Chapter V

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Chapter VI

Appendices

Appendix I : Chemicals

Hormones

human chorionic gonadotropin (hCG)	: Sigma Chemical company, St. Louis, U.S.A.
ovine-prolactin (o-PRL)	: Sigma Chemical company, St. Louis, U.S.A.
prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$)	: Sigma Chemical company, St. Louis, U.S.A.

Dye

trypan blue	: BDH Chemical Ltd.

Enzyme

collagenase	: Sigma Chemical company, St. Louis, U.S.A.
typsin	: Sigma Chemical company, St. Louis, U.S.A.

Medium

Dubecco's Modified Eagle Medium	: GIBCo, Grand Island, New York, U.S.A.
Nutrient Mixture-F10 (Ham)	: GIBCo, Grand Island, New York, U.S.A.

Others

antisera	: Matched Reagent Programme, WHO.
atropine	: The goverment pharmaceutical organizaiton, Bangkok, Thailand.
benzene	: Merck Ltd., Darmstadt, Germany.
charcoal	: Matched Reagent Programme, WHO.

diethyl ether : Merck Ltd., Darmstadt, Germany

disodium hydrogen phosphate (anhydrous)
: Merck Ltd., Darmstadt, Germany

dioxane, A.R. grade : Mallineckrodt Inc., Germany

ethanol : Merck Ltd., Darmstadt, Germany

fungizone (25 meq/ml) : GIBCo, Grand Island, New York, U.S.A.

fetal bovine serum : Grand Island Biological Co., New Zealand.

gelatin : Difco laboratories, detroit, Michigan, U.S.A.

Hank's balanced salt solution (HBSS) : Grand Island Biological Co., New York, U.S.A.

heparin : Leo Pharmacentical products, Balelrup-Denmark.

hydrochloric acid : Merck Ltd., Darmstadt, Germany.

ketamine : Parke-Davis PTV, Ltd., Sydney, N.S.W. Australia.

liquidflor : New England Nuclear 549 Albany St., Boston.

N-2-hydroxyethyl piperazine-N-2 thanesulfonic acid (HEPES : Sigma chemical company, St. Louis, U.S.A.

penicillin-streptomycin(pen 10,000 iu/ml, sterp 10,000 mcg/ml : Grand Island Biological company, Ohio, U.S.A.,

sodium bicarbonate : Merck Ltd., Darmstadt, Germany.

sodium chloride : Merck Ltd., Darmstadt, Germany.

sodium hydroxide, A.R. grade : BDH Chemical Ltd.

thiomerosal (ethylmercurithiosalicylate) : Sigma Chemical company, St. Louis, U.S.A.

toluene : Merck Ltd., Darmstadt, Germany.

tracer; (2,4,6,7,16,17,-H^3)-estradiol and (1,2,6,7-^3H) progesterone : The Radiochemical center, Amersham.

Appendix II : Instruments

autoclave : Forma Scientific Mariette Ohio

balance : W.M. Ainsworth and Sons Inv., Benver, Colo, U.S.A.

centrifugator : Cay Adams, Decton, Dickinson and Company, Parsipany, N.

drying cabinet, series 2000 : Termarks, U.S.A.

dissecting kit : Vithayasom Bangkok, Thailand

inucbator (u Trol-CO_2 incubator, Model 329: Forma Scientific Mariette, Ohio.

laporascope : Wolf, Germany

laminar flow hood, series no NBB-199 : Bellco Glass, Inc. Vineland, New Jersey.

magnetic stirrer : S-18521 Thermolyne Corporation, Jowa, U.S.A.

Mettler balance : E200 Mettler Instrument, Switzerland

micropipette eppendrof : 3130, Germany

mixer : Thermolyne Corporation, subsidiary of Syborn corporation, Dubuguelowa, U.S.A.

pH-meter : 5985 Cole Parmer Instrument Company, Chicago, Illinoi, 60648, U.S.A.

pipettegun : Clay Adam, U.S.A.

rackbeta liquid scintillation counters, model 1218-811 : Wallac Oy Turku, Finland.

refrigerated centrifuge, Model PR-J : International equipment company Mass, U.S.A.

ultrasonic cleanser : W.M. Ainsworth and Sons Inc., Benver Colo, U.S.A.

multi-well dish	: Nuncon, Denmark
millipore size 0.22, 0.45 μm	: Crop Bedford Mass, U.S.A.

Appendix III : Reagent preparation

Hormones

Human chorionic gonadotrophin (hCG, 1.0 iu/25 µl); hCG in lyophilized form (2500 iu/vial) was dissolved completely in tri-distilled water to the final concentration 40 iu/ml (1.0 iu/25 µl).

Ovine-prolactin (o-PRL, 10 µg/25 µl); 250 iu/8.1 mg in o-PRL in lyophilized form was prior dissolved in 0.01 M $NaHCO_3$ 200 µl and added with tri-distilled water up to the final concentration 400 µg/ml (10 µg/25 µl).

Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$, 500 ng/25 µl); $PGF_{2\alpha}$ 1 mg/vial in lyophilized form was prior dissolved in 200 l of 95% ethanol and was dissolved in tri-distilled water to final concentration 20 µg/ml (500 ng/25 µl).

Each hormone was steriled by filtering through 0.45 µm millipore filter, aliquoted 0.5 ml/tube and stored in deep freezer (-20°C) for each experiment.

Enzyme

- Collagenase 0.1% : collagenase 0.01 grams was dissolved in culture medium 10 ml by gentle shaking and prepared freshly before use.

- Trypsin 0.1%:trypsin 0.02 grams was dissolved in HBSS 10 ml by gentle stirring and kept at -10°C.

Dye

- Trypan blue 0.2%: trypan blue 0.02 grams was dissolved in HBSS 10 ml by gentle shaking and prepared freshly before use.

Appendix IV : Steriled technique

- Glasswares and cotton were steriled in oven at 150°C for a half and hour.

- Glasswares which attached with rubber, millipore and dissecting kit were autoclaved at 15 inch-pressure, 240°C for 30 minutes.

- Medium and balance salt solution were steriled by filtering through 0.22 μm millipore filter.

- Hormones were steriled by filtering through 0.45 μm millipore filter.

Appendix V : Reagents for steroid hormones assay

Assay buffer (buffer S, BS)

Dissolved gelatin 1.0 gm in warm distilled water 300 ml completely, then added the chemicals as below;

Sodium dihydrogenophosphate-1-hydrate ($NaHPO_4$, M.W. 120) 3.10 gm.

Disodium hydrogenphosphate (anhydrous) (Na_2HPO_4, M.W. 142) 11.6 gm.

Sodium chloride 8.8 gm.

Thiomersol (merthiolate) 0.1 gm.

Added up the solution to 1 litre with distilled water, mixed until the chemicals dissolved completely. Adjusted the pH to the range 7.2-7.4 and stored at $4^{o}C$. This solution is used as a diluent for all the reagents in steroid assays and should be stable for up to 1 month at $4^{o}C$.

Charcoal suspension

Dissolved dextran 0.0625 gm in 100 ml of assay buffer (BS) completely, then added activated charcoal 0.625 gm and shaked vigorously by magnetic stirrer for 30 seconds. Stored at $4^{o}C$. The charcoal suspension should be stable for up to 1 month and needs to be thoroughly mixed during usage.

Counting solution

Mixed toluene 3 litres, liquidflor 128 ml and dioxane 600 ml homogenously and stored in brown bottles.

Steroid standard

Progesterone standard

Progesterone standard bottled in concentration of 24 nmol/litre and served as stock P standard. Aliquoted stock P standard 0.5 ml, then added BS 4.5 ml and mixed carfully. This P standard would contain 2.4 pmol/ml (2400 fmol/ml or 1200 fmol/0.5 ml) and was kept as standard I. Prepared five serial dilution (1:1, standard: BS) of standard I shortly before use with series concentration of 1200, 600, 300, 150, 75 and 37.5 fmol/0.5 ml, respectively.

Estradiol standard

Estradiol standard bottled in concentration of 16 nmol/litre and served as stock E_2 standard. Aliquoted stock E_2 standard 0.5 ml, then added BS 4.5 ml. and mixed carfully. This E_2 standard would contain 1.6 pmol/ml (1600 fmol/ml or 800 fmol/0.5 ml) and was kept as standard I. Prepared five serial dilution (1:1, standard: BS) of standard I shortly before use with series concentration of 800, 400, 200, 100, 50 and 25 fmol/0.5 ml, respectively.

Steroid antisera

Progesterone antisera and estradiol antisera were raised in intact rabbit against progesterone-3-0-carboxyme-thyloxene-BSA and estradiol-3-0-carboxymethyloxene-BSA respectively. Each was obtained from WHO Match Reagent Program and bottled in lyophilized form. Immediately before use, the contents should be reconstituted with exactly 10 ml of assay buffer and will give the final dilution as 1 : 7000 in the P assay tube and 1 : 5600 in the E_2 assay tube.

