



## CHAPTER III

### RESULTS

#### I. Study of progesterone and $\beta$ -estradiol<sub>17</sub> on ovarian development *Penaeus monodon*.

Figure 13 and 14 showing a comparative effect of progesterone and  $\beta$ -estradiol<sub>17</sub> in prawn from different sources. The result indicated that progesterone seemed to induce ovarian development in prawns from Khlong Cone and wild captured prawns, whereas  $\beta$ -estradiol<sub>17</sub> could induce only ovarian development of Khlong Cone stock. An analysis of variance showed a significantly difference among the sources. Khlong Cone stock was easily to induce by hormones than that of Chon Buri and Samut Sakorn stock (Table 3). However, no significant between hormonal source and concentration on ovarian development could be found.

#### Effect of progesterone on ovarian development of *P. monodon*.

The results of progesterone on ovarian development of *P. monodon* in various doses and sources are summarize in Tables 4, 5 and 6.

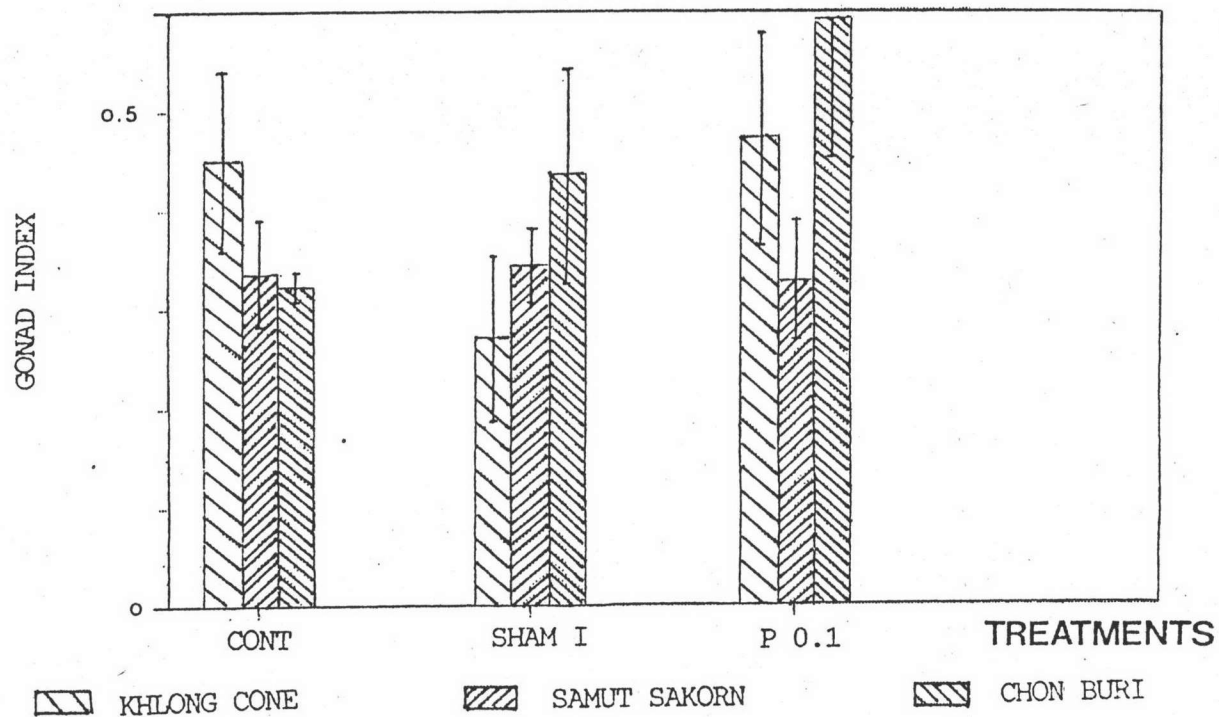


Figure 13. Comparison between 3 sources of *P. monodon* on gonad index by injected progesterone 0.1  $\mu\text{g/g}$  body weight.

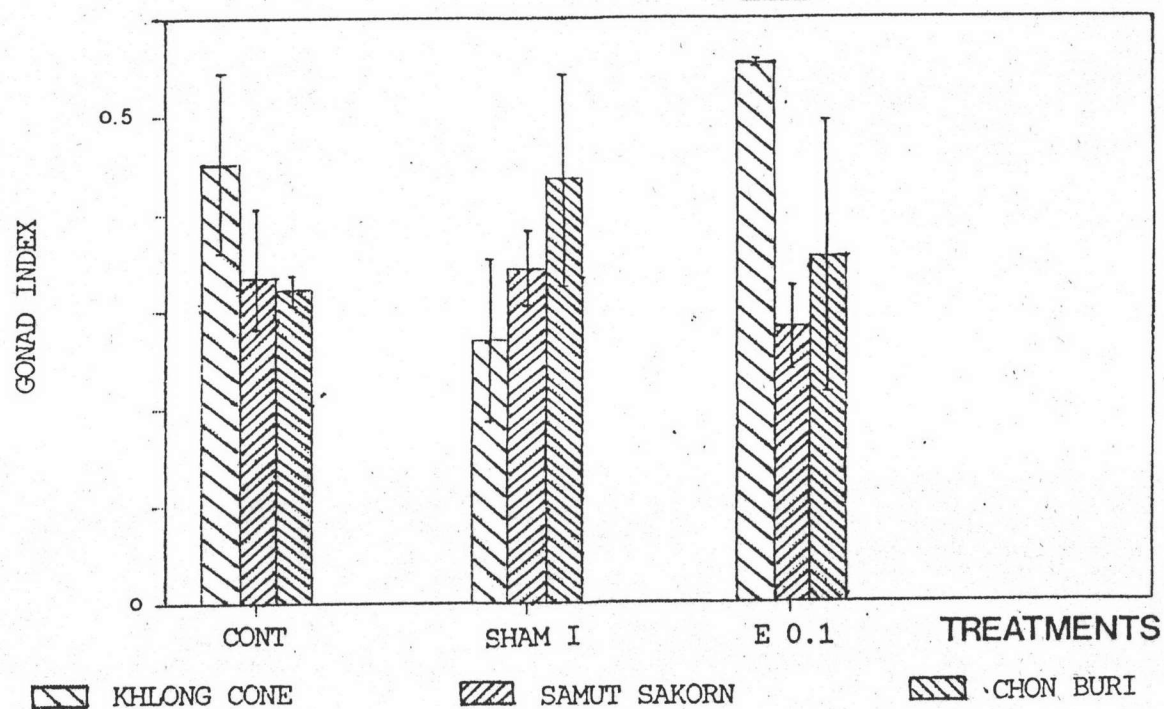


Figure 14. Comparison between 3 sources of *P. monodon* on gonad index by injected  $\beta$ -estradiol17 0.1  $\mu\text{g/g}$  body weight.

