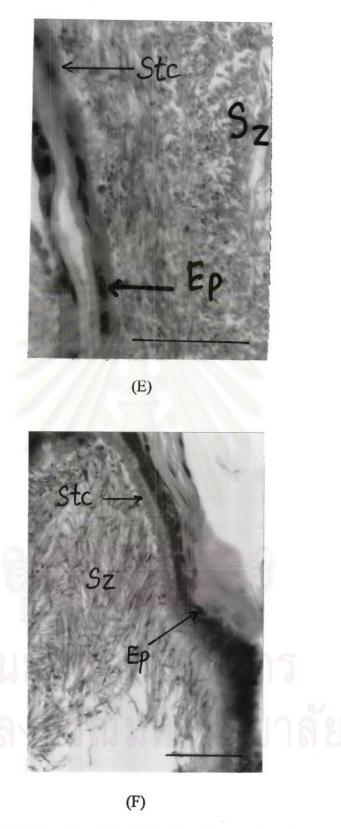


Figure 20 (continued) Tail region of epididymis of normal male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep), spermatozoa (Sz) and stereocilia (Stc). Bar scale = 50 μm

# 3.3.3 Histological changes in seminal vesicle

The seminal vesicle of all groups at the end of treatment period consisted of normal papillary pattern and simple cuboidal shape of the epithelial cell, except for TP group, the papillary of the epithelial cell lining increased folding than DW group (Figure 21A-E).

Seminal vesicle of all groups at the end of post-treatment period appeared to be normal papillary pattern with simple cuboidal shape of the epithelial cell lining, except for TP group, the papillary pattern of the epithelial cell lining consisted of numerous folding and more branching than DW group (Figure 21F).

There were no changes of the seminal vesicle histology between at the end of treatment period and at the end of post-treatment period, except the papillary pattern of the epithelial cell lining of TP group at the end of treatment period consisted of more folding pattern than that of at the end of post-treatment period.

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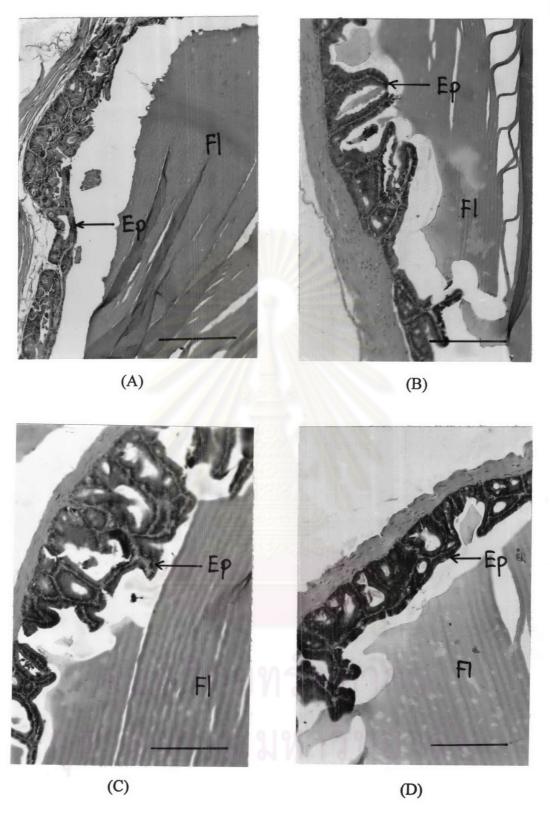


Figure 21 Seminal vesicle of normal male rats treated with distilled water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep) and fluid (Fl). Bar scale =  $200 \mu m$ 