Steroid tracers

(1,2,6,7-^{3}H) progesterone (specific activity = 80-110 (curies/mmol or 3.0-4.1 TBq/mmol) and (2,4,6,7,16,17-^{3}H) estradiol (specific activity = 140-170 curies/mmol or 5.2-6.3 TBq/mmol) each was filled in ampoule with concentration 25 µCi and was diluted up to 25 ml with analytical grade of benzene: ethanol; 9:1 and served as a stock solution of 10 µCi/ml. Stored at 4^{o}C and should be stable for 6 month. Shortly before use, the working tracer was prepared by evaporating the stock solution and diluted 1:100 in assay buffer; to obtain 100 nCi/ml in assay.

Appendix VI:

Composition of medium for luteal cell culture.

Components	DMEM Cat. No. 430-1600 mg/L	Nutrient mixture-F10 Cat. No. 430-1200 mg/L
Inorganic salts :		
$CaCl_2$ (anhydrous)	2.00	33.29
KCL	400.00	285.00
$MgSO_4$	97.69	74.64
NaCL	6400.00	7400.00
NaH_2PO_4	125	-
Na_2HPO_4	-	153.7
$CuSO_4.5H_2O$	-	0.0025
$FeSO_4.7H_2O$	-	0.834
Other components		
D-glucose	100	1100
Sodium pyruvate	110	110
Phenol red	15	1.2
Hypoxanthine	-	4.68
Lipoic acid	-	0.20
Thymidine	-	0.70
Amino acid :		
L-Arginine	84	211.0
L-Glutamine	584	146.0
Glycine	30	7.51
L-Histidine HCL.H_2O	42	23.0
L-Isoleucine	105	2.6
L-Leucine	105	13.0

continued

Components	DMEM Cat No. 430-1600 mg/L	Nutrient mixture-F10 Cat No. 430-1200 mg/L
amino-acid (continued)		
L-Lysine.HCL	146	29.0
L-Methionine	30	4.48
L-Phenylalanine	66	5.0
L-Serine	42.0	10.5
L-Threonine	95.0	3.57
L-Tryptophan	16.0	0.6
L-Tyrosine (disodium salt)	103.79	2.61
L-Valine	94	3.50
L-Cystine.2HCl	62.57	-
L-Alanine	-	9.0
L-Asparagine $2H_2O$	-	15.10
L-Cysteine	-	25.0
L-Glutamic acid	-	14.7
L-Glutamine	-	146.0
L-Histidine.HCL.H_2O	-	23.0
L-Isolucine	-	2.6
L-Leucine	-	13.0
L-Lysine.HCL	-	29.0
L-Proline	-	11.50
L-Aspartic acid	-	13.01
Vitamins :		
D-Ca.panthothenate	4.0	0.715
Choline chloride	4.0	0.698

continued

Component	DMEM Cat. No. 430-1600 mg/L	Nutrient mixture-F10 Cat. No. 430-1200 mg/L
Vitamins (continued)		
Folic acid	4.0	1.320
Nicotinamide	4.0	0.615
Pyridoxal.HCL	4.0	0.206
Riboflavin	0.40	0.376
Thiamine.HCL	4.0	1.000
Inositol	7.20	0.541
Biotin	-	0.024
Vitamin-B12	-	1.360

Appendix VII

Table. Other works in CL function in vitro.

Rhesus monkeys (Macaca mulatta)

stages	medium	hormone treatment	incubation peroid	authors
cycle : day 15-19 pregnancy : day 23-26,108-118,161-166	Ham's F10	hCG 100-1000 (ng/ml(12,000 iu/ mg)	3 hrs.	Stouffer, R.L. et al. (1976)
cycle : day 15-19 day 22-28	Ham's F10	hCG 0.1-100 ng/ml (11,600 iu/mg) mCG 60-600 µg/ml (0.2 iu hCG eq/mg) hLH 0.25-1 µg/ml (4620 iu/mg) hFSH 0.25 µg/ml) (3600 iu/mg)	6 hrs.	Stouffer, R.L. et al. (1977)
post partum and lactating stages	Ham's F10	hCG 100 ng/ml (11,600 iu/mg)	3 hrs	Stouffer, R.L. et al. (1977)
pregnancy : day 22-29, 160-165 cycle : day 16-19	Ham's F10	hCG 27 nM. (11600 iu/mg) dbc-AMP 1 mM.	3 hrs	Stouffer, R.L. et al. (1978)
cycle : day 16-18, 21-22.	Ham's F10	$PGE_{2\alpha}$ 50-5000 ng/ ml $PGF_{2\alpha}$ 50-5000 ng/ ml dbc-AMP 1 mM hCG 1 µg/ml	3 hrs	Stouffer, R.L. et al. (1979)

Rhesus monkey (Macaca mulatta) : continued

stages	medium	hormone-treatment	incubation time	authors
cycle : day 17-19	Ham's F10 Hank's M199 Ham's F10 (D-valine)	hCG 100 ng/ml (11,600 iu/mg)	10 days	Gulyas,B.J. et al.(1979)
cycle : day 16-19 day 22-22 day 23-25	Ham's F10	hCG 0.1-100 ng/ml (11,600 iu/mg)	3 hrs	Stouffer, R.L.et al. (1980)
cycle : day 17-19	Ham's F10	insulin 1.0 mU/ml thyroxine(T_4)10^{-7}M. cortisol 0.01 µg/ml cholesterol 0.1 mg/ml hCG 100 ng/ml (11,600 iu/mg)	10 days	Gulyas,B.J. et al.(1980)
cycle : day 17-19	Ham's F10	hCG 100 ng/ml (11,600 iu/mg)	3 hrs	Gulyas,B.J. et al.(1980)
cycle : day 15-19	Ham's F10	hCG 0.1-1000 ng/ml (11,600 iu/mg)	3 hrs	Sopelak,V.M. and Hodgen, G.D.(1984)

Cynomolgus monkey (Macaca fascicularis)

stages	medium	hormone treatment	incubation peroid	authors
day of delivery	Ham's F10	hCG 100 ng/ml (11,600 iu/mg)	3 hrs	Hodgen,G. et al.(1977)
cycle : day 17-20 day of delivery	Ham's F10	hCG 1-100 ng/ml (11,600 iu/mg) h-PRL 2.5-5 mg/ml	3 hrs 8 dyas	Stouffer,R.L. et al.(1980)
cycle : 7-9 days after the preovulatory E_2 surge	Ham's F10	hCG 1.16 iu/ml) E_2 10,100,100 ng/ml clomiphene 15,150, 1500 ng/ml	3 hrs	Westfahl,P.K. and Resko, J.A. (1983)

Human :

whole human corpus luteum	EMEM	hCG 5 or 7 iu/ml	5 days	Biggs,J.S.G. and Baker, T.G. (1978)
minced human corpus luteum day 1-5,6-10 and day 11 (day 1=day of ovulation)	EMEM	hCG 10 iu/ml FSH 20 mU/ml (165,000 iu/ml) abc-AMP 10 mM/ml	3 hrs	Hunter,M.G. and Baker, T.G.(1981)
cycle : early,mid and late luteal phase	DMEM	PRL 0.1-1000 ng/ml hCG 10 iu/ml	3 hrs	Tan,G.J.S. and Biggs, J.S.G.(1982)

stages	medium	hormone treatment	incubation peroid	authors
cycle:day 16-19 day 20-22 day 23-27 (corpus luteum 10-40 mg)	Kreb's Ringer bicarbonate buffer	-	3 hrs	Patwardhan,V.V. and Lanthier,A. (1984)
human granulosa luteal cells	Ham's F10	hCG 1-10 iu/ml PRL 100 ng/ml	5 days	Polan, M.R. (1984)
cycle:day 14-18 day 19-20 day 24	DMEM	hLH 0.01-1.0 iu/ml hCG 0.1-1.0 iu/ml $PGF_{2\alpha}$ 1 µmol/l dbc-AMP 1 mmol/l	2 hrs	Richardson, M.C. and Masson, G.M. (1980)

Rat :

stages	medium	hormone treatment	incubation peroid	authors
Holtzman strain pregnant: day 2	DMEM	LH 10-20 ng/ml (11,600 iu/mg) PRL 100-200 ng/ml	perifusion technique	Wu,D.H.et al. (1976)
Sprague-Dawley strain PSP L6-L7	Kreb's ringer bicarbonate buffer	LH 1 µg/ml $PGF_{2\alpha}$ 250-500 ng/ml 1-10 µg/ml	1 hr	Hall,A.K. and Robinson, J. (1979)

Rat (continued)

stages	medium	hormone treatment	incubation peroid	authors
Spragne-Dawley PSP day 1,3,7	Kreb's ringer bicarbonate buffer	LH 5 µg/ml(NIH-LH S18) $PGF_{2\alpha}$ 0.005-50 m/l	2 hrs	Khan,M.I.et al. (1977)
CD strain PSP day 4,6,8 and day 10	MEM	b-LH 10-100 ng/ml $PGF_{2\alpha}$ 10-1000 Mm	2 hrs	Wright, K. et al. (1980)
Holtzman strain PSP day 8,12	Krebs' ringer bicarbonate buffer	-	3 hrs	Terranova, P.F. (1980)
Holtzman strain pregnant:Day 2-22	Krebs' ringer bicarbonate buffer	-	2 hrs	Taya, K. and Greenwald, G.S.(1981)
Wistar strain cycle: estrus diestrus -1 diestrus -2	DMEM	LH 2 µg/ml PRL 20 µg/ml	perifusion technique (300 min)	Murakami, N. et al.(1982)
Spraque-Dawley pregnant: day 12,14,15,19 and 22	M199	-	4 hrs	Gibori, G. et al.(1982)
Holtzman strain pregnant: day 2, 4 and day 8	M199	LH 1-10 ng/ml	2 hrs	Wada,Y. and Greenwald, G.S.(1984)