Table 3. Effect of sources of broodstock on ovarian development in *P. monodon*.

Sources	mean $\pm$ SD.
A	0.73 $\pm$ 0.60 <sup>a</sup>
B	0.54 $\pm$ 0.60 <sup>ab</sup>
C	0.48 $\pm$ 0.18 <sup>b</sup>

Data presented are means and standard deviation of gonad index from various sources. Significant difference ( $P > 0.05$ ) is shown by the different letter in the column.

A is prawn from Khlong Cone, Samut Songkhram province, B is prawn from Samut Sakarn province, and C is prawn from near shore capture in Chon Buri province.

Table 4. Effect of Progesterone on gonad index of *P. monodon* from Khlong Cone.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.8235 $\pm$ 0.1507	0.4518 $\pm$ 0.1026	0.2776 $\pm$ 0.0571	2.4490 $\pm$ 1.8643
SHAMS I	0.8235 $\pm$ 0.1507	0.2713 $\pm$ 0.0886	0.4861 $\pm$ 0.1293	0.5789 $\pm$ 0.0803
SHAMS II	0.8235 $\pm$ 0.1507	1.0015 $\pm$ 0.5036	0.6349 $\pm$ 0.1414	0.7371 $\pm$ 0.0554
P 0.01	0.8235 $\pm$ 0.1507	0.8782 $\pm$ 0.1176	0.5282 $\pm$ 0.0388	0.8014 $\pm$ 0.2176
P 0.1	0.8235 $\pm$ 0.1507	0.4738 $\pm$ 0.1282	0.3024 $\pm$ 0.0743	0.4382 $\pm$ 0.2314
PII 0.1	0.8235 $\pm$ 0.1507	1.4568 $\pm$ 1.0306	0.7554 $\pm$ 0.1522	0.5800 $\pm$ 0.0140
P 0.2	0.8235 $\pm$ 0.1507	0.5398 $\pm$ 0.0785	0.7537 $\pm$ 0.1522	0.6060 $\pm$ 0.2525
P 0.4	0.8235 $\pm$ 0.1507	0.6442 $\pm$ 0.0017	1.6948 $\pm$ 1.1942	0.6802 $\pm$ 0.0244

Table 5. Effect of Progesterone on gonad index of *P. monodon* from Samut Sakorn.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.3286 $\pm$ 0.0444	0.3356 $\pm$ 0.0504	0.1902 $\pm$ 0.0178	0.4220 $\pm$ 0.1163
SHAMS I	0.3286 $\pm$ 0.0444	0.3440 $\pm$ 0.0383	0.5508 $\pm$ 0.2424	0.7455 $\pm$ 0.2706
SHAMS II	—	—	—	—
P 0.01	—	—	—	—
P 0.1	0.3286 $\pm$ 0.0444	0.3273 $\pm$ 0.0580	0.4404 $\pm$ 0.0787	0.7550 $\pm$ 0.3380
PII 0.1	0.3286 $\pm$ 0.0444	0.8748 $\pm$ 0.0794	0.4564 $\pm$ 0.0626	1.0157 $\pm$ 0.4785
P 0.2	—	—	—	—
P 0.4	—	—	—	—

Table 6. Effect of Progesterone on gonad index of *P. monodon* from Chon Buri.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.5070 $\pm$ 0.0820	0.3212 $\pm$ 0.0233	-	-
SHAMS I	0.5070 $\pm$ 0.0820	0.4362 $\pm$ 0.1583	-	-
SHAMS II	-	-	-	-
P 0.01	0.5070 $\pm$ 0.0820	0.4639 $\pm$ 0.1101	-	-
P 0.1	0.5070 $\pm$ 0.0820	0.5913 $\pm$ 0.1418	-	-
PII 0.1	-	-	-	-
P 0.2	-	-	-	-
P 0.4	-	-	-	-

Figure 15 showing effect of progesterone on gonad development of *P. monodon* from Klong Cone, Samut Songkhram province, it can be concluded that progesterone seems to have no effect on ovarian development. At high concentration ( $0.4 \mu\text{g/g}$  body weight) gonad index tends to increase at day 14. However, at longer acclimation period gonad index begins to decline. The statistical analysis (ANOVA) shows no significant difference of the effect of progesterone concentration and acclimation on gonad development ( $P > 0.05$ ).

In addition, comparison between one and twice hormone injection with control and sham I, II (one solvent injection and twice solvent injection) as shown in Figure 16, indicated that gonad index of P 0.1 and PII 0.1 seems to be higher than sham I and sham II on day 7 and decline in a longer acclimation period. A statistical analysis shows no significantly effect of single or twice injected progesterone on gonad development when compared to control, sham I and sham II.

Prawns from Samut Sakorn, the gonad index of progesterone treatment shown in Figure 17 indicates no significant change ( $P > 0.05$ ) among the treatments and acclimatation period. However, the prawn with twice injected  $0.1 \mu\text{g}$  progesterone/g body weight seems to develop higher degree of gonad index than that of the others, especially at day 7 and day 21 after injection.