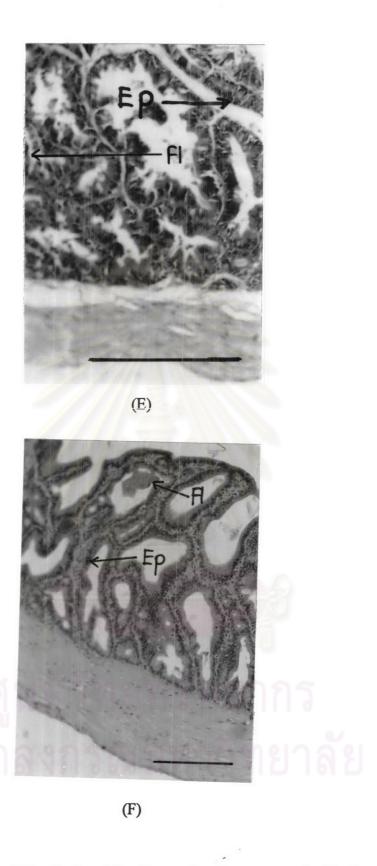


Figure 21 (continued) Seminal vesicle of normal male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep) and fluid (Fl). Bar scale =  $200 \, \mu m$ 

Experiment 4; Effects of *Mucuna collettii* on hormone-related testicular functions and reproductive organs in orchidectomized male rats.

## 4.1 Effects of M. collettii on hormone-related testicular functions

### 4.1.1 Serum testosterone levels

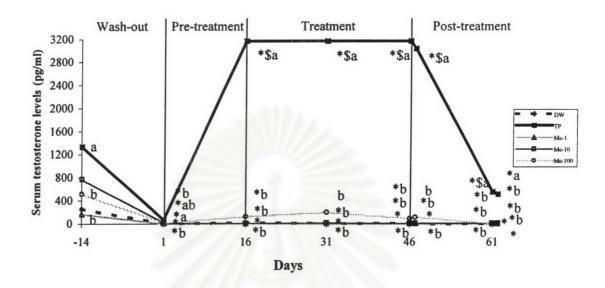
Serum testosterone levels in all 5 groups were significantly dramatically decreased after orchidectomy at D<sub>1</sub> (607.79±210.09 and 21.03±13.20 pg/ml at D<sub>.14</sub> and D<sub>1</sub>, respectively as shown in Figure 22). Serum testosterone levels in male rats after orchidectomy (D<sub>1</sub>) were significantly lower than the normal male rats (21.03±13.20 and 628.97±183.47 pg/ml in ODX rats and normal male rats, respectively). Serum testosterone levels of DW and all Mc groups were kept in the low levels throughout the study periods when compared to the day of ODX levels (D<sub>.14</sub>). No significant differences were found between that 3 periods. In TP group, serum testosterone levels were significantly increased soon after the TP injection (D<sub>16</sub>) and kept high levels throughout the treatment period (D<sub>16</sub>-D<sub>46</sub>). The levels after TP withdrawal for 2 weeks (D<sub>47</sub>-D<sub>61</sub>) were still higher than the level at D<sub>1</sub>.

Comparison to the DW group, serum testosterone levels of all Mc treated groups were no significant differences throughout the study periods, except for Mc-100 group, serum testosterone levels were significantly higher than DW at D<sub>1</sub>. In contrast, serum testosterone levels of TP group were significantly higher since D-14 and kept in the higher levels than that of DW group throughout the study periods.

Comparison to the TP group, serum testosterone levels of all Mc treated groups were significantly lower during  $D_1$ - $D_{62}$ .

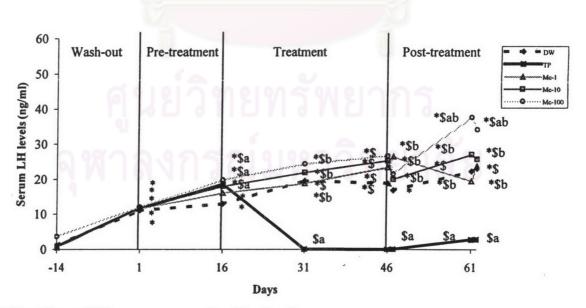


Figure 22 Changes of serum testosterone levels in orchidectomized male rats treated with distilled water, testosterone propionate and *Mucuna collettii*.



