Bulletin of Taiwan Fisheries Research Institute No.47, 1989.

荷爾蒙促進石斑魚成熟及排卵之研究

葉信利・丁雲源・郭欽明

Induced Final Maturation and Ovulation of Grouper (*Epinephelus salmonoides*, *Epinephelus fario*), with HCG or LH-RH Analogue

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For the purpose of solving the shortage and uncertain supply of mature fish from the wild, research efforts have been directed at induced breeding aimed at achieving self-sufficiency in the supply of mature grouper. Some hormone therapies, HCG, LH-RHa, Estrogen by injection and implantation methods were tested to determine the best treatment for induced final maturation and ovulation in grouper. The results were summarized as follows:

1. Female spawners (about three years old) of *Epinephelus fario* containing the mature oocytes at tertiary yolk globule stage, with the initial mean oocyte diameter of about 400-600 um, were injicted with human chorionic gonadotropin at the dosage 1000 IU/Kg body weight, to ensure successful spawning. Two or three dosages were usually injected with 24 hours interval. Spawning occurred about 48-96 hours after the first injection.

2. Spawners could remature and be induced to spawn naturally by injection with HCG in a dosage of 500-1000 IU/Kg body weight. Two or three injections 24 hours apart were found aduquate, when the females were reared in pond for one year.

3. It is apparent that age must be given considerable importance when attempting to induce maturation and ovulation of *Epinephelus fario*.

4. HCG and progesterone proved to be effective in inducing ovulation for breeding of *Epinephelus fario*.

5. The time of spawning depended upon the frequency of hormone injection and the sexual maturity was indicated by the oocyte development stage and the initial oocyte diameter and number of injections.

6. Based on the implantation study, luteinizing hormone-releasing hormone analogue (LH-RHa)

was the most effective hormone therapy for iducing maturation of the groupers *Epinephelus fario, E. salmonoides.*

7. The dosages were compared to determine the one most effective for induced maturation through hormone implantation. The results gave the range 10 mg/kg to 70 mg/kg body weight of LH-RHa.

8. The success of induced maturation through the implantation method depended upon the environmental cue and sexual maturity, indicated by the species, age, oocyte development stage, season, hormone, dosage and preparation of pellet.

前　言

台灣水產養殖業，經過十幾年來努力之開發，已有驚人之發展成就，並已逐漸傾向高經濟價值之魚種。石斑魚爲食用魚中高級經濟魚類，爲台灣今後發展鹹水養殖漁業之主要魚種之一。但石斑魚苗無論在國內或國外，均非常缺乏，一直是其無法大量推廣養殖之主因。

目前石斑魚雖然已完成許多人工繁殖及育苗工作[4][5][6]，然因其是屬於先雌後雄之兩性魚（Protogynous hermaphroditism）[8][9]，且成熟之種魚是屬於高齡魚[10]，雌魚在3齡以上，雄魚則需在8至11齡以上，捕獲甚爲困難，同時由於魚體碩大，人工繁殖之操作實爲不便，而且依賴天然種魚，來源不定受自然因素影響太大，所以惟有充足成熟種魚來源，始可預期石斑魚養殖之成果。

爲解決石斑成熟種魚來源之問題，就是能在魚塭大量育成成熟親魚，要育成雄魚則需要先經過提早促進性轉變之階段，其方法也被建立[11][12][13]，尤其是最近使用促進轉變之埋植法（Implantation）更是經濟簡便。而雌魚則是如何促其成熟爲重點，然而石斑魚在此方面之研究不多。而調節魚類的生殖作用，除環境因素（cue）影響外[15][16][17]，利用荷爾蒙刺激也是一個方法（Lam,1982,1984），又埋植法（Implantation）最近應用於魚類促進成熟及繁殖已有許多成功之報告[20][21][22][23][24][25][26][27][28][29]，而且效果佳，特別是在虱目魚之研究上，成果斐然。LH-RHa爲最近廣爲研究及應用之荷爾蒙，在鯉魚、鮭魚、香魚、臭都魚、鱸魚、虱目魚……等都有很好催熟及促使產卵效果。而HCG爲沿用已久之促進產卵之荷爾蒙，其效果也不容否認。在以往荷爾蒙皆採用注射方式，做短期性之促進目的用，而近來改採埋植方式，效果頗佳，其催熟作用時機可提前及延長作用時效。又雌性素（Estrogen）能刺激肝臟產生卵黃素（vitellogenin），進入卵細胞促使卵發育，特別是當魚類卵黃粒（yolk granule）或卵黃球（yolk globule）形成時，對魚類之卵黃蓄積作用（vitellogenesis）有影響。

所以，本試驗乃以目前台灣所養殖之主要二種石斑魚，青點石斑（*Epinephelus fario*）與鮭形石斑（*Epinephelus salmonoides*）爲對象，利用注射（injection）或埋植（Implantation）方式，使用LH-RHa，HCG，Estrogen 等荷爾蒙，針對不同年齡，發育及季節，探討荷爾蒙促使石斑魚成熟之方法，冀能找出最適荷爾蒙種類、處理時機、劑量與時間，以期早日能培養出大量成熟石斑魚。

材料與方法

材料：

1985年及1986年實驗用之石斑魚爲魚塭養成3至5齡之青點石斑（*Epinephelus fario*）28尾，平均體重4.08±0.18公斤，平均體長63.36±1.37公分，另鮭形石斑（*Epinephelus salmonoides*）爲1+至3齡之魚，平均體重 2.53±0.18 公斤，平均體長53.55±1.29公分。1987年所使用者爲4至5齡左右之青

點石斑16尾，平均體重4.88±0.31公斤，平均體長63.12±2.58公分；鮭形石斑則爲2齡左右者19尾，平均體重2.16±0.15公斤，平均體長50.85±公分。1988 年之使用魚有二，一爲前三年（1985－1987年）實驗持續所培育之石斑魚，青點石斑有15尾，魚齡在4至7齡間，鮭形石斑有7尾，魚齡在5齡左右，平均體重5.40±0.58公斤，平均體長70.06±3.03公分。另一爲1987年10月新購自魚塭養成接近3齡之青石斑33尾，經越冬飼育後，平均體重3.83±0.13公斤，平均體長 62.69±0.56公分。這些試驗用魚主要皆購自漁民養 成上市體型之石斑魚，再經一段時間馴育後當試驗材料，其來源可以大量取得。

方法：

從1985年10月至1988年7月分別使用注射（Injection）及埋植（Implantation)方式，將荷爾蒙以水溶液或藥粒（pellet）狀送入魚體內讓其吸收。

一.注射法：

選擇成熟雌性石斑魚，以其卵粒大小及卵黃蓄積程度爲準[2][12]，在卵巢內多數卵粒達到第三卵黃球期（ Tertiary yolk stage）或卵徑在400μm以上時，方選爲試驗魚，以胎盤性腺激素（human chorionic gonadotropin，HCG）及血清性腺激素（Pregnant mare serum gonadotropin PMSG），行背肌注射，促進卵巢最後成熟，達到排卵之目的，以了解排卵之過程，建立有效荷爾蒙種類及所需劑量。

二.埋植法：

不同年齡及不同性腺發育程度之石斑魚，以2－phenoxyethanol 300 ppm 麻醉後，再將含有荷爾蒙之藥粒 (pellet)，行肌肉植入方式（Intramuscular implantation），將藥粒植入，每個藥粒直徑爲2mm，長度則以所需劑量多寡不定，（藥粒製法及操作方式，於另篇報告詳細說明）[32]，藥粒（pellet）長度與重量之關係，因所含之荷爾蒙成份比例不同，而有不同。埋植時以魚體單邊植入爲主，並每尾魚以不同顏色塑膠軟管於胸鰭基底做標識，以利區別及追蹤