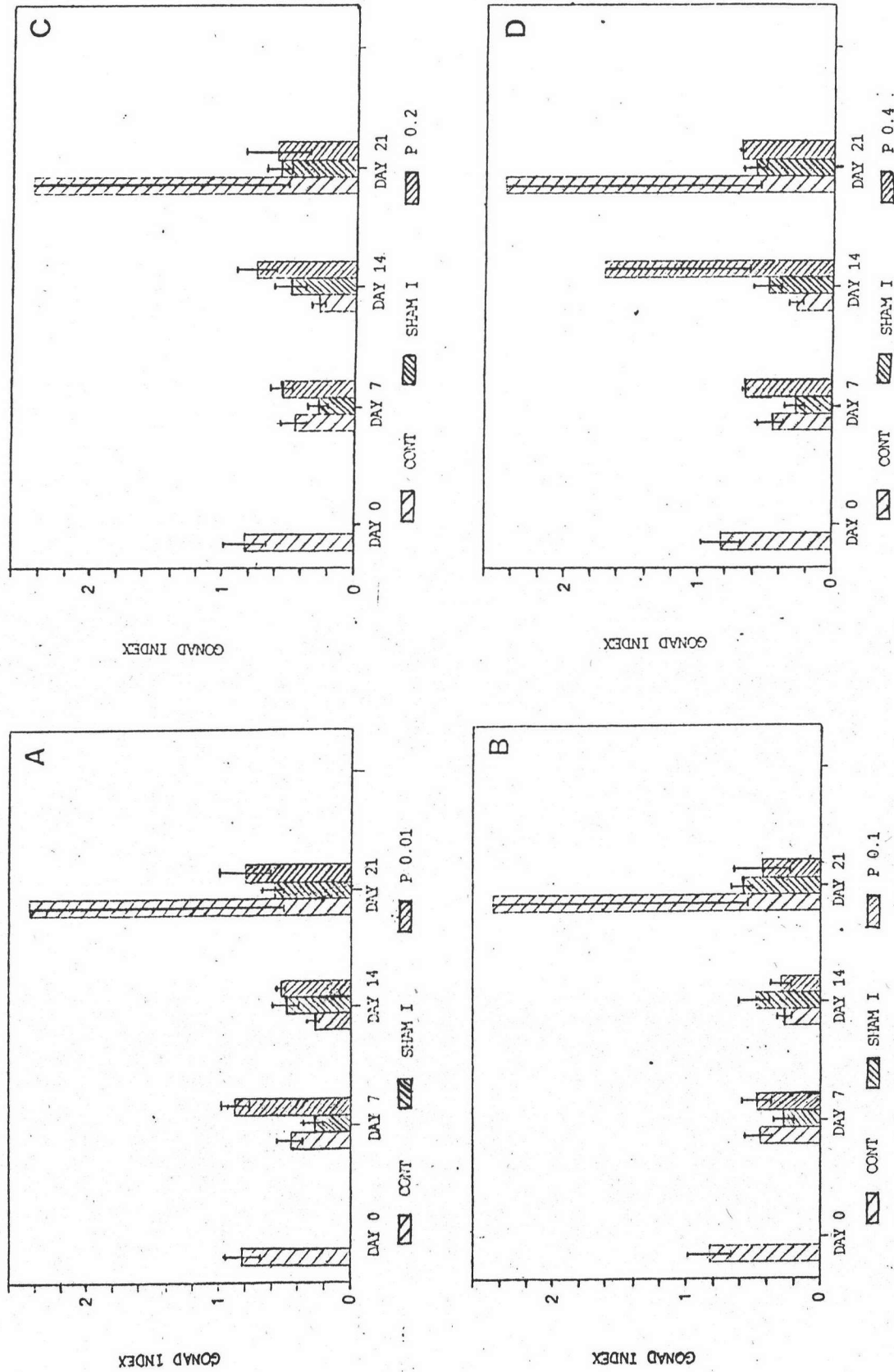


Figure 15. Comparison of ovarian index (%) of *P. monodon* from Khlong Cone in each dose of progesterone injection with control (non injected) and sham (injected with solvent of hormone).  
 (A) Progesterone 0.01 μg/g body weight. (C) Progesterone 0.2 μg/g body weight.  
 (B) Progesterone 0.1 μg/g body weight. (D) Progesterone 0.4 μg/g body weight.