Rat (continued)

stages	medium	hormone treatment	incubation peroid	authors
Holtzman strain hypophysectomy	M199	LH 10,100 ng,1 μg /ml(2.5 u/mg)	2 hrs	Kim,I.and Greenwald,G.S.(1984)
Sprague-Dawley	M199	hCG 1-100 ng/ml (11,600 iu/mg) cholera enterotoxin 0.001-10 μg/ml p-PRL 5-50 μg/ml LDL 750 μg HDL 500 μg	3 hrs	Menon, M. et al. (1985)
Sprague-Dawley PSP day 2,4,6,8 10 and day 11	EMEM	hCG 10^{-5}-10^{-3}iu/ml	66 hrs	Kumai, A. et al. (1986)

Others :

stages	medium	hormone treatment	incubation peroid	authors
mouse: ovaries with CL	-	o-PRL 50-5000 ng r-PRL 50-100 ng FSH+LH 1 μg/ml (each)	24 hrs	McNatty, K.P. et al.(1976)
mouse: granulosa cells	RPMI-1640	hCG 0.8-100 ng/ml FSH 0.1-100 ng/ml PRL 1 μg/ml	13 days	Corredor, A.C. and Flickinger, G.L. (1983)

Others (continued)

stages	medium	hormone treatment	incubation peroid	authors
pigs: pregnant day 60	DMEM	-	2 hrs	Lemon, M. and Mauleon,P.(1982)
pig: pregnant day 70-95	Eagle's medium	hCG 0.01-10 µg/ml PRL 0.01-10 µg/ml	4 hrs	Rajkumar,K.et al. (1985)
cow: mid cycle	Ham's F12 DMEM	LH 10-100 µg/l dbc-AMP 1 mmol/l insulin 10-50iu/l	10 days	O'Shaughessy,P.J. and Wathes,D.C. (1985)
guinea pig: cycle: day 3,7, 9,10,12 and day 13	DMEM	hCG 0.001-0.1 iu/ml	2 hrs	Richardson,M.C. and Peddie,M.J. (1982)
golden hamster pregnant: day 2-16	M199	hCG 100 ng/ml	1 hr	Greenwald,G.S. et al. (1984)
pregnant: day 2,4 and day 8	M199	LH 1-10 ng/ml	2 hrs	Wada, Y. and Greenwald, G.S. (1984)

Table 14 Effects of hCG, PRL and $PGF_{2\alpha}$ on P production rate of luteal cells from different physiological stages during 3 hrs in cubation (** = $P < 0.01$, * = $P < 0.05$, significantly different from the control (a), from the hCG-treated gorup (b)).

Physiological stages		P production rate (pmol/10^5 cells/3 hrs)						
		control	hCG[a]	PRL[a]	hCG+PRL[b]	$PGF_{2\alpha}$	hCG+$PGF_{2\alpha}$	PRL+$PGF_{2\alpha}$
estrous cycle	estrus (8.00)	3.38±0.10	5.11±0.43*	5.08±0.70**	5.61±0.07**	4.11±0.17	5.26±0.52	5.48±0.20
	diestrus-1 (8.00)	6.35±0.55	9.55±0.65**	8.49±1.31**	9.85±1.15	6.65±0.55	9.80±1.10	8.08±0.22
	diestrus-2 (8.00)	3.29±0.10	3.62±0.08	3.34±0.15	3.59±0.10	3.04±0.35	3.26±0.04	3.21±0.21
	diestrus-2 (15.00)	2.77±0.16	2.84±0.12	2.79±0.05	2.95±0.24	2.75±0.12	2.89±0.43	2.75±0.04
pregnancy	L_2	10.31±1.89	23.92±4.69**	19.63±4.98*	32.42±6.82*	14.32±2.85	24.03±0.03	18.63±0.63
	L_6	18.44±3.50	38.49±8.01**	29.64±7.49*	44.87±7.74*	20.2±5.35	38.68±2.48	29.10±0.34
	L_{12}	14.66±0.16	29.07±0.24**	22.97±1.34*	33.57±3.27	16.17±1.14	28.57±3.34	19.63±1.18
	L_{20}	3.55±0.15	4.90±0.10 NS	4.60±0.60 NS	4.75±0.05	3.16±0.04	4.30±0.05	3.49±0.29
PSP	L_2	5.66±0.77	12.51±0.45*	8.36±1.00*	16.66±1.10*	7.46±1.50	11.93±0.87	7.76±0.60
	L_6	12.29±0.01	21.79±1.51**	20.49±5.81*	22.19±5.11	14.39±3.91	21.49±1.81	19.79±4.11
	L_{12}	2.07±0.44	2.99±0.06	2.44±0.31	2.79±0.14	1.90±0.44	2.44±0.26	2.50±0.44
laction	L_2	4.94±0.20	7.46±0.21*	6.29±0.04*	7.33±1.93	5.06±0.19	6.28±0.23	5.15±0.29
	L_{12}	6.41±0.25	16.3±2.10**	9.14±0.74**	17.57±0.72	8.89±0.39	14.87±2.42	10.07±0.62
	L_{20}	4.25±0.45	7.35±0.45**	4.40±0.10	7.35±0.25	4.05±0.55	6.85±0.65	3.97±0.43

Table 15 Effect of hCG, PRL and $PGF_{2\alpha}$ on E_2 production rate of luteal cells from different physiological stages during 3 hours incubation. (** $P < 0.01$, * $P < 0.05$, significantly different from the control).

physiological stages		E_2 production rate (fmol/10^5 cells/3 hrs)			
		control	hCG	PRL	$PGF_{2\alpha}$
estrous cycle	estrus (8.00)	234.0±11.0	360.0±15.0*	239.0±36.0	305.0±10.0
	diestrus-1 (8.00)	467.5±105.5	568.0±86.0*	296.0±6.0	435.0±23.0
	diestrus-2 (8.00)	255.0±10.00	365.0±10.1*	265.0±10.0	305.0±10.0
	diestrus-2 (15.00)	232.5±17.5	250.0±25.0 NS	225.0±20.0	201.5±16.5
pregnancy	L_2	316.0±12.0	564.0±16.0*	354.0±34.0	354.0±14.0
	L_6	600.0±93.5	856.8±128.3*	622.4±75.6	701.0±97.0
	L_{12}	680.0±70.0	897.5±127.5*	739.5±110.5	822.0±96.2
	L_{20}	710.0±75.0	828.5±36.5 NS	690.0±35.0	686.5±138.5
PSP	L_2	342.5±42.5	560.0±50.5*	330.0±20.0	370.0±20.0
	L_6	475.0±40.0	710.0±45.0**	490.0±75.0	525.0±50.0
	L_{12}	210.5±35.5	377.5±52.5 NS	197.5±72.5	172.5±47.5
Lactation	L_2	223.0±2.0	373.0±48.0**	257.5±32.5	289.0±11.0
	L_{12}	414.0±36.0	567.0±28.0**	464.0±14.0	434.0±24.0
	L_{20}	507.5±22.5	642.0±32.5*	527.5±2.5	477.5±52.5

Table 16 Effects of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal culls from cycling rats during 11 days of incubation. (**= $P < 0.01$, *= $P < 0.05$, significantly different from the control (a), from the hCG-treated group (b)).