所使用藥粒（pellets）以所含荷爾蒙成份不同，分別爲α－Tocopherol（V.E）；LH －RHa；Estrone（E1）；β－Estradiol（E2）；PU（puberogen）＋V.E（α－Tocopherol)；PE（peamex）＋V.E；E2＋V.E，PU＋E2＋V.E；PE＋E1＋V.E，PU＋PE＋V.E等10種，其中Puberogen及Peamex以 IU爲單位，每次製做以總劑量250mg爲準，Puberogen 每次爲5000IU，Peamex 同爲5000IU；V.E，E1，E2所使用粘劑cocoa butter以不超過總劑量之5%。E2＋V.E兩者比例則爲1:4。LH－RHa則參照Lee（1986）[27]之製法。

另以不使用荷爾蒙而採用同埋植操作方式之處理魚，及不處理之魚爲對照組。並以組織學研究性腺發育狀況，以探討埋植法之應用功效與各種荷爾蒙對石斑魚之催熟排卵機制，以建立有效荷爾蒙種類及所需劑量與最簡易之處理方法。

結　果

一.催熟與產卵試驗：

青點石斑（*Epinephelus fario*）從1985年12月至1988年7月，以HCG（human chorioric gonadotropin）之類的荷爾蒙進行促進最後成熟（final maturation）及排卵（Ovulation），使用劑量約由500IU／kg魚體重，至2000IU／kg魚體重間，其結果如表1至3所示。

1.1985年及1986年：

由表1知，1985年及1986年成熟之青點石斑，體重在3.5至4.8公斤間，平均卵徑450至730μm，以每針劑量3,500至8000IU，HCG（puberogen 日本製）注射處理或另加250IU，PMSG（Peamex 日本製）處理2至3次，12尾石斑魚中有8尾順利產出卵，卵爲透明性卵，卵徑800至900μm，有一大油球200μm。另成熟度與爾蒙注射次數對產卵之影響，注射1針者2尾，效果不佳；卵徑600μm以上注

表1　1985年12月及1986年4月至7月青點石斑荷爾蒙催熟與產卵試驗

Table 1　Summary of hormonal treatments for induced ovulation trials on *Epinephelus fario* in 1985-1986.

Trial No.	Body Wt. (Kg)	Age	Date	Injection dose (IU)	Specific dose (IU/Kg BW)	IOD (mm)	No. of eggs spawned (1000)	Remarks
1	3.24	3	12/12			.45-.80		
			12/24	3000PU	925PU	.45-.80		
			12/26	2500PU	772PU			
			03/06					Eggs degenerated
2	4.40	4	04/25	4000PU	909PU	.47		
			04/30			.80-.90		Over-ripe (spawned)
3	3.60	3+	04/25	4000PU	1111PU	.57		
			04/30					Eggs partially released, OD:.85-.90
4	4.45	4	05/03	4000PU+250PE	899PU+56PE	.52-.60		
			05/04	4000PU+250PE	899PU+56PE	.50-.60		
			05/06	4000PU+250PE	899PU+56PE	.70		
			05/07				400	Pelagic eggs Naturally spawned, OD: .85-.90
			05/08					Over-ripe
5	4.25	4	05/07	4500G	1059G	.45-.55		
			05/08	4500G+250PE	1059G+59PE	.49-.57		
			05/09	4500G+250PE	1059G+59PE			
			05/14					Eggs degenerated
6	4.80	4+	05/07	5000PU	1042PU	.53-.55		
			05/08	5000PU+250PE	1042PU+52PE	.53-.55		
			05/09	5000PU+250PE	1042PU+52PE			Fish demaged and died
7	4.52	4	05/27	4500PU	996PU	.53-.73		
			05/28	4500PU	996PU	.50-.60		
			05/29				370	Stripped
			05/30					Died
8	3.50	3+	05/27	3500PU+250PE	1000PU+71PE	.60		
			05/28	7000PU	2000PU			
			05/30				400	Naturally spawned
9	3.60	3+	05/27	3500PU+250PE	972PU+69PE	.60-.70		
			05/28	3500PU	972PU			
			05/29				400	Stripped
			05/30					Died
10	3.55	3+	06/03	3500PU+250PE	986PU+70PE	.42-.50		
			06/04	7000PU	1972PU			
			06/06					Did not spawn
11	3.50	3+	06/05	3500PU+250PE	1000PU+71PE	.50-.53		
			06/06	3500PU+250PE	1000PU+71PE	.50		
			06/07	25 mg PG	7.1 mg PG	.53		
			06/09				400	Naturally spawned
12	4.78	4	07/08	5000PU+250PE	1046PU+52PE	.49-.51		
			07/09	8000PU	1674PU	.80-.90		
			07/10	5000PU			330	Naturally spawned
			07/11				349.5	Same
			07/12				380	Same

* PU : Puberogen (trade name) ; PE : Peamex (trade name); PG : Progesterone; IOD : Initial oocyte diameter ; OD : Oocyte diameter in mm.

表 2　1987 年青點石斑荷爾蒙催熟及產卵試驗

Table 2　Summary of hormonal treatments for induced ovulation trials on *Epinephelus fario* in 1987.

Trial No.	Body Wt. (Kg)	Age	Date	Injection dose (IU)	Specific dose (IU/Kg BW)	IOD (mm)	No. of eggs spawned (1000)	Remarks
1	4.65	4	06/10	5000PU	1075PU	.37-.43		
	4.30		07/29	4000PE	930PE	.37-.41		
			07/30	4000PE	930PE	.37		
			07/31			.33-.40		
2	5.20	4	06/13	5000PU+250PE	961PU+48PE	.39-.47		
			06/14	5000PU+250PE	961PU+48PE	.45-.50		
			06/15	5000PU+250PE	961PU+48PE	.50-.67		
			06/16	5000PU+250PE	961PU+48PE	.73-.88		
			06/18	25 mg PG	4.8 mg PG	.73-.88		Did not spawn
3	6.10	5+	06/13	6000PU	984PU	.50-.55		
			06/14	6000PU	984PU	.40		Eggs partially Over-ripe
			06/15	6000PU	984PU	.40-.58		
			06/16			.40		Eggs partially Over-ripe
4	5.40	4	06/13	5500PU	1018PU	.44-.50		
			06/14	5500PU	1018PU	.47-.52		
			06/15	5500PU	1018PU	.68-.80		
			06/16	5500PU	1018PU	.85		
			06/18				841	Naturally spawned OD : .90
5	5.54	4+	06/13	5500PU	993PU	.42-.51		
			06/14	5500PU	993PU	.42-.60		
			06/15	5500PU	993PU	.40-.45		
			06/16	5500PU	993PU	.56-.60		
			06/18	20 mg PG	3.6 mg	.40-.90		Did not spawn
6	4.30	4	06/13	4000PU+250PE	930PU+58PE	.43-.50		
			06/14	4000PU+250PE	930PU+58PE	.47-.80		
			06/15	4000PU+250PE	930PU+58PE	.50-.60		
			06/16	4000PU+250PE	930PU+58PE	.83-.90		
			06/18			.90		Over-ripe
7	5.20	4	06/25	5000PU	962PU	.42-.55		
			06/26	5000PU	962PU	.50-.60		
			06/27	5000PU	962PU		579	Naturally spawned OD:.80
			06/28				333.7	OD:.90
			06/29	25 mg PG	4.8 mg		82.5	OD:.90
			06/30				266.9	Died
8	4.80	4	06/26	5000PU	1005PU	.44-.57		
			06/27	5000PU	1005PU	.50-.57		
			06/28				75	Naturally spawned OD: .80
			06/29	5000PU	1005PU		90	OD: .90
9	4.55	4	06/26	4000PU+500PE	880PU+110PE	.42-.50		
			06/27	4000PU+1000PE	880PU+220PE	.37-.47		
			06/29	4000PU+1000PE	880PU+220PE	.33		
10	5.20	4	06/26	5000PU	961PU	.45-.50		
			06/27	5000PU	961PU	.42-.90		
			06/30	25 mg PG	4.8 mg PG		755.2	Naturally spawned OD : .90
11	2.70	2+	06/26	3000PU	1111PU	.48-.60		
			06/27	3000PU	1111PU	.50-.57		
			06/29	3000PU	1111PU	.84		
			06/30	3000PU	1111PU		474.1	Naturally spawned OD: .90
			07/01				33.3	OD: .90
12	5.70	4+	07/29	5000PU	878PU	.40-.41		
			07/30	5000PU	878PU	.32-.37		
			07/31					Eggs degenerated

* PU: Puberogen (trade name); PE: Peamex (trade name); PG: Progesterone; IOD : Initial oocyte diamter ; OD: Oocyte diamter

表 3　1988 年青點石斑荷爾蒙催熟及產卵試驗

Table 3　Summary of hormonal treatments for induced ovulation trials on *Epinephelus fario* in 1988.