- \* Significant difference compared to D<sub>-14</sub> levels.
- \$ Significant difference compared to D<sub>1</sub> levels.
- <sup>a</sup> Significant difference of TP and Mc groups compared to DW group.
- <sup>b</sup> Significant difference of Mc groups compared to TP group.

Figure 23 Changes of serum LH levels in orchidectomized male rats treated with distilled water, testosterone propionate and *Mucuna collettii*.



- \* Significant difference compared to D<sub>-14</sub> levels.
- \$ Significant difference compared to D<sub>1</sub> levels.
- <sup>a</sup> Significant difference of TP and Mc groups compared to DW group.
- <sup>b</sup> Significant difference of Mc groups compared to TP group.

#### 4.1.2 Serum LH levels

In agreement with the low levels of testosterone, serum LH levels in all 5 groups were significantly increased after orchidectomy (1.35±0.58 and 11.63±0.16 ng/ml at D<sub>-14</sub> and D<sub>1</sub>, respectively). Basal serum of LH levels in male rats after orchidectomy (D<sub>1</sub>) were significantly higher than the normal male rats (11.63±0.16 and 0.46±0.09 ng/ml in ODX rats and normal male rats, respectively) as shown in Figure 23. When compared to D<sub>-14</sub>, serum LH levels of DW and all Mc treated groups were gradually increased since D<sub>1</sub> until the last day of study (D<sub>16</sub>-D<sub>62</sub>). Except serum LH levels of DW group at D<sub>16</sub> and D<sub>47</sub> and Mc-1 group at D<sub>16</sub> were no significantly differences. Serum LH levels of TP group were significantly increased at D<sub>1</sub> and D<sub>16</sub> and returned to D<sub>-14</sub> levels at (D<sub>31</sub>-D<sub>62</sub>).

Comparison to the DW group, serum LH levels of Mc-1 group were no significantly differences throughout the study periods. However, serum LH levels of Mc-10 group at  $D_{16}$  and Mc-100 group at  $D_{16}$  were significantly higher than the DW group, these higher levels were agreed with the non-different levels between  $D_1$  and  $D_{16}$  of the DW group. Serum LH levels of TP group were significantly higher only  $D_{16}$ , while the LH levels were significantly lower than the DW group ( $D_{31}$ - $D_{62}$ ).

Comparison to the TP group, serum LH levels of all Mc treated groups were significantly higher after Mc administration for 15 days (D<sub>31</sub>-D<sub>46</sub>) and the higher levels were still remained after the cessation of treatment (D<sub>47</sub>-D<sub>62</sub>).

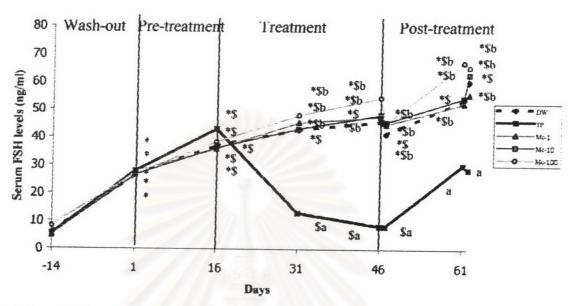
## 4.1.3 Serum FSH levels

In agreement with the high levels of LH, serum FSH levels in all 5 groups were significantly increased after orchidectomy (5.74 $\pm$ 0.65 and 27.15 $\pm$ 0.28 ng/ml at D<sub>-14</sub> and D<sub>1</sub>, respectively as shown in Figure 24). Basal serum of FSH levels (D<sub>1</sub>) in male rats after orchidectomy were significantly higher than the normal male rats (27.15 $\pm$ 0.28 and 6.06 $\pm$ 0.31 ng/ml in ODX rats and normal male rats, respectively). Serum FSH levels of DW and all Mc treated groups after ODX were abruptly increased since D<sub>1</sub> and kept in the higher levels until the last day of experiment (D<sub>62</sub>). Serum FSH levels of TP group were significantly increased at D<sub>1</sub>-D<sub>16</sub>, while the FSH levels were significantly decreased during D<sub>31</sub>-D<sub>47</sub>.