stages	day of incubation	P secretion ($pmol/10^5$ cells)						
		control	hCG[a]	PRL[a]	hCG+PRL[b]	$PGF_{2\alpha}$[a]	hCG+$PGF_{2\alpha}$	PRL+$PGF_{2\alpha}$
estrus	1	21.0±0.6	20.1±1.5	22.2±4.2	20.2±2.2	22.1±3.5	21.0±4.6	19.8±1.8
	3	35.6±2.9	60.8±4.8**	52.6±1.8*	76.2±7.8*	58.2±5.8	62.5±4.5	52.6±4.5
	5	26.0±1.2	49.5±11.1**	46.2±6.6*	58.7±15.4*	48.9±11.9**	52.0±8.0	62.9±9.1
	7	22.6±6.2	27.4±4.8	25.8±3.6	48.1±6.7	26.4±7.6	40.5±3.5	30.6±6.2
	9	19.8±5.0	23.9±4.9	21.2±6.4	26.6±3.1	24.1±7.9	34.3±4.1	24.3±3.7
	11	12.2±1.4	13.5±2.5	12.5±1.8	20.7±7.3	15.5±4.6	21.2±8.5	15.9±3.3
diestrus-1	1	34.4±2.4	36.3±2.1	36.9±6.3	32.5±10.9	30.8±2.0	38.4±3.2	33.5±2.5
	3	45.3±0.5	71.5±12.5**	68.7±11.3	75.6±8.4	59.2±4.8	73.3±2.7	69.0±15.0
	5	33.9±0.3	52.6±1.8*	50.2±4.2	56.4±7.6	60.7±17.1**	66.1±2.1	70.7±9.9
	7	23.8±5.8	31.6±3.6	27.9±5.9	32.8±1.6	24.7±8.3	38.4±9.9	28.8±10.8
	9	18.6±7.0	25.59±3.9	23.2±5.2	31.5±3.7	20.8±8.8	28.7±4.3	21.2±4.8
	11	11.0±3.0	15.0±5.6	11.17±2.9	19.1±2.3	6.4±1.0	19.3±10.2	16.7±5.9
late diesturs-2	1	15.4±2.0	15.4±3.4	14.0±2.0	17.8±3.0	18.8±6.8	15.2±0.8	13.4±1.0
	3	20.2±2.2	23.8±4.2	21.8±3.8	23.2±5.6	19.2±1.2	20.6±1.1	20.4±2.0
	5	18.7±2.2	18.6±1.0	21.2±1.9	20.0±1.6	16.5±5.1	12.9±0.5	14.8±1.2
	7	14.5±4.4	16.8±2.6	13.2±3.8	16.4±4.4	13.5±4.9	14.9±5.1	14.8±4.8
	9	9.4±1.0	10.6±1.5	8.1±1.5	7.9±1.7	8.4±1.6	8.3±3.3	6.7±0.2
	11	6.2±2.1	7.3±2.8	5.5±1.3	6.8±1.4	5.1±0.9	5.2±1.7	4.2±1.3

Table 17 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells from cyclic rats during 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/10^5 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
estrus	1	0.54±0.02	0.48±0.04	0.55±0.05	0.52±0.02
	3	0.62±0.04	0.75±0.09*	0.62±0.02	0.68±0.06
	5	0.55±0.01	0.66±0.11*	0.54±0.62	0.61±0.01
	7	0.40±0.04	0.52±0.02*	0.48±0.02	0.50±0.04
	9	0.31±0.07	0.45±0.01*	0.30±0.01	0.43±0.03
	11	0.30±0.04	0.35±0.01	0.30±0.10	0.35±0.50
diestrus-1	1	0.61±0.01	0.60±0.01	0.65±0.03	0.66±0.02
	3	0.65±0.01	0.74±0.02*	0.66±0.02	0.64±0.07
	5	0.59±0.03	0.69±0.03*	0.61±0.07	0.59±0.05
	7	0.47±0.07	0.58±0.06*	0.52±0.06	0.51±0.15
	9	0.41±0.01	0.46±0.06	0.44±0.04	0.41±0.07
	11	0.27±0.06	0.32±0.04	0.34±0.04	0.30±0.05
diestrus-2	1	0.40±0.05	0.40±0.04	0.39±0.03	0.40±0.10
	3	0.37±0.05	0.33±0.03	0.32±0.01	0.35±0.01
	5	0.27±0.01	0.29±0.01	0.25±0.04	0.24±0.04
	7	0.24±0.02	0.26±0.02	0.29±0.01	0.23±0.07
	9	0.19±0.03	0.16±0.06	0.18±0.06	0.17±0.03
	11	0.14±0.02	0.17±0.03	0.15±0.03	0.11±0.01

* = $P < 0.05$ significantly different from the control

Table 18 Responsiveness of rat luteal cells from estrous cycle to hCG, PRL, $PGF_{2\alpha}$ on P and E_2 secretion ($pmol/10^5$ cells/7 days) during 7 day incubation (mean ± S.E., n = 3).

stages / treatments	estrus		diestrus-1		diestrus-2	
	P	E_2	P	E_2	P	E_2
control	103.2±15.2	2.11±0.01	137.4±5.9	2.32±0.17	68.7±5.8 NS	1.25±0.13 NS
hCG	157.8±9.6**	2.50±0.11**	192.1±22.8**	2.60±0.20*	74.7±3.9 NS	1.28±0.00
PRL	146.8±22.9**	2.19±0.15	183.7±21.5**	2.43±0.16	70.1±5.6	1.26±0.09
hCG+PRL	188.1±22.9**	-	202.3±17.3	-	75.9±6.8 NS	-
PGF_2	184.4±41.3**	2.32±0.01	164.9±6.3*	2.43±0.23	65.5±18.5	1.22±0.16
hCG+$PGF_{2\alpha}$	176.2±6.5	-	196.2±10.4	-	66.2±9.2	-
PRL+$PGF_{2\alpha}$	150.9±10.8	-	197.4±11.5	-	63.4±3.6	-

* = $P < 0.05$, ** $P < 0.01$ significantly different, NS = non-significantly differnt from the control

Table 19 Effects of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells from pregnant rats during 11 days of incubation (** = P < 0 01, * = P < 0.05, significantly different from the control (a), from the hCG-treated group (b)).

stages	day of incubation	P secretion (pmol/10^5 cells)						
		control	hCG[a]	PRL[a]	hCG+PRL[b]	$PGF_{2\alpha}$[a]	hCG+$PGF_{2\alpha}$[b]	PRL+PGF_{α}
L_2	1	41.2±6.9	48.0±0.0	43.4±11.8	46.6±15.8	39.1±8.9	47.7±8.3	46.7±4.5
	3	45.9±1.3	111.0±4.2**	56.0±4.8*	130.8±22.0**	73.2±6.8**	121.0±7.0	63.3±14.9
	5	34.2±0.6	59.0±7.4*	45.4±3.8	70.2±1.0*	47.6±6.0	68.2±0.6	49.6±4.8
	7	17.9±2.3	28.4±3.6	21.6±4.0	32.6±1.0*	30.2±13.8	40.1±5.9	25.1±3.7
	9	12.0±3.4	14.4±0.0	15.2±3.6	16.4±6.4	19.2±7.2	21.0±5.4	16.5±1.5
	11	9.0±1.2	14.8±0.8	7.6±0.6	11.3±4.3	14.6±1.0	14.6±1.4	10.9±1.9
L_6	1	56.9±2.3	56.7±0.7	67.6±0.4	55.7±5.1	61.6±13.6	52.9±1.7	55.7±10.9
	3	66.4±2.8	126.0±28.9**	89.7±5.1	191.8±61.5**	86.1±14.8*	125.4±31.0	86.5±6.8
	5	32.8±2.4	68.5±4.5*	51.6±9.8*	90.2±19.6	69.9±0.6*	77.8±10.6	62.0±7.8
	7	17.6±6.4	26.9±9.5	19.7±0.5	31.0±3.7	25.9±2.6	60.9±18.6	22.0±8.9
	9	10.2±3.2	20.0±6.8	16.8±0.4	26.3±3.2	12.1±1.4	21.6±6.1	17.1±0.2
	11	7.5±2.7	15.7±4.4	15.4±0.9	19.0±6.2	7.6±3.5	17.9±5.2	12.5±1.9
L_{12}	1	55.5±3.3	51.8±7.0	57.2±10.8	47.1±2.3	49.4±9.4	50.0±16.0	52.0±14.0
	3	59.4±0.6	127.3±0.7**	107.0±13.0**	138.7±10.7	75.3±10.8	113.3±5.3	110.0±2.0
	5	32.5±6.7	59.4±5.4**	58.0±10.0**	67.4±11.0	35.4±7.4	58.0±0.8	51.0±2.2
	7	14.6±1.0	38.3±5.7**	27.0±3.8	48.2±4.6	25.5±5.5	37.0±3.8	29.4±3.4
	9	15.0±3.0	25.9±3.3	20.0±0.4	32.1±0.1	17.2±4.4	22.0±1.0	21.8±2.6
	11	11.6±0.8	15.7±3.5	13.2±4.4	18.4±0.8	12.7±1.8	17.0±4.8	14.6±5.8
L_{20}	1	22.2±1.0	23.9±1.4	23.5±0.3	23.7±1.3	24.1±1.1	22.6±0.2	23.8±2.6
	3	11.0±3.8	36.0±2.4**	23.5±8.5**	3.38±8.6	10.4±2.4	21.8±1.6**	21.7±3.3
	5	7.8±3.1	17.7±0.9**	9.4±2.2	23.4±8.6	6.9±1.3	10.6±1.4	7.0±1.4
	7	2.7±1.3	5.3±2.5	3.1±1.3	6.4±3.6	5.4±1.8	6.6±1.0	3.2±0.4
	9	2.9±1.7	6.2±4.6	4.3±1.6	8.0±1.2	3.1±0.7	5.6±0.4	2.5±0.7
	11	2.7±0.9	5.6±1.3	5.1±1.5	5.9±1.0	2.2±0.0	4.2±0.9	2.5±1.3