Trial No.	Body Wt. (Kg)	Age	Date	Injection dose (IU)	Specific dose (IU/Kg BW)	IOD (mm)	No. of eggs spawned (1000)	Remarks
1	3.15	3	05/11	3000PU	952PU	.43-.47		
			05/12	3000PU	952PU	.55		
			05/13	3000PU	952PU	.90		
			05/14				46.7	Naturally spawned OD:.90
	3.45	3	06/29	4000PU	1160PU	.48-.50		
			06/30	4000PU	1160PU	.53-.63		
			07/01	4000PU	1160PU	.80		
			07/02	4000PU	1160PU		250	Naturally spawned OD:.90
			07/03				34.8	Stripped OD:.90
			07/04					Eggs degenerated
2	3.52	3+	05/11	3500PU	997PU	.40-.43		
			05/12	3500PU	997PU	.48-.53		
			05/13	3500PU	997PU	.87		
			05/14				47.5	Naturally spawned OD:.90
			05/16				11.8	Same
	3.86	3+	07/11	4000PU	1037PU	.45-.52		
			07/13	5000APL	1295APL	.65-.72		
			07/14			.80		
			07/15				80.9	Naturally spawned OD:.90
			07/16					Eggs degenerated
3	3.20	3	05/11	3500PU	1094PU	.12-.30		
	3.42		07/11	3000PU	877PU	.45-.55		
			07/13	3000PU	877PU	.52-.70		
			07/14	1000PU+1000PE	292PU+292PE		114.7	Naturally spawned and stripped; OD:.90
			07/16					Eggs degenerated
4	3.40	3	05/11	4000PU	1176PU	.52-.55		
			05/12	3500PU	1029PU	.47-.53		
			05/13	3500PU	1029PU	.70		
			05/16				7.2	Naturally spawned OD:.90
			05/17				79.4	Same
			05/18					Eggs degenerated
	3.30		06/30	3000PU	909PU	.52-.56		
			07/01	3000PU	909PU	.40-.45		
			07/02	3000PU	909PU	.45		
			07/03	3000PU	909PU	.47		
			07/04	3000PU	909PU	.47		
			07/07	4000PU	1212PU	.50-.52		
			07/08	4000PU	1212PU	.45-.52		
			07/09	4000PU	1212PU	.45-.50		
			07/13			.42		Eggs degenerated
5	3.50	3+	05/13	3500PU	1000PU	.43-.45		
	3.70		06/30			.36-.38		
	3.82		07/11			.37-.42		

*PU: Puberogen (trade name); PE: Peamex (trade name); PG: Progesterone;
IOD: Initial oocyte diamter; OD: Oocyte diamter in mm; APL: Chorionic gonadotropin U.S.P.

表3 續
Table 3 Continued.

Trial No.	Body Wt. (Kg)	Age	Date	Injection dose (IU)	Specific dose (IU/Kg BW)	IOD (mm)	No. of eggs spawned (1000)	Remarks
6	3.80	3+	05/13	3500PU	921PU	.47-.63		
	3.70		06/30	4000PU	1081PU	.42-.48		
			07/01	4000PU	1081PU	.41-.48		
			07/02	4000PU	1081PU			
			07/03	2000PU	541PU	.90	11.6	Naturally spawned OD:.90
			07/05				24.6	Same
			07/07			.80		Eggs degenerated
7	2.20	3	05/13	2500PU	1136PU	.47-.51		
			05/16	2500PU	1136PU	.85-.87		
			05/17				91.5	Naturally spawned OD:.90
			05/18				2.1	Same
	2.50		06/30	3000PU	1200PU	.43-.48		
			07/01	3000PU	1200PU	.48-.52		
			07/02	3800PU	1200PU	.80-.85		
			07/03	2000PU	800PU	.80	170	Naturally spawned OD:.90
			07/05				21.6	Same
			07/07					Eggs degenerated
8	4.46	3+	05/16	4500PU	1000PU	.43-.47		
			05/18	5000PU	1121PU	.40		
			07/11			.35-.38		
9	3.80	3+	06/11	2000PU	526PU	.46-.52		
			06/14	4000PU	1052PU	.42		
			06/15	4000PU	1052PU	.43-.50		
			06/20	4000PU	1052PU	.44-.46		
			06/21	4000PU	1052PU	.48-.43		
			06/22	4000PU	1052PU	.50		
			06/24					Eggs degenerated
10	4.40	3+	06/29	4000PU	909PU	.30-.47		
			06/30			.53		
			07/11			.80-.90		Over-ripe
11	4.50	3+	06/30	4000PU	889PU	.47-.54		
			07/01	4000PU	889PU	.40		
			07/02	4000PU	889PU			
			07/03	4000PU	889PU			Over-ripe
			07/07					Eggs degenerated
12	4.22	3+	07/11	4000PU	948PU	.48-.50		
			07/13	4000PU	948PU	.45-.70		
			07/14	2000APL	474APL		138.2	Naturally spawned OD:.90
			07/15				50	Same
13	4.50	3+	07/11	5000PU	1111PU	.40-.42		
			07/13	5000APL	1111APL	.42-.68		
			07/15				87	Naturally spawned OD:.90
			07/16				4.5	Same
14	3.70	3+	07/11	4000PU	1081PU	.45-.50		
			07/13	4000PU	1081PU	.47-.80		
			07/14	2000PU	541PU		115.7	Naturally spawned OD:.90
			07/15				14.8	Same
			07/16				32.8	Same
15	4.90	4	07/11	5000PU	1020PU	.45		
			07/13	5000PU	1020PU	.55-.78		
			07/14	2000APL	408APL		211	Naturally spawned OD:.90
			07/15				88	Same
			07/16	2000APL	408APL			Eggs degenerated

*PU: Puberogen; PE: Peamex; PG: Progesterone; IOD: Initial oocyte diamter
OD: Oocyte diamter in mm; APL: Chorionic gonadotropin U.S.P.

表3 續
Table 3 Continued.