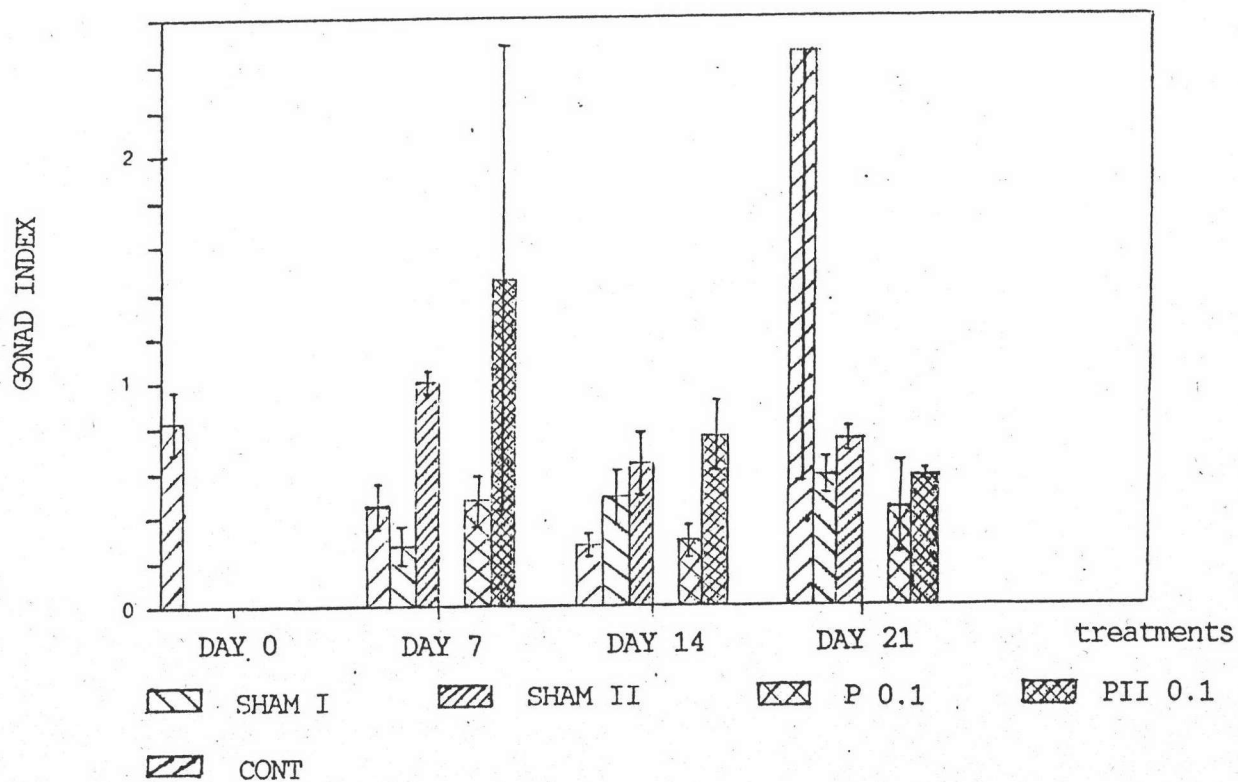


Figure 16. Comparison of gonad index (%) of *P. monodon* from Khlong Cone between 1<sup>st</sup> and 2<sup>nd</sup> progesterone injection (0.1  $\mu$ g/g body weight) with control and 1<sup>st</sup> and 2<sup>nd</sup> injection sham.



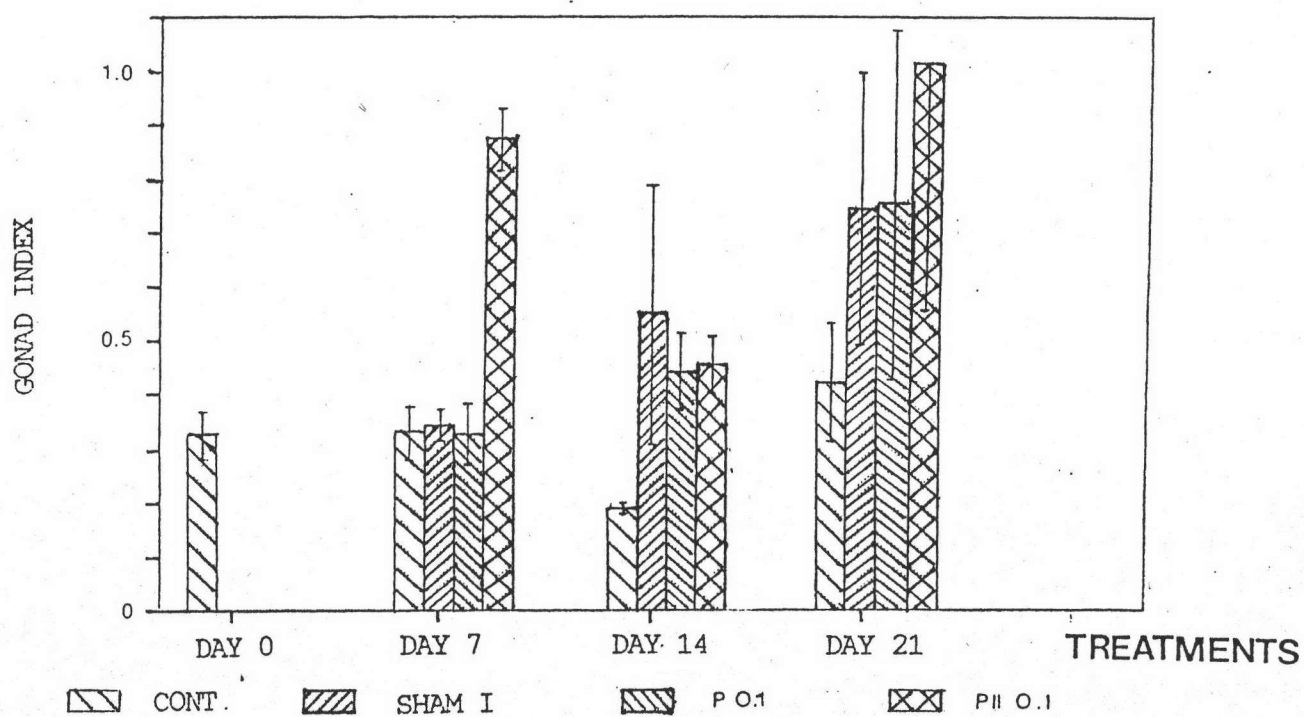


Figure 17. Comparison of gonad index (%) of *P. monodon* from Samut Sakorn between 1<sup>st</sup> and 2<sup>nd</sup> progesterone injection (0.1  $\mu$ g/g body weight) with control and sham.

Prawns from wild (near shore captured) in Chon Buri, the result of progesterone treatment on gonad development is shown in Table 6. It is clear that within 7 days after injecting progesterone, there is no significant change of gonad index among the treatments.

#### Effect of $\beta$ -estradiol17 on ovarian development of *P. monodon*.

The effect of  $\beta$ -estradiol17 on ovarian development of *Penaeus monodon* varied doses, sources and acclimation period are shown in Tables 7, 8 and 9.

Prawns from Khlong Cone, Table 7, gonad index value of control and sham I were compared with each dose of  $\beta$ -estradiol17 which is shown as histogram in Figure 18 A, 18 B, 18 C and 18 D. From Figure 18 indicated that gonad index value of each dose was not differentiated and time was not effect on gonad index. There were not significant different ( $P>0.05$ ) of gonad index among doses and time.

Comparing between control, sham I, sham II, E 0.1 and EII 0.1 as shown in Figure 19, the histogram indicates that the control prawn at day 21 has the highest ovarian development. However, analysis of variance shows no significantly different in ovarian development among the treatments ( $P>0.05$ ).