Table. 20 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells from pregnant rats during 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/10^5 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
L_2	1	0.59±0.07	0.59±0.07	0.63±0.01	0.58±0.02
	3	0.69±0.05	0.82±0.04**	0.67±0.01	0.72±0.02
	5	0.60±0.09	0.72±0.06**	0.62±0.02	0.68±0.10
	7	0.54±0.04	0.64±0.04*	0.51±0.01	0.50±0.04
	9	0.45±0.08	0.53±0.02*	0.45±0.05	0.40±0.02
	11	0.33±0.01	0.42±0.02	0.33±0.03	0.38±0.11
L_6	1	0.83±0.04	0.86±0.04	0.82±0.06	0.81±0.07
	3	0.80±0.06	1.05±0.20**	0.82±0.01	0.83±0.03
	5	0.36±0.04	0.55±0.03*	0.39±0.04	0.40±0.43
	7	0.28±0.07	0.40±0.01	0.34±0.06	0.36±0.00
	9	0.26±0.03	0.37±0.01	0.28±0.01	0.27±0.06
	11	0.23±0.01	0.34±0.03	0.24±0.01	0.22±0.02
L_{12}	1	1.68±0.22	1.74±0.26	1.73±0.17	1.60±0.16
	3	1.52±0.04	1.94±0.26**	1.55±0.15	1.71±0.03
	5	0.85±0.07	1.22±0.18*	0.92±0.08	0.88±0.04
	7	0.78±0.04	0.90±0.10	0.80±0.04	0.77±0.03
	9	0.64±0.08	0.84±0.04	0.70±0.08	0.85±0.07
	11	0.63±0.05	0.67±0.06	0.72±0.02	0.75±0.09
L_{20}	1	1.92±0.16	1.84±0.08	1.82±0.26	1.80±0.28
	3	1.10±0.22	1.29±0.21	1.14±0.12	0.84±0.04
	5	0.97±0.09	0.98±0.02	0.96±0.08	0.77±0.06
	7	0.61±0.07	0.63±0.05	0.68±0.08	0.61±0.06
	9	0.60±0.06	0.59±0.06	0.58±0.06	0.54±0.06
	11	0.51±0.03	0.55±0.01	0.54±0.02	0.36±0.04

** = $P < 0.01$, * = $P < 0.05$, significantly different from the control

Table 21 Effects of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells from pseudopregnant rats during 11 days of incubation. (** = $P < 0.01$, * = $P < 0.05$ significantly different from the control (a), from the hCG-treated group (b)).

stages	day of incubation	P secretion (pmol/10^5 cells)						
		control	hCG[a]	PRL[a]	hCG+PRL[b]	$PGF_{2\alpha}$[a]	hCG+$PGF_{2\alpha}$[b]	PRL+$PGF_{2\alpha}$
L_2	1	34.4±1.6	33.2±2.8	29.6±1.2	30.3±2.3	36.0±1.7	36.9±3.9	35.9±3.7
	3	53.4±0.6	98.2±3.8**	94.4±8.4**	104.5±3.5	74.8±17.2*	91.6±6.0	95.8±13.8
	5	39.3±7.3	61.7±16.6**	48.0±8.4	83.8±17.4*	64.2±15.8**	64.0±8.0	53.3±8.9
	7	25.4±0.2	34.0±2.0	29.1±0.6	36.2±0.6	36.2±7.8	37.6±10.4	31.7±3.5
	9	14.5±0.9	23.2±4.4	18.8±1.2	30.2±5.0	22.8±3.6	29.5±4.9	29.0±2.6
	11	7.4±2.7	9.5±7.5	7.4±3.0	13.3±5.1	10.2±2.6	13.1±4.9	11.5±5.3
L_6	1	49.0±5.0	54.0±12.0	49.5±13.5	49.5±13.5	47.4±11.4	43.0±11.0	53.5±9.5
	3	52.5±7.6	101.0±3.0**	92.0±20.0**	120.9±4.9	78.4±9.6*	102.0±10.0	98.0±6.0
	5	35.2±4.0	68.4±23.6*	64.8±15.2*	71.8±22.0	67.4±1.4*	89.0±3.0	84.6±9.4
	7	20.7±6.3	37.8±18.2	31.4±10.6	54.8±32.8	48.0±12.0	57.8±11.8	42.2±15.8
	9	13.9±2.1	24.2±7.8	17.4±5.0	25.6±6.8	26.3±5.8	30.4±0.8	17.2±4.0
	11	7.7±0.3	23.0±5.8	15.4±5.4	14.2±1.0	19.4±5.0	17.2±7.6	17.0±8.6
L_{12}	1	13.3±2.7	12.6±2.6	10.4±2.4	11.4±3.0	11.0±1.8	10.3±0.9	10.0±2.0
	3	15.7±0.5	18.0±0.4	15.8±0.2	18.4±2.4	15.3±0.4	15.9±1.7	14.3±1.3
	5	12.9±2.3	13.6±1.7	9.5±2.1	9.8±1.8	8.6±1.6	9.4±1.6	7.2±1.6
	7	9.1±3.7	8.4±2.4	6.3±1.1	5.6±0.4	5.6±0.4	5.7±0.9	6.0±2.8
	9	7.9±2.5	8.4±2.4	6.1±0.9	6.2±1.0	5.7±0.5	5.8±0.9	4.7±0.9
	11	4.6±1.0	5.0±0.6	5.0±1.2	4.7±0.2	4.1±0.5	4.4±0.9	3.2±0.3

Table. 22 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells from PSP rats during 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/10^5 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
L_2	1	0.80±0.04	0.86±0.02	0.84±0.04	0.82±0.06
	3	0.88±0.12	1.05±0.19*	0.90±0.02	0.90±0.08
	5	0.76±0.08	0.90±0.06	0.71±0.07	0.79±0.12
	7	0.67±0.11	0.67±0.17	0.69±0.09	0.70±0.14
	9	0.57±0.15	0.63±0.15	0.57±0.11	0.60±0.04
	11	0.37±0.05	0.55±0.13	0.39±0.09	0.56±0.12
L_6	1	1.01±0.09	0.97±0.04	1.01±0.14	0.98±0.06
	3	0.89±0.17	1.29±0.22*	0.88±0.14	0.94±0.07
	5	0.61±0.07	0.97±0.05*	0.75±0.08	0.86±0.08
	7	0.53±0.13	0.74±0.14	0.54±0.19	0.71±0.17
	9	0.54±0.20	0.68±0.20	0.56±0.18	0.67±0.13
	11	0.51±0.13	0.58±0.14	0.59±0.04	0.61±0.12
L_{12}	1	0.54±0.12	0.60±0.12	0.55±0.03	0.54±0.06
	3	0.49±0.01	0.53±0.05	0.51±0.02	0.38±0.03
	5	0.44±0.01	0.48±0.36	0.43±0.08	0.35±0.03
	7	0.42±0.03	0.40±0.03	0.39±0.07	0.26±0.04
	9	0.35±0.07	0.33±0.09	0.35±0.06	0.25±0.02
	11	0.29±0.03	0.36±0.07	0.34±0.06	0.20±0.02

* = $P < 0.05$ significantly different from the control

Table 23 Effects of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells from lactating rats drung 11 days of incubation (** = $P < 0.01$, * = $P < 0.05$ significantly different from the control (a), from the hCG-treated group (b)).

stages	day of incubation	P secretion (pmol/10^5 cells)						
		control	hCG[a]	PRL[a]	hCG+PRL	$PGF_{2\alpha}$[a]	hCG+$PGF_{2\alpha}$[b]	PRL+$PGF_{2\alpha}$
L_2	1	26.4±0.8	24.8±1.6	30.0±2.0	26.8±4.4	30.4±0.0	29.2±2.8	30.8±2.7
	3	30.6±2.6	39.4±4.2*	37.1±3.3*	40.9±4.7	33.7±4.5	38.4±2.3	38.9±0.9
	5	19.8±0.2	27.2±0.2*	26.1±3.4*	31.4±1.0	22.8±0.5	25.6±4.4	26.9±0.9
	7	11.0±4.0	25.7±5.7**	20.8±2.8**	25.7±5.5	14.5±4.1	14.5±2.5	21.26±2.0
	9	2.8±0.0	3.8±0.2	3.3±0.1	6.0±0.8	5.9±2.5	5.6± 1.6	6.7±1.3
	11	2.9±5.0	4.9±1.1	4.6±0.4	5.1±0.7	3.7±1.3	4.5±1.1	5.0±1.0
L_{12}	1	34.5±0.4	32.6±0.2	38.4±2.4	31.4±1.4	39.2±5.2	38.4±4.8	30.3±2.1
	3	38.8±0.4	54.0±2.4**	44.3±1.3	61.8±2.6	48.1±4.9*	54.7±5.3	47.1±2.3
	5	28.7±8.5	47.2±0.8**	45.3±3.3*	45.6±3.6	42.0±0.4**	49.5±3.5	47.8±3.8
	7	13.6±2.8	22.7±1.7*	20.3±1.7	23.1±2.1	20.6±6.6	24.6±1.2	24.3±3.7
	9	14.0±1.6	17.5±0.7	14.5±3.3	14.6±5.0	12.4±0.2	12.7±0.5	13.1±1.1
	11	10.0±0.0	17.3±0.3	8.5±1.3	14.3±4.5	9.9±1.8	14.5±2.7	8.6±2.3
L_{20}	1	14.1±0.8	14.4±0.8	14.8±1.2	14.4±0.0	15.6±0.4	14.8±0.8	14.8±1.2
	3	17.4±1.4	24.8±0.6**	19.3±4.5	23.6±1.6	17.0±3.4	19.5±5.0*	13.9±2.9
	5	12.4±2.4	15.4±2.2	11.5±2.9	15.6±0.4	9.9±2.1	12.0±2.8	9.4±3.4
	7	9.6±3.2	12.3±0.9	9.7±2.3	13.5±0.1	5.9±0.8	9.1±2.1	8.4±3.2
	9	9.5±2.7	10.5±0.5	9.5±0.5	10.8±0.4	7.2±2.1	9.9±1.7	8.5±3.5
	11	8.1±4.5	7.7±1.5	8.0±3.7	10.0±2.3	7.3±5.2	6.2±2.2	8.5±4.3