Trial No.	Body Wt. (Kg)	Age	Date	Injection dose (IU)	Specific dose (IU/Kg BW)	IOD (mm)	No. of eggs spawned (1000)	Remarks
16	4.72	4	07/11	4000PU	847PU	.45-.80		
			07/13	4000PU	847PU	.47-.76		
			07/14	2000PU	424PU		150	Naturally spawned OD:.90
			07/16	1000APL	212APL		57.3	Same
			07/18					Eggs degenerated
17	8.40	7	05/24	8000PU	952PU	.37-.65		
			05/26	8000PU	952PU	.53-.80		
			05/29				250	Naturally spawned OD:.90
18	9.00	7+	05/24	8000PU	889PU	.37-.40		
			05/26	8000PU	889PU	.35-.43		
19	7.10	6	05/31	7000PU	986PU	.43		
			06/02	7000PU	986PU	.80-.90		
			06/04				527.4	Naturally spawned OD:.90
			06/05				39.2	Same
20	7.85	6+	06/10	8000PU	1019PU	.48-.75		
			06/13	4000PU	510PU		1607.7	Naturally spawned OD:.90
			06/15			.60-.70		
			06/20			.75-.80		Over-ripe
21	6.55	6	06/10	6000PU	916PU	.48		
			06/13	6000PU	916PU	.50-.55		
			06/15	6000PU	916PU	.80-.90		Didnot spawn
			06/20					Eggs degenerated
22	5.72	5	06/10	3000PU+1000PE	524PU+175PE	.48-.50		
			06/13	6000PU	1049PU	.50-.80		
			06/15	6000PU	1049PU	.40		
			06/20					Eggs degenerated
23	4.50	4	06/10	2000PU+1000PE	445PU+223PE	.51		
			06/13	2000PU+1000PE	445PU+223PE			
			06/15	6000PU	1333PU	.48		
			06/20					Eggs degenerated
24	5.80	5	06/10	2000PU+1000PE	345PU+173PE	.45-.48		
			06/13	6000PU	1034PU	.58-.60		
			06/15	6000PU	1034PU	.67		
			06/17	6000PU	1034PU	.85		Died
25	8.40	7	06/10	2000PU+1000PE	238PU+119PE	.60		
			06/13			.40-.50		
			06/15					Eggs degenerated
26	6.50	5+	06/11	3000PU	462PU	.42-.48		
			06/13	6000PU	924PU	.58-.62		
27	8.15	7	06/11	4000PU+2000PE	491PU+245PE	.42		
			06/14	4000PU+2000PE	491PU+245PE	.50-.60		
			06/15	4000PU+2000PE	491PU+245PE	.56		
			06/20	8000PU	982PU	.51-.60		
			06/21	8000PU	982PU	.50-.60		
			06/22	8000PU	982PU	.55		
			06/24	8000PU	982PU	.50-.80		
			06/27					Eggs degenerated
28	7.40	6+	06/11	4000PU+3000PE	541PU+405PE	.48-.51		
			06/14	4000PU+3000PE	541PU+405PE	.65-.80		
			06/15	4000PU+3000PE	541PU+405PE	.80-.90		Pre-ovulation
			06/20					Eggs degenerated

*PU: Puberogen; PE: Peamex; PG: Progesterone; IOD: Initial oocyte diamter
OD: Oocyte diamter in mm.; APL: Chorionic gonadotropin U.S.P.

射2針者5尾，除一尾未產卵，一尾因季節關係外，餘皆採得卵。其中2尾隔日死亡，死因可係強迫擠卵而受傷。卵徑600μm以下者注射3針爲5尾，3尾順利產出卵，卵數皆超30萬粒，其中一尾打第2針後已產出約33萬粒，立即補打第3針，則24小時內再排出約35萬粒卵。注射HCG或混合使用PMSG皆能達到產卵。使用劑量以每約1000IU／kg魚體重，間隔24小時注射，若只打2針者，則第2針劑量加倍，其效果並無明顯差異。

2.1987年試驗

青點石斑在1987年之體重試驗魚爲2.78至6.10公斤間，平均卵徑370至600μm，此批魚爲去年培育所留下之魚。以每尾魚接受HCG（Puberogen）1000IU／1kg魚體重左右之劑，或另加Peamex 250IU處理2至4次，12尾魚中有4尾魚順利產卵，產卵數17萬粒至120萬粒左右。另2尾魚卵過熟，1尾卵萎縮退化，餘有些之卵只進行生殖泡移動（Germinal vesicle migration）及卵徑增大（表2）而已。在使用荷爾蒙方面，此次添加Peamex之魚有3尾，但皆未有產卵現象，其中1尾最初卵徑爲390至470μm，另2尾卵徑爲420至500μm。另有4尾魚在注射HCG後間隔24小時又注射1針Progesterone，其中3尾沒反應，1尾產出卵。而產出卵者係已先產出卵，再接受注射後續又排出卵，可現Progesterone之效果不顯著。

3.1988年之結果

1988年之成熟青點石斑，分爲二種類型，一爲3齡左右者14尾，體重在2.2至4.5公斤間，另一爲4至7齡魚14尾，體重在4.5至9.0公斤間（表3）。3齡魚卵徑分布在400至630μm間，以Puberogen注射每尾1000IU／kg魚體重，或添加Peamex（1尾），另有2尾各打1針APL（chorionic gonadotropin，U.S.P)，14尾魚中有9尾經注射後順利排出卵，3尾魚卵萎縮，2尾魚沒反應。產卵魚中又有3尾經第一次產卵後，經1個半月至2個月時間，又再度成熟，並經注射後又產卵（表3）。試驗魚中，產卵數最爲3萬5千粒最多爲33萬粒。使用之荷爾蒙以HCG爲主，以Peamex配合效果同樣顯著，在注射次數上，以2至3針效果最佳，超過4針者，往往使卵粒退化（表3）。注射間隔以第1針與第2針間差24小時爲佳，第2針以後，則時間可稍延長再注射第3針較具彈性，可視需採卵時機而定。

4至7齡魚14尾（表3），卵徑分布爲370μm至750μm間，同樣以HCG注射，或添加Peamex，以每尾魚500IU／kg及1000IU／kg魚體重之劑量處理，結果有5尾順利產卵，產卵數最少21萬，最多160萬左右，另3尾沒反應，1尾死亡，5尾卵退化。在年齡差別上以4齡魚產卵尾數最佳有2尾，其次6，6＋,7齡各1尾魚。在注射間隔上，以第1針與第2針間隔48小時效果較好，6尾中有4尾產卵，2尾沒反應；而間隔72小時的8尾中，只有1尾產卵，餘6尾卵退化，一尾死亡。在荷爾蒙種類使用上，添加Peamex效果不顯著（表3），6尾中除1尾達到預排卵期（Pre－ovulation）外餘卵皆退化。

4.三年催熟產卵之比較：

1985年至1988年青點石斑之催熟產卵試驗結果比較如表4至8所示。試驗中以1988年之B群（3齡魚）催熟後產卵數百分比最高，達64.3%，其次爲1986年及1987年，而以1988年之A群魚（4至7齡魚）較差（表4）。反之卵退化情形則以1988年A群魚35.7%最高，沒有反應則以1987年33.3%爲最。

由年齡分布來看催熟產卵數之結果（表5），青點石斑2＋魚齡左右之魚就可成熟，而以3至4齡魚成熟者較多，進行催熟產卵也以階段之成功率較高，約佔50%以上。

荷爾蒙之種類與催熟產卵之效果如表6所示，以HCG之Puberogen爲主劑，添加Peamex或以APL或Progesterone或Gona. 取代之效果，以Puberogen單獨使用便可達到催熟與產卵之目的，約佔54%，至於添加Peamex之效果不定，約有30.8%，而以APL取代Puberogen效果亦佳，Progesterone對末期排卵有作用，Gona．效果不顯。

注射次數與催熟產卵之關係如表7所示，注射4針以下者，能使石斑魚產卵，反之若打4針以上者，幾乎卵未能產出。注射次數中又以打2針者之效果最顯，3針次之。而且在注射1至3針之魚中，於第1次產卵後，立即補打1針，續產卵者，1針組有1尾，2針組6尾，3針組5尾。

打針次數與第1針注射後幾日產卵之關係如表8，打1針者在3日及5日後產卵，打2針者集中在2，3

表 4　青點石斑催熟產卵數百分比之年度比較

Table 4　Precentage of results of hormonal treatments for induced ovulation trials on *Epinephelus fario* in different years (1985-1988).

YEAR / Results		1985-1986	1987	1988 A	1988 B
SPAWNED	Naturally	41.6%	41.6%	35.7%	64.3%
	Stripped	16.7%			
Eggs degenerated		8.3%	8.3%	35.7%	7.1%
Over-ripe		8.3%	16.7%		14.3%
Did not-spawn8		8.3%	33.3%	21.4%	14.3%
Died		8.3%		7.1%	
Total no. of fish		12	12	14	14

A: Fish of ages 4-7　　B: Fish of age 3.

表 5　青點石斑催熟產卵數之年齡比較

Table 5　Results of hormonal treatments for induced ovulation trials on groupers(*Epinephelus fario*)of varions ages.