Table 7. Effect of  $\beta$ -Estradiol17 on gonad index of *P. monodon* from Khlong Cone.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.8235 $\pm$ 0.1507	0.4518 $\pm$ 0.1026	0.2776 $\pm$ 0.0571	2.4490 $\pm$ 1.8643
SHAMS I	0.8235 $\pm$ 0.1507	0.2713 $\pm$ 0.0886	0.4861 $\pm$ 0.1293	0.5789 $\pm$ 0.0803
SHAMS II	0.8235 $\pm$ 0.1507	1.0015 $\pm$ 0.5036	0.6349 $\pm$ 0.1414	0.7371 $\pm$ 0.0554
E 0.01	0.8235 $\pm$ 0.1507	0.5709 $\pm$ 0.1718	0.4125 $\pm$ 0.3175	0.5147 $\pm$ 0.4204
E 0.1	0.8235 $\pm$ 0.1507	0.5527 $\pm$ 0.0077	0.6451 $\pm$ 0.2152	0.6015 $\pm$ 0.1459
EII 0.1	0.8235 $\pm$ 0.1507	0.2796 $\pm$ 0.0702	0.7743 $\pm$ 0.3322	0.6654 $\pm$ 0.0050
E 0.2	0.8235 $\pm$ 0.1507	0.4837 $\pm$ 0.1525	0.7956 $\pm$ 0.0124	0.8401 $\pm$ 0.1362
E 0.4	0.8235 $\pm$ 0.1507	0.7433 $\pm$ 0.3051	0.6358 $\pm$ 0.2097	1.7436 $\pm$ 0.6579

Table 8. Effect of  $\beta$ -Estradiol17 on gonad index of *P. monodon* from Samut Sakorn.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.3286 $\pm$ 0.0444	0.3356 $\pm$ 0.0504	0.1902 $\pm$ 0.0178	0.4220 $\pm$ 0.1163
SHAMS I	0.3286 $\pm$ 0.0444	0.3440 $\pm$ 0.0383	0.5508 $\pm$ 0.2424	0.7455 $\pm$ 0.2706
SHAMS II	-	-	-	-
E 0.01	-	-	-	-
E 0.1	0.3286 $\pm$ 0.0444	0.2842 $\pm$ 0.0420	0.4698 $\pm$ 0.0601	0.7789 $\pm$ 0.0263
EII 0.1	0.3286 $\pm$ 0.0444	0.7525 $\pm$ 0.3935	0.5096 $\pm$ 0.0529	0.7620 $\pm$ 0.1626
E 0.2	-	-	-	-
E 0.4	-	-	-	-

Table 9. Effect of  $\beta$ -Estradiol17 on gonad index of *P. monodon* from Chon Buri.

TREATMENTS	OVARIAN INDEX (MEAN $\pm$ SD)			
	INITIAL	DAY 7TH	DAY 14TH	DAY 21ST
CONTROL	0.5070 $\pm$ 0.0820	0.3212 $\pm$ 0.0223	-	-
SHAMS I	0.5070 $\pm$ 0.0820	0.4362 $\pm$ 0.1583	-	-
SHAMS II	-	-	-	-
E 0.01	0.5070 $\pm$ 0.0820	0.6617 $\pm$ 0.2163	-	-
E 0.1	0.5070 $\pm$ 0.0820	0.3584 $\pm$ 0.1310	-	-
EII 0.1	-	-	-	-
E 0.2	-	-	-	-
E 0.4	-	-	-	-

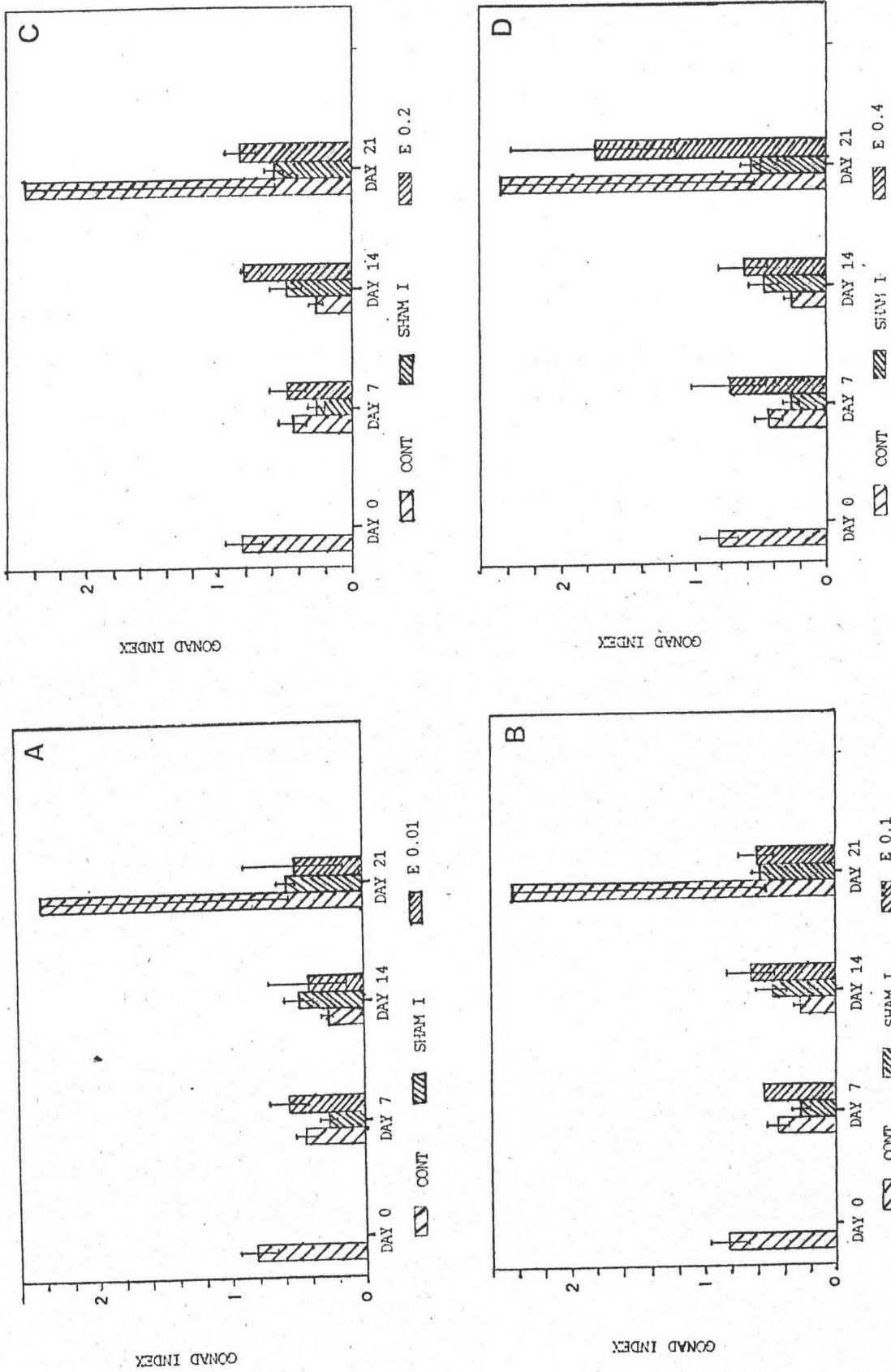


Figure 18. Comparison of ovarian index (%) of *P. monodon* from Khlong Cone in each dose of  $\beta$ -estradiol17 injection with control (non injected) and sham (injected with solvent of hormone).