Table. 24 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells from lactating rats during 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/10^5 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
L_2	1	0.44±0.01	0.45±0.01	0.56±0.04	0.47±0.01
	3	0.49±0.08	0.64±0.13*	0.51±0.06	0.52±0.03
	5	0.48±0.08	0.55±0.11	0.50±0.08	0.49±0.09
	7	0.42±0.12	0.46±0.05	0.39±0.02	0.43±0.02
	9	0.40±0.01	0.37±0.06	0.40±0.02	0.32±0.01
	11	0.32±0.02	0.30±0.07	0.35±0.02	0.28±0.03
L_{12}	1	0.57±0.03	0.57±0.03	0.59±0.08	0.54±0.03
	3	0.61±0.05	0.71±0.12	0.60±0.08	0.54±0.04
	5	0.48±0.08	0.64±0.05**	0.50±0.01	0.51±0.01
	7	0.41±0.01	0.50±0.08*	0.42±0.01	0.47±0.03
	9	0.32±0.08	0.49±0.04**	0.39±0.07	0.39±0.07
	11	0.28±0.04	0.43±0.05**	0.33±0.05	0.24±0.04
L_{20}	1	0.77±0.05	0.76±0.02	0.76±0.06	0.79±0.07
	3	0.69±0.09	0.80±0.06*	0.68±0.08	0.63±0.03
	5	0.56±0.06	0.60±0.02	0.56±0.04	0.47±0.05
	7	0.50±0.09	0.49±0.08	0.46±0.05	0.45±0.05
	9	0.38±0.07	0.38±0.05	0.37±0.07	0.37±0.05
	11	0.36±0.05	0.37±0.04	0.37±0.01	0.33±0.03

** = $P < 0.01$, * = $P < 0.05$, significantly different from the control

Table 25 Responsiveness of rat luteal cells to hCG, PRL, $PGF_{2\alpha}$ on P and E_2 secretion (pmol/10^5 cells/7 days) during 7 day inucbation (mean±S.E., n = 3).

Stages / treat	L_2		L_6		L_{12}		L_{20}	
	P	E_2	P	E_2	P	E_2	P	E_2
pregnancy control	139.1±15.7	2.42±0.03	173.7±6.4	2.20±0.12	161.8±14.7	4.83±7.1	43.6±7.1	4.67±0.47 NS
hCG	247.2±0.57**	2.72±0.06	278.0±48.9**	2.82±0.20**	277.0±26.3**	5.80±1.13**	82.88±5.7* NS	4.75±0.13
PRL	166.3±9.6*	2.43±0.01	228.6±5.2**	2.37±0.13	249.2±53.2**	5.02±0.65	45.2±4.2	4.61±0.43
hCG+PRL	280.2±14.5 aNS	-	368.7±57.3 a**	-	301.4±10.2 NS	-	87.3±10.1 NS	-
$PGF_{2\alpha}$	190.1±35.5*	2.48±0.45	243.4±44.6**	2.40±0.05	185.2±32.7	4.96±0.09	46.8±6.6 -a*	4.01±0.28
hCG+$PGF_{2\alpha}$	277.0±21.8	-	287.4±49.2	-	287.4±49.2	-	61.5±1.9	-
PRL+$PGF_{2\alpha}$	184.7±2.7	-	226.4±26.6	-	232.9±24.2	-	55.6±3.5	-
PSP control	152.5±6.9	3.11±0.35	157.3±32.4	3.04±0.02	51.0±3.8 NS	1.89±0.15 NS	-	-
hCG	227.2±19.6**	3.49±0.35*	261.2±56.8**	3.97±0.35**	52.6±2.6 NS	2.01±0.24	-	-
PRL	201.1±5.9*	3.17±0.30	237.7±59.3**	3.18±0.06	41.9±1.5	1.88±0.28	-	-
hCG+PRL	234.9±25.3	-	322.0±54.6 a*	-	45.2±2.3	-	-	-
$PGF_{2\alpha}$	211.2±37.9*	3.21±0.35	241.2±44.7**	3.49±0.49	40.5±3.8 NS	1.54±0.11	-	-
hCG+$PGF_{2\alpha}$	230.1±6.3	-	291.8±42.1	-	41.3±2.4	-	-	-
PRL+$PGF_{2\alpha}$	216.7±6.7	-	278.3±40.7	-	35.5±7.2	-	-	-
Lactation control	91.8±0.6	1.85±0.17	-	-	115.9±8.1	2.07±0.22	53.8±11.1	2.52±0.26 NS
hCG	104.3±0.5**	2.13±0.04*	-	-	156.8±4.5*	2.43±0.12**	66.9±2.4** NS	2.66±0.20
PRL	113.9±8.3**	1.86±0.13	-	-	148.4±3.9**	2.11±0.36	55.2±8.1	2.47±0.23
hCG+PRL	124.8±6.5 a*	-	-	-	161.8±3.7	-	67.1±2.7 NS	-
$PGF_{2\alpha}$	101.4±1.2*	1.91±0.02	-	-	149.9±5.5**	2.15±0.07	47.8±0.6 *a	2.25±0.29
hCG+$PGF_{2\alpha}$	107.7±7.5	-	-	-	167.2±2.5	-	55.4±9.0	-
PRL+$PGF_{2\alpha}$	119.3±2.2	-	-	-	149.6±6.5	-	48.0±9.8	-

* = $P < 0.05$, ** = $P < 0.01$, significantly different from the control

a = compared with the hCG treated group.

NS = non-significantly different.

Table 26 Effects of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells during menstrual cycle of monkey #101 in 11 days of incubation (** = $P < 0.01$, * = $P < 0.05$ significantly different from the control (a), from the hCG-treated group (b)).

stages	day of incubation	P secretion (pmol/5x10^4 cells)						
		control	hCG[a]	PRL	hCG+PRL	$PGF_{2\alpha}$[a]	hCG+$PGF_{2\alpha}$[b]	PRL+$PGF_{2\alpha}$
early luteal phase (15 days prior menses of cycle 37 days)	1	8.79±1.31	8.41±1.01	8.41±0.55	7.99±0.04	7.61±1.31	7.20±1.15	8.41±0.51
	3	9.20±1.15	24.95±0.43**	10.47±0.16	26.57±0.20	16.41±0.45**	25.22±2.00	14.44±1.15
	5	8.41±1.32	21.88±1.32*	9.10±0.10	10.80±0.21	14.81±0.70**	12.80±1.50	12.79±1.31
	7	5.60±0.44	8.22±0.52	5.73±0.70	5.99±1.10	3.79±1.51	5.26±0.52	4.52±0.65
	9	4.81±0.20	5.13±1.10	5.32±0.05	7.13±0.52	5.54±0.07	3.98±1.10	7.04±0.43
	11	4.17±0.35	7.39±0.04	5.96±0.22	7.36±0.43	4.71±1.10	3.38±0.10	3.98±0.55
mid luteal phase (8 days prior menses of cycle 30 days)	1	82.01±5.81	87.99±9.40	87.99±9.60	84.01±10.50	79.20±9.60	83.98±6.00	80.0±10.60
	3	75.99±7.49	106.59±15.80**	77.90±1.40	106.80±15.24	96.34±1.40*	109.20±15.80	96.59±13.50
	5	65.99±8.01	102.52±4.00**	67.71±4.90	109.52±5.11	90.22±11.40*	110.89±9.4	96.59±15.50
	7	30.99±7.74	53.92±11.0**	32.80±5.75	60.0±11.93	46.12±3.00	54.59±9.5	44.11±6.80
	9	20.99±5.35	54.33±5.81**	28.69±3.91	47.01±7.76	38.86±5.40	50.41±5.75	39.90±1.00
	11	14.01±3.34	51.24±3.91**	23.57±1.81	49.36±10.60	24.59±5.80	48.31±6.80	30.45±5.40
late luteal phase (2 days prior menses of cycle 32 days)	1	30.00±5.35	37.99±4.90	26.02±5.00	34.01±2.30	26.02±3.60	32.01±5.00	26.02±4.32
	3	30.41±3.00	49.20±9.80**	30.51±4.90	46.88±3.90	28.99±2.60	35.29±6.80*	27.68±2.60
	5	18.79±1.70	63.57±9.50**	19.01±2.10	57.80±8.90	16.01±2.80	42.90±7.60**	16.20±3.50
	7	13.79±0.90	36.91±1.30**	12.45±0.45	26.82±4.00	12.29±0.90	28.12±7.64	2.93±0.40
	9	11.82±0.20	27.01±1.60*	10.92±0.30	13.85±1.00	12.01±0.30	24.14±5.00	12.42±0.30
	11	11.82±0.05	10.45±2.80	12.36±0.40	2.74±0.30	12.96±0.90	5.03±1.24	12.20±0.05
luteolytic phase (2 days after menses of cycle 33 days).	1	1.66±0.05	1.59±0.25	2.01±0.90	1.59±0.05	2.01±0.10	1.82±0.10	2.10±0.09
	3	1.34±0.40	1.47±0.04	1.43±0.04	1.40±0.03	1.69±0.14	1.56±0.09	1.62±0.19
	5	1.31±0.25	1.40±0.45	1.50±0.44	1.31±0.20	1.31±0.09	1.27±0.03	1.43±0.05
	7	0.83±0.05	0.67±0.00	0.89±0.03	0.76±0.04	1.02±0.05	0.73±0.12	1.12±0.03
	9	0.41±0.03	0.80±0.03	0.45±0.00	0.92±0.01	0.32±0.04	0.22±0.01	0.32±0.01
	11	0.45±0.01	0.86±0.01	0.54±0.02	0.70±0.04	0.48±0.01	0.29±0.01	0.06±0.00