AGE / Results		2+	3	3+	4	4+	5	5+	6	6+	7	7+	Total
SPAWNED	Naturally	1	4	8	8				1	1	1		24
	Stripped			1	1								2
Eggs degenerated			1	1	2	1	1			1	2		9
Over-ripe				2	2			1					5
Did not spawn				3	3	1		1	1			1	10
Died						1	1						2
Total no. of fish		1	5	15	16	3	2	2	2	2	3	1	52

，4日後產卵，打3針者在3，4，5日產卵，打4針者在5日後產卵，亦即通常集中在打完最後1針之隔日至後2天產卵，亦即此段時間可以採卵。

二.埋植催熟試驗：

青點石斑及鮭石斑從1985年10月至1988年6月，以含不同荷爾蒙成分之藥粒處理，埋植劑量由3.024mg／kg至71.800mg／kg魚體重，其結果如下所述。

1.1985年及1986年青石斑埋植試驗：

16尾青點石斑魚齡爲2＋至5＋，魚體重爲2.80公斤至6.25公斤，魚體長55.2公分至74.4公分，所使用荷爾蒙爲LH－RHa，Estrone，β－Estradiol，α－Tocopherol＋Puberogen等5種，埋植劑量

表 6　荷爾蒙種類對青點石斑催熟產卵數之影響

Table 6　Results of various hormone treatments for induced ovulation trials on *Epinephelus fario* in 1985-1988.

Results	Hormone	PUBER.	PEAM.	APL.	GONA.	PROG.	PU+PE	G+PE
SPAWNED	Naturally	20		3		2	4	
	Stripped	1					1	
Eggs degenerated		7		1	1		4	1
Over-ripe		3					1	
Did not spawn		5	1			3	3	
Died		1						1
Total no. of fish		37	1	5	1	5	13	2

*PUBER. : Puberogen (from Japan); PEAM.: Peamex (from Japan)
APL.: Chorionic gonadotropin U.S.P.;
GONA. : Chorionic gonadotropin (from chinese)
PROG. : Progesterone; PU+PE : Puberogen + Peamex;
G+PE : GONA. + Peamex

表 7　注射次數對青點石斑催熟產卵數之影響

Table 7　Results of hormonal treatments for induced ovulation trials on *Epinephelus fario*, by number of times of injection, in 1985-1988.

Number of times of injection / Results	1	2	3	4	5	6	7	8
SPAWNED Naturally	2(1)*	14(6)*	10(5)*	1				
SPAWNED Stripped		2						
Eggs degenerated	1	2	4			1	1	1
Over-ripe	1		1	2			1	
Did not spawn	1	4	3		2			
Died			1	1				
Total no. of fish	5	22	19	4	2	1	1	1

*() : Number of times the fish spawned after continuing injection.

表8　荷爾蒙注射次數對青點石斑產卵時間之影響

Table 8　The effect of frequency of hormonal injection for induced ovulation on the spawning time of *Epinephlus fario* in 1985-1988.

Days after 1st injection / Number of Injections	2	3	4	5	Total
1		1		1	2
2	5	6	5	1	17
3		4	3	2	9
4				1	1
Total no. of fish	5	11	8	5	29

爲9.424mg／kg至71.800mg／kg魚體重，結果如表9所示。在1985年之三尾魚中，催熟效果不明，但對於藥粒有吸收現象。在1986年4月所試驗埋植LH－RHa之青點石斑4尾，則有明顯使卵徑增大，以及部份卵進行生殖泡遷移（Germinal vesicle migration）現象，其中有1尾再使用HCG注射後順利產出卵（表9）。在5月至7月所做之試驗，則使魚之卵退化掉。另於11月埋植之6尾魚中，於次年（1987年）之6，7月所做之追蹤，埋植PU＋V.E中有1尾卵已進入第三卵黃期，另1尾爲周邊仁期（perinucleolar stage），另對照組及空白組則仍只處於卵母細胞及周邊仁期而已。所以埋植LH－RHa及PU＋V.E對於催熟有正面效果。

2.鮭形石斑之1986年埋植試驗

由1986年4月至11月進行22尾埋植試驗，魚齡在1＋至3齡間，魚體重在1.05公斤至3.60公斤間，體長42.0公分至61.3公分。埋植劑量爲10.437mg／kg至43.851mg／kg魚體重，使用荷爾蒙有HH－RHa，β－E2，PU＋E2＋V.E，E2＋V.E，PU＋V.E，另有空白組及對照組（表10）。使用LH－RHa者在1至2個月後卵皆進化至卵原細胞（Oogonia stage）及周邊仁期。使用β－E2者在一個月後亦停留在卵原細胞期，其餘處理之魚，則卵細胞仍處於未發育期，對照組及空白組亦同，可見LH－RHa及β－E2對於鮭形石斑仍有其催熟作用，而PU＋E2＋V.E，E2＋V.E及PU＋V.E則於此實驗效果有待研究，而且埋植劑量多寡對結果亦無明顯差異。

3.石斑魚1987年之埋植試驗

青點石斑爲2至5齡魚4尾，體重在2.20公斤至6.80公斤體長52.4公分至75.5公分。鮭形石斑爲2齡左右之魚19尾，體重爲0.9公斤至2.93公斤，體長在40.2至57.2公分間。埋植之荷爾蒙，青點石斑爲V.E＋E1＋PE，劑量爲8.030mg／kg及16.272mg／kg魚體重。鮭形石斑則埋植有V.E，V.E＋E2，V.E＋PE，V.E＋E2＋PU，LH－RHa及E1等6種，劑量爲5.040 mg／kg至21.053 mg／kg魚體重。經埋植後7個月，青點石斑性腺發育仍未發現明顯發育。鮭形石斑也於6個月後性腺發育同樣未能有明顯差異，在7個月後情形亦同，可說是埋植催熟效果皆不顯著。（表11）

4.1988年石斑魚埋植試驗

從5月至6月青點石斑有20尾，年齡主要爲3齡左右的有17尾，4至6齡3尾，魚體重爲2.70公斤至8.40公斤，體長爲56.6公分至81.3公分，使用荷爾蒙爲V.E＋PU＋PE，V.E＋PU，V.E＋PE，LH－RHa等4種，植入劑量從3.024 mg／kg至18.815 mg／kg魚體重。而鮭形石斑有7尾，魚齡爲3及5齡左右，體重從2.54公斤至6.60公斤，體長53.7公分至75.8公分，使用荷爾蒙爲V.E＋PE及V.E ＋PU＋

表 9　1985年及1986年青點石斑之催熟埋植試驗

Table 9　Results of hormonal pellet implantation on the grouper (*E. fario*) in 1985-1986.

Date*	Species**	Age	BW (Kg)	BL (cm)	Oocyte diameter (mm)	Hormone type	Dose (mg/Kg BW)	Results
Oct.26,1985	E.f.	3	3.50	61.0	.15	ESTRONE	15.571	
						LH-RHa	9.429	
Oct.26,1985	E.f.	4	5.00	70.5	.20	LH-RHa	66.667	
Oct.28,1985	E.f.	3	3.24	58.5	.45-.80	LH-RHa	71.800	Mar. 6. All oocytes were at vitellogenic stages Mar. 14. Oocytes were at oil-droplet-yolk vesicle stages; residue pellets were 54 mg
Apr.25,1986	E.f.	3+	4.45	62.1	.50-.53	LH-RHa	58.100	Apr. 30. Oocytes ranged from 500-700 micron in size May 2. Oocytes at size 520-600 um May 7. Spawned after HCG injection
Apr.25,1986	E.f.	3+	4.20	61.1	.46-.53	LH-RHa	30.200	Apr. 30. Oocytes ranged from 500-600 micron in size May 2. Oocytes at size 490-530 um May 21. Oocytes degene-rated.
Apr.25,1986	E.f.	3+	3.80	61.0	.50-.55	LH-RHa	41.700	Apr. 30. Some oocytes progressed to germinal vesicle migration stage. May 2. Oocytes found only at yolk vesicle and yolk globule stage. May 21. Oocytes degene-rated.
Apr.25,1986	E.f.	3	3.10	59.4	.32	------		Primary yolk stages
May 8, 1986	E.f.	4	5.60	71.0	.38-.47	LH-RHa	50.900	May 21. Oocytes sampled and were degenerated.
June 6,1986	E.f.	3+	3.55	57.3	.50	LH-RHa	36.300	June 10. Oocytes degene-rated.
July,8,1986	E.f.	3	3.30	60.5		β-E2	9.424	July 15. Oocytes were at previtellogenic stages July 21. Died
Nov.7, 1986	E.f.	4	5.00	67.2	.20	PU.+V.E	19.200	June 10, 1987. Not can-nulated. July 29, 1987. Oocytes were at perinucleolar stage
Nov.7,1986	E.f.	3	3.80	63.0	.25	PU.+V.E	32.100	June 13, 1987. Oocytes were at tertiary yolk stage
Nov.15,1986	E.f.	4+	5.90	69.8	.15	SHAM		June 9, 1987. Oocytes at perinucleolar stage July 29, 1987. Not can-nulated.
Nov.21,1986	E.f.	5+	6.25	74.4		SHAM		June 9, 1987. Not can-nulated. July 29, 1987. Oocytes ranged from 270-300 micron in size
Nov.21,1986	E.f.	3+	3.80	62.5		----		June 13, 1987. Oocytes at size 130-220 um
Nov.21,1986	E.f.	2+	2.80	55.2	.20	----		June 10, 1987. Oocytes were at perinucleolar stage