Comparison to the DW group, serum FSH levels of TP group were significantly lower after TP administration for 15 days (D<sub>31</sub>-D<sub>46</sub>) until the study was over (D<sub>47</sub>-D<sub>62</sub>). Serum FSH levels of all Mc treated groups were no significant differences from the DW group throughout the study periods.

Comparison to the TP group, serum FSH levels of Mc treated groups were significantly higher during  $D_{31}$ - $D_{62}$ .

Figure 24 Changes of serum FSH levels in orchidectomized male rats treated with distilled water, testosterone propionate and Mucuna collettii.



<sup>\*</sup> Significant difference compared to D-14 levels.

<sup>b</sup> Significant difference of Mc groups compared to TP group.

<sup>\$</sup> Significant difference compared to D<sub>1</sub> levels.

a Significant difference of TP and Mc groups compared to DW group.

## 4.2 Effects of Mucuna collettii on epididymis and seminal vesicle

When compared the weights and absolute weights of epididymis and seminal vesicle at the end of post-treatment period to the end of treatment period, no significant differences were found in DW and all Mc treated groups (Table 15). In TP group, seminal vesicle weight and its absolute weight at the end of treatment period were significantly higher than at the end of post-treatment period.

Comparison to the DW group, the weights and absolute weights of epididymis and seminal vesicle of Mc treated groups were no significant differences in both periods of treatment. In TP group, the weights and absolute weights of epididymis and seminal vesicle were significantly higher at the end of treatment period and still remained at the end of post-treament periods.

Comparison to the TP group, the weights and absolute weights of epididymis and seminal vesicle of all Mc treated groups were significantly lower than the TP group in both periods of treatment.

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Table 15 Changes of the weights and absolute weights of epididymis and seminal vesicle at the end of treatment and at the end of post-treatment periods in orchidectomized male rats treated with distilled water, testosterone propionate and Mucuna collettii.

Group	ď	Organs weight	weight			Absolute weight	weight	
		(grams)	ms)			$(x 10^{-3})$	grams)	
	Epididymis	lymis	Seminal vesicle	vesicle	Epididymis		Seminal vesicle	vesicle
	Treatment	Post-	Treatment	Post-	Treatment	Post-	Treatment	Post-
	11	treatment		treatment		treatment		treatment
DW	0.26+0.03	0.29+0.05	0.09+0.01	0.08+0.01	0.71+0.82	0.79+0.14	0.24+0.02	0.22+0.02
TP	0.83+0.09ª	0.60+0.04ª	3.17±0.14ª	1.44+0.20*a	2.07+0.20 <sup>8</sup>	1.57+0.15ª	8.04+0.36ª	3.79+0.51*a
Mc-1	0.25±0.04 <sup>b</sup>	0.35±0.02 <sup>b</sup>	0.08±0.01 <sup>b</sup>	0.08+0.10 <sup>b</sup>	0.65+0.09 <sup>b</sup>	0.87+0.05 <sup>b</sup>	0.21+0.03 <sup>b</sup>	0.20+0.02 <sup>b</sup>
Mc-10	0.30±0.05°	0.33±0.01 <sup>b</sup>	0.11±0.01 <sup>b</sup>	0.08+0.00 <sup>b</sup>	0.82+0.13 <sup>b</sup>	0.88+0.05 <sup>b</sup>	0.27+0.04 <sup>b</sup>	0.20+0.01 <sup>b</sup>
Mc-100	0.26±0.03°	0.33±0.02 <sup>b</sup>	0.09±0.01 <sup>b</sup>	0.07±0.10 <sup>b</sup>	0.65±0.09 <sup>b</sup>	0.83±0.05b	$0.21 \pm 0.02^{b}$	$0.18\pm0.02^{b}$
	9			T N N N N N N N N N N N N N N N N N N N				

Significant difference compared to at the end of treatment period.

<sup>&</sup>lt;sup>a</sup> Significant difference of TP and Mc groups compared to DW group.
<sup>b</sup> Significant difference of Mc groups compared to TP group.