Table. 27 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells during menstrual cycle of monkey #101 in 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/5x10^4 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
early luteal phase (15 days prior menses of 37 days)	1	0.16±0.02	0.15±0.01	0.12±0.01	0.12±0.01
	3	0.13±0.03	0.13±0.02	0.09±0.02	0.12±0.01
	5	0.11±0.02	0.19±0.03*	0.08±0.01	0.12±0.01
	7	0.09±0.01	0.12±0.01*	0.06±0.01	0.12±0.02
	9	0.08±0.02	0.10±0.00	0.06±0.02	0.09±0.01
	11	0.08±0.01	0.10±0.01	0.06±0.01	0.07±0.01
mid luteal phase (8 days prior menses of cycle 30 days)	1	0.05±0.03	0.54±0.04	0.50±0.01	0.51±0.01
	3	0.32±0.01	0.67±0.02**	0.25±0.01	0.33±0.01
	5	0.25±0.01	0.48±0.02**	0.14±0.01	0.27±0.02
	7	0.23±0.02	0.40±0.01**	0.11±0.01	0.27±0.02
	9	0.20±0.01	0.39±0.20**	0.11±0.02	0.25±0.02
	11	0.15±0.01	0.35±0.20**	0.11±0.01	0.20±0.01
late luteal phase (2 days prior menses of cycle 32 days)	1	0.36±0.01	0.38±0.01	0.36±0.01	0.37±0.01
	3	0.28±0.02	0.45±0.01**	0.24±0.02	0.41±0.02
	5	0.22±0.02	0.28±0.01*	0.22±0.20	0.23±0.01
	7	0.21±0.01	0.28±0.20	0.15±0.02	0.14±0.02
	9	0.18±0.01	0.25±0.20	0.09±0.02	0.13±0.01
	11	0.11±0.01	0.14±0.01	0.06±0.01	0.12±0.01
luteolytic phase (2 days after menses of cycle 33 days)	1	0.13±0.00	0.13±0.01	0.11±0.01	0.15±0.01
	3	0.14±0.01	0.16±0.01	0.17±0.01	0.09±0.01
	5	0.09±0.01	0.16±0.01	0.13±0.01	0.14±0.01
	7	0.16±0.01	0.16±0.01	0.12±0.01	0.12±0.01
	9	0.14±0.01	0.15±0.01	0.17±0.01	0.10±0.01
	11	0.12±0.01	0.08±0.01	0.16±0.01	0.14±0.01

** = $P < 0.01$, * = $P < 0.05$ significantly different from the control.

Table 28 Effects fo hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells during menstrual cycle of monkey #75 in 11 days of incubation. (** = P < 0.01, * = P < 0.05 significantly different from the control (a), from the hCG-treated group (b).

stages	day of incubation	P secretion (pmol/5x10^4 cells)						
		control	hCG[a]	PRL	hCG+PRL	$PGF_{2\alpha}$[a]	hCG+$PCF_{2\alpha}$[b]	PRL+$PGF_{2\alpha}$
early luteal phase (18 days prior menses of cycle 45 days)	1	6.21±0.25	6.21±1.00	4.01±0.00	4.01±0.50	4.20±0.75	4.80±0.25	4.00±0.75
	3	10.80±1.00	48.00±5.00**	12.91±0.25	46.00±2.75	34.59±5.25**	42.00±5.76	23.09±1.24
	5	17.20±0.20	49.4±3.75**	18.25±1.00	48.79±5.24	17.77±1.00	32.77±6.00	16.50±2.00
	7	13.22±1.40	23.57±2.00	13.12±0.74	29.80±1.00	14.78±0.74	25.96±0.00	15.83±1.76
	9	9.65±0.42	15.99±1.03	12.36±0.56	22.04±1.75	13.66±1.26	16.47±1.24	13.44±2.75
	11	6.40±0.47	13.12±2.47	8.22±1.25	14.27±1.00	8.12±0.00	8.19±0.75	6.50±1.74
mid luteal phase (9 days prior menses of cycle 34 days)	1	64.78±7.75	78.41±10.00	61.59±3.20	76.82±2.64	60.00±10.82	60.00±2.00	64.01±7.23
	3	52.80±3.00	151.21±8.75**	65.61±2.68	142.80±11.30	77.07±4.80*	121.78±4.65	80.64±4.60
	5	39.17±8.26	139.43±7.64**	30.76±2.10	125.99±5.60	55.73±5.89*	113.78±11.11	41.98±8.00
	7	18.03±1.75	63.57±3.66**	17.45±2.85	36.12±2.01	20.70±4.76	50.00±2.10	19.75±5.08
	9	6.75±0.75	19.30±2.27	8.03±0.27	15.54±4.76	6.75±0.15	16.82±4.33	8.66±3.00
	11	5.89±1.25	12.83±0.99	5.54±0.90	12.77±1.64	6.37±1.26	13.41±3.28	6.21±1.00
late luteal phase (4 days prior menses of cycle 33 days)	1	50.00±4.90	47.99±12.00	47.99±7.30	50.00±1.70	41.97±11.00	38.41±4.90	38.41±7.30
	3	37.61±5.40	68.79±3.50**	40.19±8.40	75.60±7.8	35.32±3.50	58.79±6.80*	38.63±1.63
	5	27.30±1.40	68.89±8.00*	30.35±3.50	70.57±3.6	24.71±2.60	58.79±1.00*	34.43±3.60
	7	14.49±1.20	33.20±7.80	14.36±2.00	35.70±4.90	13.12±5.30	30.45±5.80	17.85±5.00
	9	10.00±3.90	29.71±3.60	10.03±2.64	31.08±5.30	12.77±4.00	28.34±2.10	11.08±3.24
	11	4.49±0.50	31.78±2.60	5.13±0.66	30.45±0.90	6.47±1.60	25.0±1.40	4.78±2.80

Table 29 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells during menstrual cycle of monkey #75 in 11 day incubation (mean±S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/5x10^4 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
early luteal phases (18 days prior menses of cycle 45 days)	1	0.27±0.02	0.29±0.01	0.25±0.03	0.27±0.03
	3	0.26±0.02	0.29±0.03	0.20±0.02	0.27±0.01
	5	0.20±0.03	0.45±0.03**	0.23±0.03	0.20±0.01
	7	0.12±0.02	0.35±0.03**	0.11±0.02	0.17±0.02
	9	0.11±0.01	0.15±0.01	0.19±0.01	0.15±0.03
	11	0.10±0.10	0.16±0.02	0.17±0.01	0.12±0.01
mid luteal phase (9 days prior menses of cycle 34 days)	1	0.52±0.01	0.58±0.02	0.59±0.03	0.59±0.05
	3	0.46±0.04	0.61±0.02*	0.40±0.02	0.45±0.03
	5	0.16±0.02	0.89±0.02**	0.21±0.01	0.16±0.01
	7	0.13±0.02	0.49±0.02**	0.11±0.01	0.16±0.02
	9	0.12±0.01	0.26±0.01	0.10±0.02	0.10±0.01
	11	0.08±0.17	0.11±0.01	0.09±0.02	0.07±0.02
late luteal phase (4 days prior menses of cycle 33 days)	1	0.43±0.03	0.45±0.02	0.46±0.03	0.42±0.01
	3	0.37±0.01	0.69±0.04**	0.35±0.05	0.49±0.02
	5	0.26±0.02	0.51±0.04**	0.24±0.05	0.31±0.01
	7	0.19±0.01	0.25±0.07*	0.16±0.03	0.16±0.02
	9	0.12±0.02	0.30±0.03	0.09±0.03	0.13±0.02
	11	0.07±0.01	0.16±0.01	0.07±0.01	0.08±0.01