*Beginning of pellet implantation ; ** E.f.: *E. fario*;
PU.: Puberogen ; V.E : α-Tocopherol ; E2:β-Estradiol

表 10 1986 年鮭形石斑之催熟埋植試驗

Table 10 Results of hormonal pellet implantation on the grouper *Epinephelus salmonoides* in 1986.

Date*	Species**	Age	BW (Kg)	BL (cm)	Oocyte diameter (mm)	Hormone type	Dose (mg/Kg BW)	Results
Apr.11,1986	E.s.	1+	1.32	44.5		LH-RHa	22.575	July 4. All oocytes were at previtellogenic stage Dec. 7. Same
Apr.11,1986	E.s.	1+	1.05	42.5		LH-RHa	28.571	July 4. Oocytes were not developed Nov. 7. Oocytes at oogonial stage
Apr.11,1986	E.s.	1+	1.42	46.0		LH-RHa	42.253	May 15. Oocytes were all not developed and not being cannulated.
Apr.11,1986	E.s.	1+	1.35	46.2		LH-RHa	43.851	May 6. Oocytes were not developed May 14. Oocytes were at oogonial stage July 4. Same Nov. 7. Same
Apr.11,1986	E.s.	1+	1.10	42.0		LH-RHa	23.181	May 6. Oocytes were not developed May 14. Previtellogenic oocytes were found July 4. Not cannulated. Nov. 7. Same
Apr.25,1986	E.s.	3	3.10	56.2	.02			Oogonial stages
June 19,1986	E.s.	2	1.77	49.3		β-E2	20.903	July 18. Oocytes at oogonial stage Aug. 9. Died
June 19,1986	E.s.	2	1.60	47.8		β-E2	10.437	July 18. Oogonial stage Sept. 2. Same
Nov.7,1986	E.s.	2	2.90	56.1		PU.+E2+V.E	21.700	June 9, 1987 Oocytes not cannulated.
Nov.7,1986	E.s.	3	3.50	58.8	.03	PU.+E2+V.E	12.100	June 9, 1987. Same
Nov.7,1986	E.s.	2+	2.50	56.7		SHAM		June 9, 1987. Oocytes were not developed.
Nov.7,1986	E.s.	2	3.00	57.4		SHAM		June 16, 1987. Oocytes were at perinucleolar stage
Nov.8,1986	E.s.	3	3.20	57.2	.03	E2+V.E	17.000	
Nov.8,1986	E.s.	2+	3.00	56.6		E2+V.E	29.000	
Nov.8,1986	E.s.	2+	2.95	55.7		PU.+E2+V.E	29.100	
Nov.8,1986	E.s.	2+	2.90	56.0	.02	PU.+E2+V.E	29.100	
Nov.8,1986	E.s.	2+	3.00	57.8		PU.+V.E.	29.300	
Nov.8,1986	E.s.	3	3.60	60.5	.02	PU.+V.E	30.500	
Nov.8,1986	E.s.	3	3.20	57.1		PU.+V.E	30.000	
Nov.8,1986	E.s.	3	3.60	61.3		SHAM		June 9, 1987. Oocytes not developed.
Nov.8,1986	E.s.	3	3.35	58.8	.05	SHAM		June 9, 1987. Same
Nov.8,1986	E.s.	2	2.10	53.7		----		

*Beginning of pellet implantation; ** E.s.: E. salmonoides
PU. : Puberogen; V. E : α-Tocopherol; E2 : β-Estradiol

表 11　1987 年以不同荷爾蒙對石斑魚之催熟埋植試驗

Table 11 Results of hormonal pellet implantation on the groupers *Epinephelus salmonoides* and *E. fario* in 1987.

Date*	Species**	Age	BW (Kg)	BL (cm)	oocyte diameter (mm)	Hormone type	Dose (mg/Kg BW)	Results
Apr.28,1987	E.f.	2	2.20	52.4		V.E+E1+PE.	16.272	Dec. 11. Oocytes not cannulated
Apr.28,1987	E.f.	3	3.30	59.1	.06	V.E+PU.+PE.	8.030	Dec. 11. Oocytes not cannulated
June 9,1987	E.f.	5	6.80	75.5	.90	----		June 16. Died
June 9,1987	E.f.	5	6.40	69.8	.90	----		June 25. Died
Apr.29,1987	E.s.	2	2.18	51.9		V.E	10.550	June 30. Oocytes were at oogonial stages; residue pellets were 20 mg
Apr.30,1987	E.s.	2+	2.40	53.5		V.E+E2	5.040	May 22. Oocytes not developed
Apr.30,1987	E.s.	2+	2.42	53.2		V.E+E2	10.041	May 19. Same
Apr.30,1987	E.s.	2+	2.60	53.4	.03	V.E+E2	20.769	
Apr.30,1987	E.s.	2+	2.80	53.3	.05	V.E+E2	5.357	Dec. 11. Oocytes not cannulated.
Apr.30,1987	E.s.	2+	2.92	54.8	.03	V.E+E2+PU.	5.479	
Apr.30,1987	E.s.	2	2.15	51.4		V.E+E2+PU.	9.767	
Apr.30,1987	E.s.	2	1.90	49.3		V.E+E2+PU.	21.053	Dec. 11. Oocytes not develped
May 4,1987	E.s.	2	2.20	53.2		V.E+E2	5.000	Dec. 11. Oocytes not cannulated.
May 4,1987	E.s.	2	2.68	55.4		V.E+E2	10.448	Dec. 11. Same
May 4,1987	E.s.	2	2.95	57.2		V.E+E2	20.373	Dec. 11. Oocytes not developed
May 4,1987	E.s.	2+	2.75	54.9		V.E+E2+PU.	5.455	Dec. 11. Same
May 4,1987	E.s.	2+	2.24	52.1	.06	V.E+E2+PU.	12.946	Dec. 9. Oocytes at perinucleolar stages
May 4,1987	E.s.	2	2.75	55.6		LH-RHa	10.291	Dec. 11. Oocytes not cannulated
May 14,1987	E.s.	2	1.12	42.2		E1	19.643	
May 14,1987	E.s.	2	1.20	43.7		V.E	12.500	
May 14,1987	E.s.	2	1.40	44.5		V.E+PE.	16.286	Nov. 26. Same
May 14,1987	E.s.	2	1.48	46.3		V.E+E2	12.568	
May 14,1987	E.s.	1+	0.90	40.2		V.E+E2+PU.	11.111	Nov. 26. Oocytes not developed

* Beginning of pellet implantation; **E.s.: *E. salmonoides*; E.f.: *E. fario*; V.E: α-Tocopherol
E2: β-Estradiol; PU.: Puberogen; E1: Estrone; PE. : Peamex