4.3 Effects of *Mucuna collettii* on histological changes in epididymis and seminal vesicle

## 4.3.1 Histological changes in epididymis

The epididymal tissue of all groups at the end of treatment period revealed a few number of spermatozoa remaining throughout the regions of epididymis. However, spermatozoa of the TP group showed more density than DW and Mc treated groups. The diameter of head, body and tail of epididymis was narrow and the thickness of simple cuboidal epithelial cell lining with stereocilia was increased, 3-4 layers, but the height of epithelial cell lining tended to be lower (Figure 25A-E).

The histology of epididymis of DW and Mc treated groups at the end of treatment period revealed a few number of spermatozoa remaining throughout the epididymal tubules. The diameter of head, body and tail of epididymis was narrow and the thickness of epithelial lining was increased, 3-4 layers. In contrast, the diameter of epididymis of TP treated group tended to be wider in the certain regions of epididymal tubules, but the height of epithelial cell lining tended to be lower (Figure 25F).

The epididymis of rat at the end oftreatment period groups were compared to at the end of post-treatment period, there were no changes of epididymal tissue in all groups.

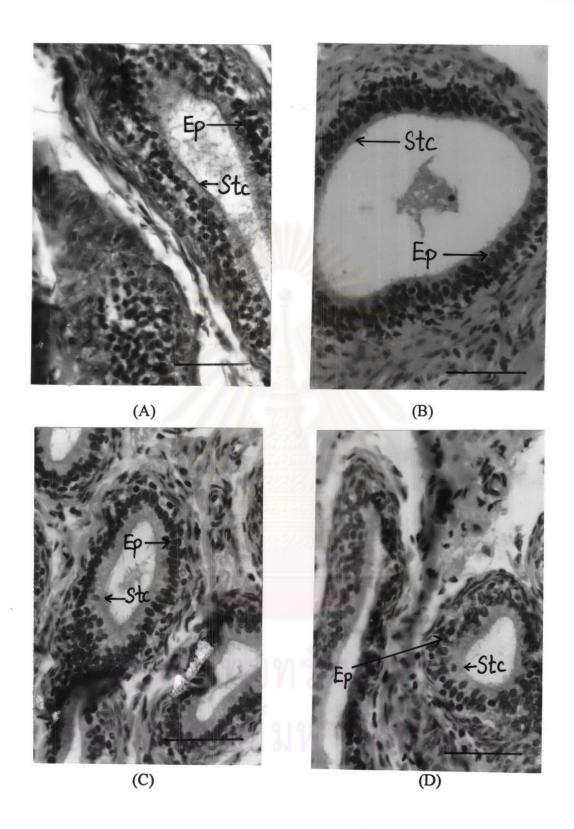


Figure 25 Tail region of epididymis of orchidectomized male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining. Epithelial cells (Ep) and stereocilia (Stc). Bar scale =  $50 \mu m$ 

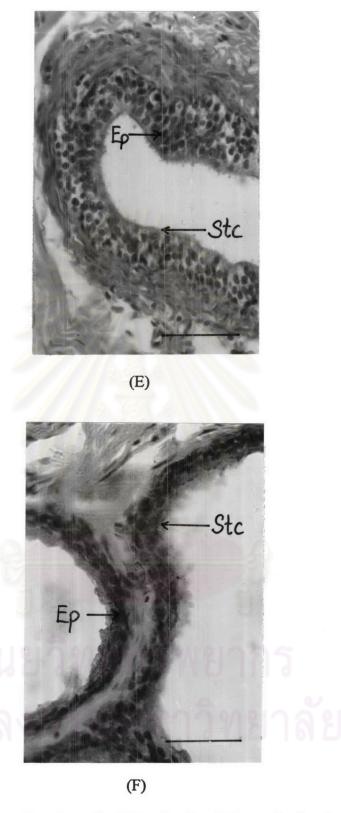


Figure 25 (continued) Tail region of epididymis of orchidectomized male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining. Epithelial cells (Ep) and stereocilia (Stc). Bar scale =  $50 \mu m$ 

## 4.3.2 Histological changes in seminal vesicle

The histology of seminal vesicle of all groups at the end of treatment period showed a simple folding pattern with simple cuboidal epithelial cell lining, except for TP group, the papillary pattern of the epithelial cell lining consisted of the increasing folding and the amount layer of seminal fluid were also higher than DW group (Figure 26A-E).