(A)  $\beta$ -estradiol17 0.01  $\mu$ g/g body weight. (C)  $\beta$ -estradiol17 0.2  $\mu$ g/g body weight.  
 (B)  $\beta$ -estradiol17 0.1  $\mu$ g/g body weight. (D)  $\beta$ -estradiol17 0.4  $\mu$ g/g body weight.

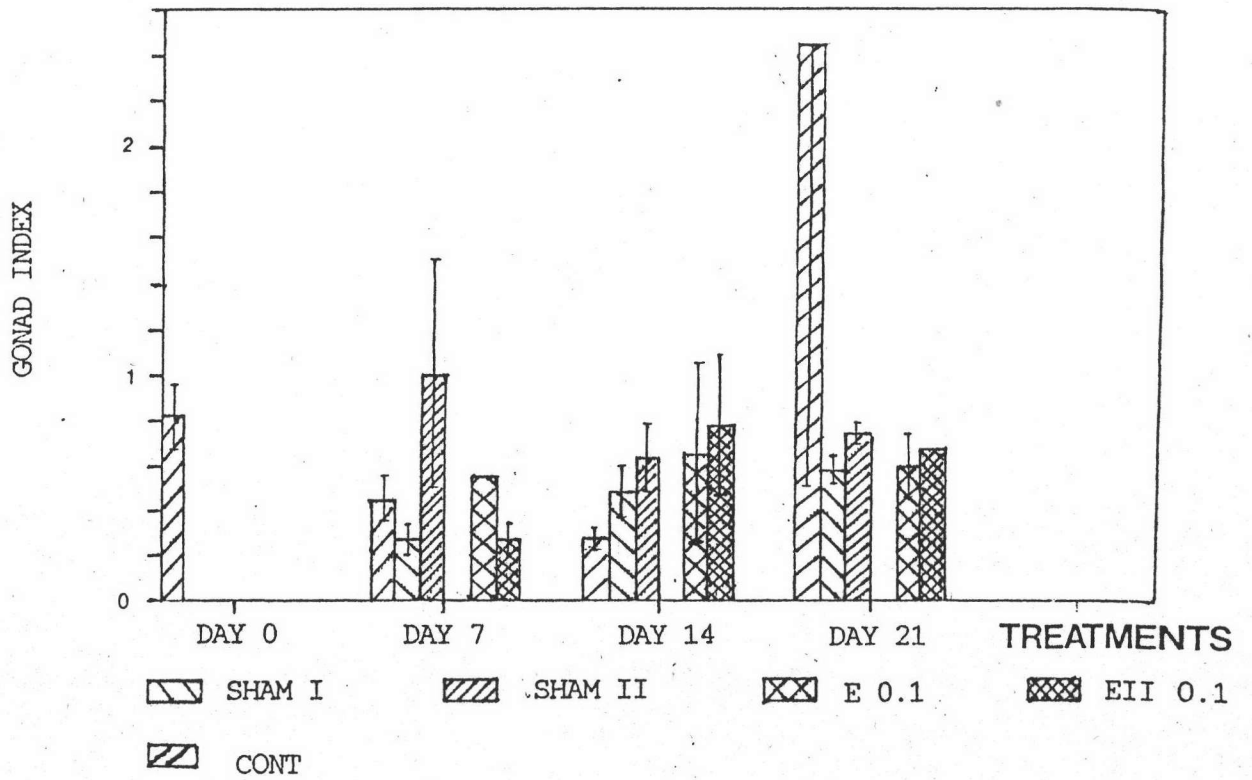


Figure 19. Comparison of gonad index (%) of *P. monodon* from Khlong Cone between 1<sup>st</sup> and 2<sup>nd</sup>  $\beta$ -estradiol17 injection (0.1  $\mu$ g/g body weight) with control and 1<sup>st</sup> and 2<sup>nd</sup> injection sham.

In prawns from Samut Sakorn (Table 8 and Figure 20) the effect of  $\beta$ -estradiol<sub>17</sub> on ovarian development seems to be higher in sham and  $\beta$ -estradiol<sub>17</sub> injected groups than that of the control one, especially at day 14 and day 21. However, the ANOVA shows no significant ( $P > 0.05$ ) of ovarian development among treatments and acclimation periods.

Prawns from Chon Buri, the gonad index value of  $\beta$ -estradiol<sub>17</sub> treated group on day 7 after injection were shown in Table 9. There is no significant difference ( $P > 0.05$ ) among the treatments. However, prawn injected with  $\beta$ -estradiol<sub>17</sub> 0.01  $\mu\text{g/g}$  body weight seem to have highest ovarian development.

## II. Study of 2-deoxyecdysone and $\beta$ -ecdysone on moulting in *Penaeus monodon*.

### Moulting cycle of *Penaeus monodon*.

Moulting cycle are divided to 5 stage such as : intermediate postmoult(A), postmoult(B), intermoult(C), premoult(D) and moult or ecdysis(E). The duration of moult cycle of *Penaeus monodon* 20-40 g. are 18 days. The detail of moulting cycle in *Penaeus monodon* is described in Figure 21.

Normally, the duration from stage B, C and D<sub>1</sub>" to ecdysis were 15 days, 12 days and 8 days, respectively.