PE，劑量爲9.652 mg／kg至10.411 mg／kg魚體重（表12）。在經埋植50天後，青點石斑以V.E+PU+PE埋植魚有3尾達到第三卵黃球期，2尾在油球卵黃期（oil－droplet－yolk vesicle stage），一尾在卵原細胞期。V.E+PU者也有2尾達到第三卵黃球期，餘皆處於卵黃堆積期之前的卵母細胞。而以LH－RHa者則2尾在埋植37天後卵細胞由原來周邊仁期，迅速發育至第三卵黃球期，其中一尾經HCG注射後產卵。而對照組大部份仍停留於卵黃蓄積前期，只有一尾例外發育至第三卵黃球期，但其並未排卵。在鮭形石斑，則埋植後之效果並無明顯差異，只有一尾埋植V.E+PE，其卵細胞發育至卵黃蓄積前期（Previtellogenic stage）。所以，1988年埋植以青點石斑較有明顯變化，使用LH－RHa及V.E+PU+PE或V.E+PU，皆能達到催熟之效果。

討　論

從1931年Houssay使用腦下垂體加入法（hypophysation）以來，此法即被廣泛應用於控制產卵[63]，且經不斷研究，充分證明不同促性腺激素（gonadotropin），包括腦下垂體（pituitary），人類促性腺激素（human chorionic gonadotropin，HCG），哺乳類黃體生成素（luteinizing hormone，LH）或濾泡刺激素（follicle stimulating hormone，FSH）等，均能促成排卵或產卵，但卵細胞對這些不同促性腺激素之反應效果因魚種不同而有差異，也因卵細胞之成熟度不同而不同[64]。所以使用荷爾蒙改變內分泌系統來調節魚類的生殖作用爲一種重要方法[19]。但荷爾蒙種類之選擇及如何使用，往往是決定性的關鍵。在本試驗中，由HCG及其他荷爾蒙注射魚體催熟產卵之探討，由每年之結果不同，亦顯示出季節、魚齡、卵徑、荷爾蒙種類、注射次數、注射間隔時間與荷爾蒙所用劑量都會對石斑魚之排卵有影響。生殖季節前期至中期比後期成功率高；魚齡太小及太大效果不若在生物最小成熟體型後1，2年者爲佳，這似乎跟石斑魚爲兩性魚類之先雌後雄之性分化有關。而卵雖然達卵徑420μm以上或第三卵黃期即可進行催熟產卵，但以剛開始多數卵之卵徑較大者，所需注射次數少，而且產卵率及產卵數都高。而卵徑較小者，小於600μm，雖同樣可得到催熟產卵之目的，但其所需荷爾蒙劑量就多，而且產卵率不穩定，尤其在剛開始卵徑小於420μm或左右者，常有使卵細胞在多次注射後反而退化萎縮現象，此情形在其他魚類亦常見。

對於荷爾蒙種類、劑量、注射次數、時間間隔在試驗結果都明顯指出與魚發育有密切關係，不同的組合，其結果亦互異，尤其同樣雖是HCG但日本製puberogen，與美製之A.P.L，與台製之Gona，其效果却差很多，可能係純度不同之關係。注射時間間隔則不僅關係著荷爾蒙劑量之多寡，亦可應用於產卵時間之控制或採卵時機之把握，在本次研究中，就是利用較低劑量之荷爾蒙，以及延長注射間隔時間，探討如何延時產卵，但亦容易發生卵細胞退化之現象，可能爲開始催熟時之劑量沒有控制好，無法讓卵細胞進行最後成熟（final maturation）階段。所以青點石斑促使排卵較佳之條件爲以5至6月（魚塭養成者）爲催熟時機，3至4齡爲親魚，卵徑至少420μm以上，越大越好，若用HCG則以劑量每1000IU／kg BW，注射間隔24小時，2至3次注射爲宜。同樣地鮭形石斑之催熟產卵亦是卵徑450μm以上，荷爾蒙劑量1000 IU／kg BW，注射2至3針，間隔24小時內爲佳。

本次注射催熟產卵試驗荷爾蒙係以HCG爲主，但近年來對於LH－RHa功效之研究已頗有進展[35][36][37][38][39][40][41]，而且在以荷爾蒙誘導成熟過程需較長期的刺激，以往利用多次注射來達成目的[42]，但重覆操作之壓迫（stress），會使荷爾蒙效力受影響，但若以藥粒（pellet）植入魚體內可減少處理次數與比注射方式能有較長期效果（Crim，1984，葉，1988）[14][18]，所以使緩慢釋出荷爾蒙做爲慢性處理減少操作次數亦爲另一模式[18]。就LH－RHa之主要功能爲使下視丘的GtH產生細胞活性化，以促進腦下腺GtH的分泌，進而促進卵黃形成（vitellogenesis）並誘發成熟及排卵，且其應用於鯉魚（*Cyprinus carpio*）[30]，鱒魚（*Salmo trutta*）[31]已有顯著的功效，又LH－RHa具有持續性的功能，適宜與膽固醇（cholesterol）製成藥粒（pellet）[22]植入魚體。在本試驗以埋植方法之

表 12　1988 年青點石斑及鮭形石斑之催熟產卵埋植試驗

Table 12 Results of hormonal pellet implantation on the groupers, *Epinephelus salmonoides* and *E. fario* in 1988.

Date*	Species**	Age	BW (Kg)	BL (cm)	Oocyte diameter (mm)	Hormone type	Dose (mg/Kg BW)	Results
May 6,1988	E.f.	3+	4.60	64.1	.46-.47	V.E+PU.+PE.	9.913	May 15. Oocytes at size 430-500 um June 30. Oocytes at tertiary yolk stage.
May 6,1988	E.f.	3+	3.76	62.4	.12-.20	V.E+PU.+PE.	9.973	May 10. Previtellogenic oocytes were found. June 30. Same
May 6,1988	E.f.	4	4.75	64.4	.50	V.E+PU.+PE.	4.463	May 10. Oocytes at tertiary yolk stage. June 29. Oocytes were at size 200-300 um.
May 6,1988	E.f.	3+	3.40	59.8	.33-.37	V.E+PU.+PE.	6.294	May 10. Oocytes at oil-droplet-yolk vesicle stage June 30. Oocytes ranged from 250-280 micron in size
May 11,1988	E.f.	3+	4.20	64.3	.06-.12	V.E+PU.+PE.	3.024	June 29. Oocytes at oogonial stages
May 13,1988	E.f.	3	2.87	59.7	.06-.12	V.E+PU.	15.000	June 29. All oocytes were at previtellogenic stage
May 13,1988	E.f.	3	3.40	62.2	.06-.12	SHAM		June 30. All oocytes were at previtellogenic stage
May 13,1988	E.f.	3+	3.80	61.8	.47-.63	SHAM		May 16. Oocytes ranged from 430um upward in size May 18. Oocytes degenerated June 30. Tertiary yolk stage oocytes were found
May 16,1988	E.f.	3+	4.00	62.8	.20-.50	V.E+PU.	10.400	July 11. Oocytes were at tertiary yolk stage. July 14. Spawned after HCG injection
May 16,1988	E.f.	3	3.66	62.4	.06	V.E+PE.	14.290	
May 16,1988	E.f.	3+	4.40	65.8	.06-.20	V.E+PU.+PE.	14.091	July 11. Oocytes ranged from 400-420 micron in size July 15. Spawned after HCG injection
May 23,1988	E.f.	3	3.42	61.8	.40	V.E+PU.	8.889	June 30. Degenerated
May 23,1988	E.f.	3+	4.60	66.1	.20	V.E+PU..	7.043	July 11. Oocytes were at tertiary yolk stage. July 14. Spawned after HCG injection
May 23,1988	E.f.	3	3.50	61.8	.20	V.E+PE.	18.171	June 30. Oogonial stage
May 23,1988	E.f.	3+	4.12	64.5	.20	LH-RHa	8.422	June 29. Oocytes were at tertiary yolk stage. July 11. Oocyte over-ripe
May 23,1988	E.f.	3+	4.70	65.8	.12	LH-RHa	10.128	June 30. Oocytes ranged from 260-380 micron in size. July 11. Oocytes at size 450-800 um July 14. Spawned after HCG injection
May 23,1988	E.f.	3	2.70	56.6	.12	V.E+PU.	18.815	
May 26,1988	E.f.	6+	8.40	81.3	.20	V.E+PU.	3.405	
June 10,1988	E.f.	3+	4.20	62.1	.06	----		June 13. Oogonial stage
June 10,1988	E.f.	5	6.45	70.5		----		June 13. Not cannulated
May 24,1988	E.s.	5	6.60	75.0	.06	V.E+PE.	9.652	
May 24,1988	E.s.	3+	4.00	65.3		SHAM		
May 24,1988	E.s.	3	2.54	53.7	.06	SHAM		
May 25,1988	E.s.	5+	6.70	75.8	.15	V.E+PE.	9.970	May 30. All oocytes were at previtellogenic stage
May 25,1988	E.s.	5	6.08	72.8	.20	V.E+PU.+PE.	10.411	
June 3,1988	E.s.	5	5.80	73.4		----		June 29. Not cannulated
June 3,1988	E.s.	5	6.10	74.4		----		June 30. Same