The seminal vesicle of all groups at the end of post-treatment period also appeared a simple folding pattern with simple cuboidal epithelial cell lining, except the papillary pattern of the epithelial cell lining of TP group was increased folding than DW group (Figure 26F).

There were no changes of the seminal vesicle histology at the end of treatment period when compared to the rat at the end of post-treatment period, except the papillary pattern of the epithelial cell lining of TP group at the end of treatment period was increased folding pattern than that of the rat at the end of post-treatment period.

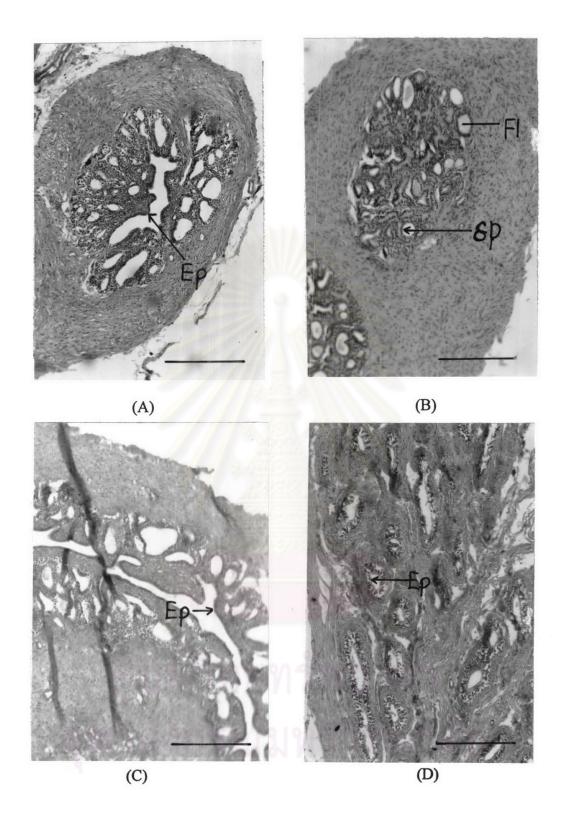


Figure 26 Seminal vesicle histology of orchidectomized male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep). Bar scale =  $200 \, \mu m$ 

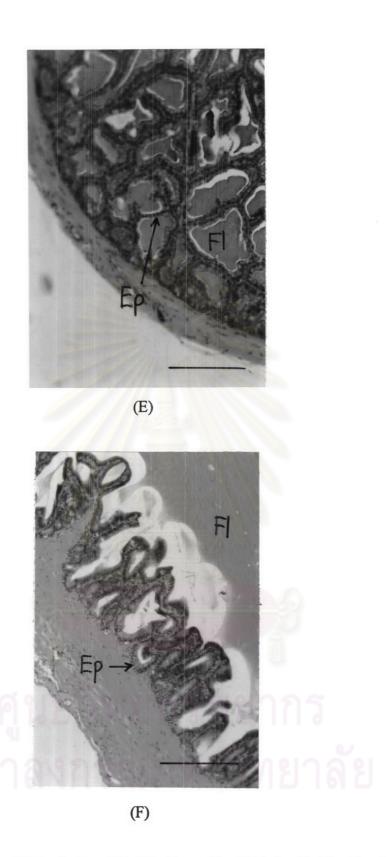


Figure 26 (continued) Seminal vesicle histology of orchidectomized male rats treated with distilled-water (A), Mc-1 (B), Mc-10 (C), Mc-100 (D) and TP at the end of treatment (E) and TP at the end of post-treatment periods (F) with H & E staining in epithelial cells (Ep). Bar scale =  $200 \, \mu m$