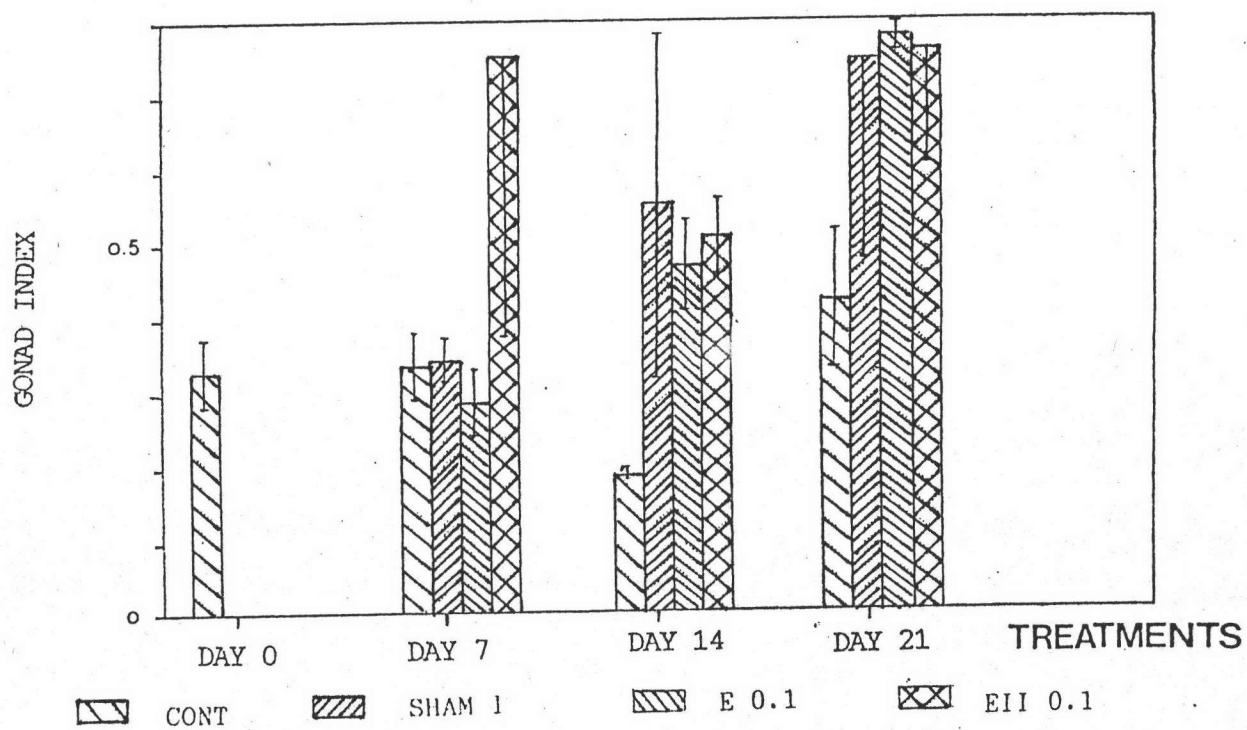


Figure 20. Comparison of gonad index (%) of *P. monodon* from Samut Sakorn in 1<sup>st</sup> and 2<sup>nd</sup>  $\beta$ -estradiol17 injection ( $0.1 \mu\text{g/g}$  body weight) with control and sham.

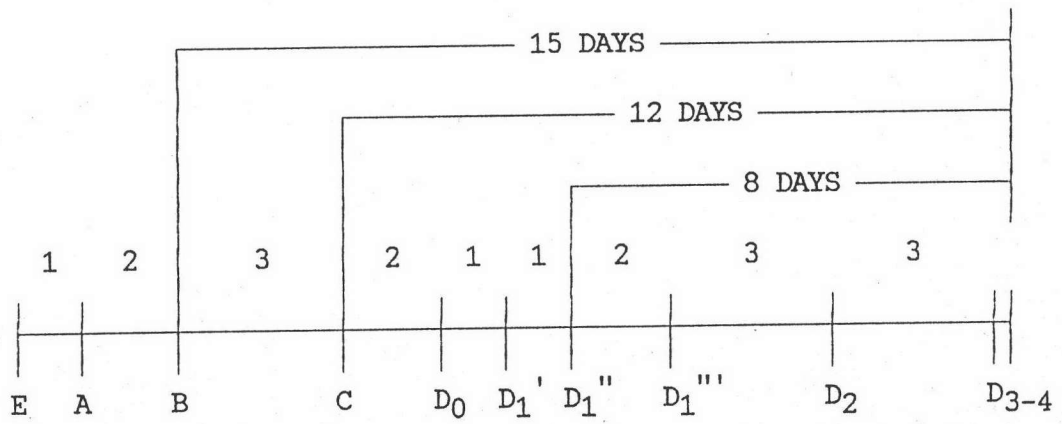


Figure 21. The duration and stages of moult cycle of *P. monodon*  
20-40 g.

The effect of 2-deoxyecdysone and  $\beta$ -ecdysone on moulting of *Penaeus monodon*.

The experiment were studied in moulting stage B, C and D<sub>1</sub>" by injected hormone 2-deoxyecdysone or  $\beta$ -ecdysone doses of 0.01, 0.1, 0.2 and 0.4  $\mu$ g/g body weight. Results of this experiment are shown as Figure 22.

It is clear that either 2-deoxyecdysone or  $\beta$ -ecdysone could induce moulting in B, C and D<sub>1</sub>" stages of moulting. At B-stage, moulting duration decreases from 15 days to average 9.5 days in both hormone-treated group, and to 14 days in sham group. At C-stage, moulting duration decreases from 12 days to average 7.1 days in both hormone-treated group, and to 10 days in sham group. And at D<sub>1</sub>"-stage, moulting duration decreases from 8 days to average 3.8 days in both hormone-treated group, and to 6 days in sham group.

The reduction of moulting duration cycle in each stage of this experiment showing no difference between 2-deoxyecdysone and  $\beta$ -ecdysone (see Table 10). Comparing to level of injected doses, the result shows that either 2-deoxyecdysone and  $\beta$ -ecdysone at doses between 0.01 and 0.1  $\mu$ g/g body weight could provide shortest moulting duration, especially in stage C and D<sub>1</sub>". This indicates that the optimal level of 2-deoxyecdysone and  $\beta$ -ecdysone recommended is not more than 0.01  $\mu$ g-hormone/g body weight.