* Beginning of pellet implantation; **E.s.: *E. salmonoides*; E.f.: *E. fario*; V.E: α-Tocopherol E2: β-Estradiol; PU.: Puberogen; E1: Estrone; PE. : Peamex

結果，亦指出以LH－RHa之藥粒較具有催熟效果，其他以β－E2，V.E＋PU＋PE，或V.E＋PU雖亦能達到催熟之目的，然却不穩定。在以荷爾蒙粒植入 land locked salmon，虹鱒(Crim et al 1983)[20]，Atlantic salmon (Crim and Glebe 1985)[23]，Sea bass，rabbitfish（Harvey et al 1985）之試驗中，亦以LH－RHa之藥粒能加速這些魚之生理循環。同樣以埋植法試驗於虱目魚誘導成熟，亦曾使用SPH (salmon pituitary homogenate)，SG－G100 (salmon gonadotropin) HCG (human chorionic gonadotropin) 等荷爾蒙但效果未顯 (Lacanilao et al 1984)[43]，但後來改用LH－RHa及LH－RHa＋17 α－MT 則效果非常佳 (Lee，1986)[29]，而單獨使用liquid或crystalline之17α－MT 刺激虱目魚成熟無效，這都說明了LH－RHa之功效，所以在埋植法之應用上，藥粒所含荷爾蒙種類對預期效果之好壞影響很大。

魚類本身性腺發育程度對於受刺激之反應關係重大，不論是環境因素 (Cue) 或是荷爾蒙處理。故不管以注射或埋植方式，在魚類催熟產卵之應用，都必需在性腺發育至某一程度及在一定年齡以上才行。像虱目魚在5齡以上始能刺激其成熟[19][44]，未成熟虹鱒的腦下腺對LH－RHa亦無任何反應（Crim and Evans，1980)[45]，本試驗中亦出現同樣情形，對於已屆成熟年齡或較大體型之青點石斑以LH－RHa刺激，通常都可順利使卵細胞發育至第三卵黃期，但在鮭形石斑則其效果就不若如此顯著，其因可能就是其大都爲2至3齡左右之魚，離其成熟年齡尙久，未到其成熟體型[6]，故在挑選試驗用親魚做爲埋植催熟對象時，應選擇接近生物最小成熟體型後之魚來做。

埋植時機及操作時之季節對於石斑魚催熟來說亦是重要之因素，在成功使石斑魚達到成熟階段之操作時機皆在其生殖季4月至7月之初期，亦即4月至5月時埋植爾蒙藥粒，很快的促使石斑魚成熟，而在其他月份，則未能有如此效果，此說明了雖然藥粒 (pellet) 能慢慢釋放出荷爾蒙成份促進成熟，但最好能配合魚類本身之生理週期，與環境之因素 (Cue)，才能發揮效果。

另一對於埋植法影響效果甚大的爲所作藥粒 (pellet) 之形式與操作方法，藥粒形式在虱目魚、烏魚已獲得促進產卵的效果[25][26][46]，在鮭魚、虹鱒及鱸魚使排卵或產卵同步化[47][48]，而據Lee (1986) 以silastic tube 內裝LH－RHa＋17α－MT對促進虱目魚成熟亦有效，又有以silastic capsule含testosterone埋入幼鱒腹腔內可刺激腦下腺產生GtH (Crim and Evans，1982)[15]。所以埋植之pellet之型式有必要進一步探討，以silastic tube來將各種荷爾蒙混合裝在一起，避免因不同荷爾蒙混在一起製成pellet時，因不同物理性質而遭破壞，像本試驗之各種HCG混合製成之pellet功效不顯，有可能就是在製成pellet 之過程中功效大打折扣[32]。又據Sherwood et al (1987) 研究藥粒釋放效果因膽固醇含量而有明顯的差異，含膽固醇75%之藥丸其釋放程度遠比膽固醇含量95%者快速，且於排卵前血清中促性腺激素急遽增加 (ovulatory surge) 以促進排卵作用。本試驗在LH－RHa之膽固醇含量皆爲95%，而結果亦指出只能讓青點石斑卵細胞成熟達第三卵黃期，而無法排卵，必需再靠HCG之刺激才能使石斑魚產卵，可能就是這個原故，所以，低膽固醇含量之pellet適合於促進排卵之用，至於卵黃生成過程，GtH需有持續性之刺激，則以高膽固醇含量之成分較適宜。

總之，本研究以注射法及埋植性之刺激分別探討促使石斑魚成熟及排卵之技術，雖在促進排卵方面，已成功的能使青點石斑自然將卵產出，但是在埋植法之應用上，效果並不如預期的佳，特別是在鮭形石斑上效果部份不顯，故如何來克服此方面的問題都是今後研究的重點。

摘　要

如何培育出大量成熟石斑魚，爲解決石斑魚人工繁殖成功及大量推廣養殖之重要關鍵，本試驗以注射方式及埋植法利用HCG，LH－RHa及Estrogen等荷爾蒙探討促進石斑魚成熟與排卵之技術，從1985年至1988年青點石斑及鮭形石斑之實驗結果如下：

一.青點石斑3齡左右，卵徑400至600μm者，施以HCG催熟，劑約1000 IU／kg 魚體重，間隔24小時

內注射1次，每尾注射2至3針，則於第1針後第48至96小時內可產卵，產卵數約$3x10^5$粒。

二.成熟或產過卵之青點石斑再放回魚塭培育，次年可再度成熟，以HCG處理，劑量500 IU／kg至1000 IU／kg 魚體重，2至3針注射可排卵，產卵數約17至160萬粒。

三.青點石斑2＋魚齡就可成熟，而以3至4齡者多，進行催熟產卵成功率佔50%以上。

四.使用於促進排卵荷爾蒙以HCG爲主，progesterone對末期排卵有作用。

五.產卵時間受打針數影響，注射1針者在第1針後3日及5日產卵，2針者集中在3、4日後產卵，3針者在4、5日後產卵。

六.埋植催熟試驗，荷爾蒙種類以LH－RHa及PU＋V.E對青點石斑催熟有很大功效。鮭形石斑則以LH－RHa及β－E2較具效果。

七.荷爾蒙LH－RHa埋植之劑量以每10 mg／kg至70 mg／kg魚體重之範圍對催熟有效，其他種類荷爾蒙之劑量多寡影響效果不明顯。

八.魚種、魚齡、性腺發育、季節、荷爾蒙種類、劑量、植入方式等對埋植法之催熟成敗有很大影響。

謝　辭

本試驗工作，得以完成，非常感謝王村籐先生之鼎力協助，及分同仁慨借器材，提供意見，謹此致以最深的謝忱。

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