** = $P < 0.01$, * = $P < 0.05$ significantly different from the control

Table 30 Effect of hCG, PRL and $PGF_{2\alpha}$ on P secreting ability of luteal cells during menstrual cycle of monkey #24 in 11 days of incubation. (** = $P < 0.01$, * = $P < 0.05$ significantly different from the control)

stages	day of incubation	P secretion (pmol/5x10^4 cells)						
		control	hCG	PRL	hCG+PRL	$PGF_{2\alpha}$	hCG+$PGF_{2\alpha}$	PRL+$PGF_{2\alpha}$
mid luteal phase (8 days prior menses of cycle 22 days)	1	5.60±2.10	6.00±1.30	6.60±1.15	6.00±2.10	6.00±1.10	7.00±1.95	6.00±0.95
	3	6.80±0.99	36.80±5.15**	6.56±0.99	31.20±7.60	17.60±2.55*	36.00±4.55	17.20±2.50
	5	4.00±1.51	40.80±3.90**	4.00±1.12	36.80±3.15	7.40±1.95	48.00±5.10	6.80±0.15
	7	2.00±0.61	24.20±4.50**	3.10±0.75	24.40±7.10	3.90±0.75	28.80±7.51	3.20±0.55
	9	3.40±0.93	14.80±2.65	3.00±0.25	9.20±2.00	4.80±1.00	18.00±3.70	3.00±1.50
	11	2.60±0.78	12.40±1.15	3.00±0.45	7.40±1.55	3.80±0.93	14.00±2.00	3.20±0.78
late luteal phase (5 days prior menses of cycle 21 dyas)	1	36.40±5.10	44.01±5.59	36.98±5.35	44.01±5.10	33.60±4.40	31.21±4.30	31.21±5.68
	3	27.20±3.35	56.18±7.25**	27.82±5.25	62.99±6.55	36.91±3.30	50.41±1.60	42.01±2.00
	5	17.20±4.10	59.46±6.00**	17.83±3.25	67.20±7.10	33.63±2.50**	52.52±4.20	40.32±8.76
	7	7.80±2.00	31.78±1.95**	7.41±1.00	33.19±6.00	18.85±1.60	30.73±5.89	19.33±4.76
	9	5.41±0.95	31.15±2.50**	4.94±0.95	36.75±2.65	16.40±2.71	27.01±6.22	16.37±1.58
	11	3.38±1.50	27.61±3.75**	3.39±0.05	30.45±1.25	17.64±1.00	21.15±2.85	12.71±3.66
luteolytic phase (the first day of menses).	1	2.56±0.10	2.88±0.55	2.16±0.35	1.60±0.21	2.20±0.34	2.32±0.12	2.96±0.22
	3	2.00±0.07	2.36±0.16	1.76±0.12	2.20±0.04	2.10±0.12	1.60±0.16	2.20±0.20
	5	1.32±0.05	2.08±0.10	1.50±0.05	2.08±0.65	1.22±0.17	1.48±0.10	1.30±0.04
	7	1.20±0.08	1.40±0.04	1.22±0.16	1.24±0.17	1.20±0.05	1.24±0.08	1.26±0.21
	9	1.10±0.10	1.40±0.43	1.24±0.43	1.50±0.21	1.2±0.24	1.10±0.21	1.20±0.43
	11	1.00±0.04	1.10±0.21	1.10±0.04	1.20±0.05	1.10±0.50	1.00±0.04	0.98±0.16

Table 31 Effects of hCG, PRL and $PGF_{2\alpha}$ on E_2 secreting ability of luteal cells during menstrual cycle of monkey #24 in 11 day incubation (mean ± S.E., n = 3)

stages	day of incubation	E_2 secretion (pmol/5x10^4 cells)			
		control	hCG	PRL	$PGF_{2\alpha}$
mid luteal phase (8 days prior menses of cycle 22 days)	1	0.23±.02	0.26±0.02	0.23±0.03	0.23±0.02
	3	0.20±0.03	0.36±0.02**	0.21±0.03	0.21±0.03
	5	0.17±0.02	0.31±0.01**	0.14±0.02	0.18±0.02
	7	0.18±0.00	0.16±0.01	0.16±0.01	0.19±0.01
	9	0.15±0.01	0.19±0.02	0.14±0.02	0.13±0.01
	11	0.13±0.02	0.16±0.01	0.12±0.01	0.13±0.01
late luteal phase (5 days prior menses of cycle 21 days)	1	0.46±0.01	0.50±0.01	0.48±0.04	0.48±0.04
	3	0.52±0.01	0.72±0.03**	0.47±0.05	0.55±0.05
	5	0.30±0.03	0.43±0.03**	0.30±0.02	0.33±0.01
	7	0.23±0.02	0.36±0.01**	0.29±0.02	0.24±0.02
	9	0.17±0.04	0.35±0.02**	0.16±0.03	0.17±0.01
	11	0.17±0.01	0.35±0.02**	0.18±0.01	0.15±0.02
luteolytic phase (the first day of menses	1	0.27±0.02	0.31±0.03	0.31±0.02	0.30±0.07
	3	0.23±0.01	0.30±0.01	0.21±0.02	0.25±0.02
	5	0.23±0.02	0.28±0.02	0.21±0.02	0.24±0.04
	7	0.22±0.01	0.25±0.02	0.23±0.03	0.23±0.02
	9	0.23±0.01	0.25±0.07	0.24±0.02	0.20±0.02
	11	0.21±0.02	0.15±0.02	0.20±0.03	0.19±0.03

** = $P < 0.01$, * $P < 0.05$, significantly different from the control.

Table 32 Responsiveness of monkey luteal cells to hCG, PRL and $PGF_{2\alpha}$ on P and E_2 secretion (pmol/5x10^4 cells/7 days) during 7 days incubation (mean±S.E., n = 3).

stages / treatments	early luteal phase		mid luteal phase		late luteal phase		luteolytic phase	
	P	E_2	P	E_2	P	E_2	P	E_2
#101								
control	32.0±4.8	0.48±0.01	254.9±31.2	1.30±0.21	92.9±12.1	1.07±0.02	5.1±1.1	0.52±0.12
hCG	63.5±7.9*	0.59±0.08*	351.0±53.1**	2.09±0.13*	187.7±44.2**	1.38±0.04*	NS 5.1±0.8	0.51±0.03
PRL	33.6±3.8	0.35±0.03	266.4±41.1	1.01±0.10	87.9±20.6	0.97±0.02	5.8±0.7	0.52±0.06
hCG+PRL	51.4±8.2	-	354.3±38.1		165.5±43.8		NS 5.1± 0.9	
$PGF_{2\alpha}$	42.6±6.2*	0.48±0.04	311.9±42.8**	1.38±0.04	NS 83.1±18.1	1.15±0.13	6.0±0.5	0.50±0.04
hCG+$PGF_{2\alpha}$	50.5±7.5		aNS 358.7±38.1		a* 138.4±22.2		5.4±0.7	
PRL+$PGF_{2\alpha}$	40.16±6.2		317.3±41.2		82.8±18.3		6.2±0.7	
#75								
control	47.4±5.2	0.84±0.15	174.8±0.18	1.27±0.18	129.3±16.6	1.25±0.30		
hCG	127.2±41.2**	1.39±0.01*	432.6±39.5**	2.56±0.09*	218.9±12.1**	1.91±0.12*		
PRL	48.3±8.5	0.79±0.18	175.4±18.2	1.31±0.14	132.9±19.2	1.20±0.11		
hCG+PRL	128.6±4.3		381.7±41.1		231.9±27.2			
$PGF_{2\alpha}$	71.3±9.5*	0.90±0.12	213.5±18.2*	1.36±0.05	115.1±12.1	1.38±0.18		
hCG+$PGF_{2\alpha}$	105.5±15.5		aNS 445.6±29.2		a* 186.4±33.2			
PRL+$PGF_{2\alpha}$	59.4±4.2		206.4±24.4		129.3±6.5			
#24								
control			18.4±3.8	0.78±0.05	88.6±5.1	1.56±0.19	7.1±0.2	0.94±0.08
hCG			107.8±12.9**	1.13±0.12*	191.4±30.0**	2.01±0.12*	NS 8.7±1.1	NS 1.14±0.13
PRL			20.2±2.8	0.73±0.19	90.4±6.4	1.54±0.12	6.6±0.7	0.96±0.02
hCG+PRL			98.4±2.3		207.4±14.3		7.1±0.3	
$PGF_{2\alpha}$			34.9±2.7*	0.80±0.01	157.0±7.9**	1.61±0.11	NS 6.7±0.8	1.02±0.04
hCG+$PGF_{2\alpha}$			119.8±14.6		aNS 164.87±21.2		6.6±0.2	
PRL+$PGF_{2\alpha}$			33.2±3.2		132.85±6.2		7.7±0.9	

* = P < 0.05, ** = P < 0.01, significantly different from the control.

a = ocmpared with the hCG-treated group

NS = non-significantly different.

BIOGRAPHY

Miss Ampa Luiengpirom was born on July 10,1949 in Nakornrajasima, Thailand. She graduated with the Bachelor degree of Science (Biology) in 1971 and the Master degree of Science (Biology) in 1973 from the Faculty of Science, Chulalongkorn University.