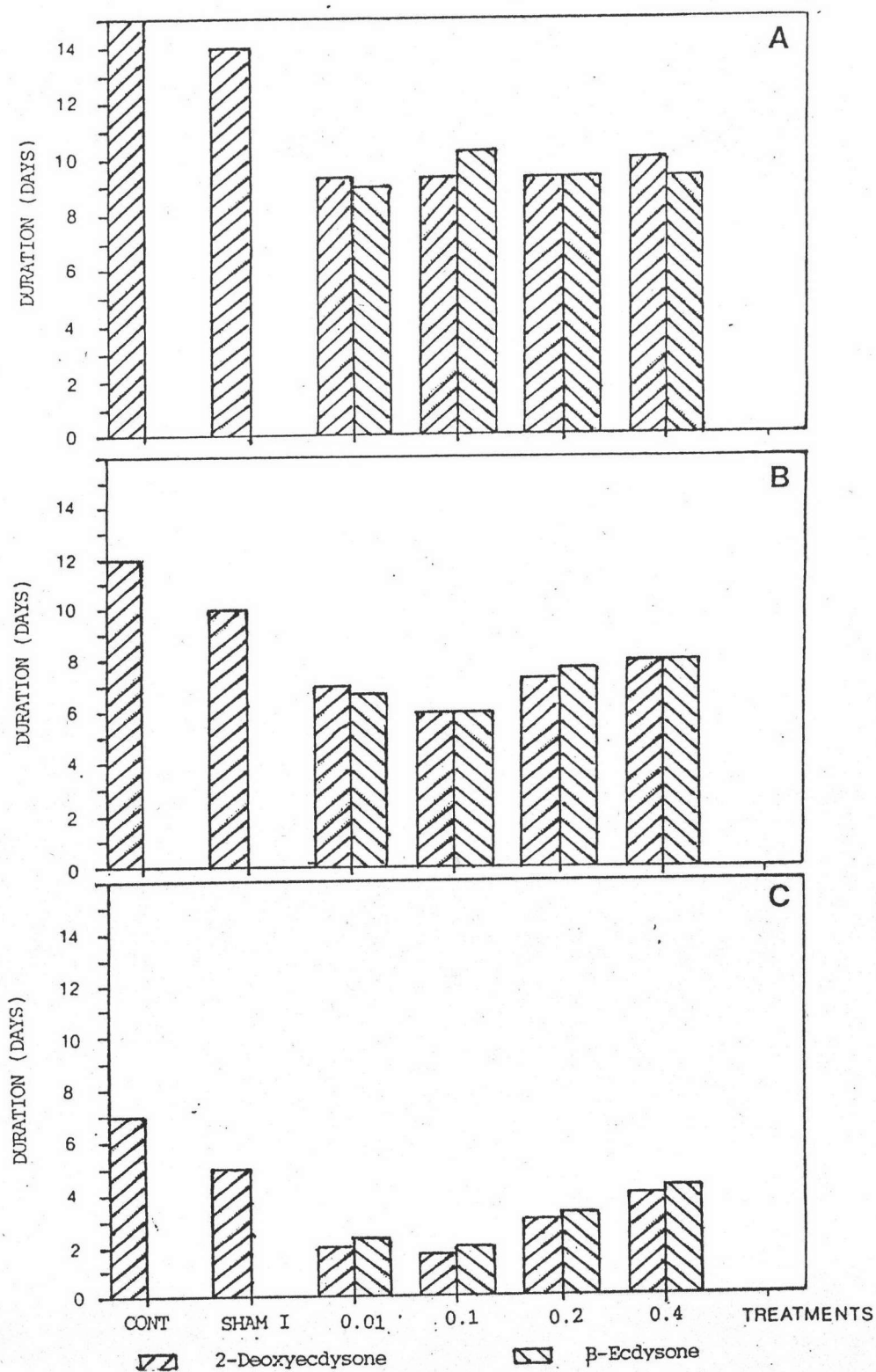


Figure 22. Comparative result of 2-Deoxyecdysone and  $\beta$ -Ecdysone on moulting duration of *P. monodon* with control (non-injected) and sham (injected with non hormonal solvent); (A) Stage B (postmoult), (B) Stage C (intermoult), (C) Stage D<sub>1</sub>" (pre-moult).

Table 10. Effect of 2-deoxyecdysone and  $\beta$ -ecdysone on moulting duration of *P. monodon*

Treatment	M E A N (Days)		
	STAGE B	STAGE C	STAGE D <sub>1</sub> "
Control	15.000 <sup>a</sup>	12.000 <sup>a</sup>	7.000 <sup>a</sup>
Sham	14.000 <sup>ab</sup>	10.000 <sup>ab</sup>	5.000 <sup>b</sup>
Ec 0.01	9.333 <sup>c</sup>	7.000 <sup>ba</sup>	2.000 <sup>c</sup>
BEc 0.01	9.000 <sup>c</sup>	6.667 <sup>c</sup>	2.333 <sup>bc</sup>
Ec 0.1	9.333 <sup>c</sup>	6.000 <sup>c</sup>	1.667 <sup>c</sup>
BEc 0.1	10.333 <sup>bc</sup>	6.000 <sup>c</sup>	2.000 <sup>c</sup>
Ec 0.2	9.333 <sup>c</sup>	7.333 <sup>bc</sup>	3.000 <sup>bc</sup>
BEc 0.2	9.333 <sup>c</sup>	7.667 <sup>bc</sup>	3.333 <sup>bc</sup>
Ec 0.4	10.333 <sup>bc</sup>	8.000 <sup>bc</sup>	4.000 <sup>b</sup>
BEc 0.4	9.333 <sup>c</sup>	8.000 <sup>bc</sup>	4.333 <sup>b</sup>

Data are presented as mean of moulting duration from various treatments. Significant differences ( $P < 0.05$ ) are indicated by different letter in each column.