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# THE RECOVERY OF OVARIAN FUNCTION DURING BREAST-FEEDING

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Summary—The pattern of breast-feeding was daily recorded and the serum concentrations of prolactin (PRL), FSH, LH, estradiol ( $E_2$ ) and progesterone (prog) were measured at weekly intervals in 26 breast-feeding mothers from the time of delivery and up to the resumption of regular ovulation or to the end of the first postpartum year. Twelve postpartum non-breast-feeding women were similarly studied as controls. An algorithm was used to characterize ovulatory events into three types: the first, with evidence highly suggestive of normal ovulation (EHSO), the second, with evidence of probable ovulation (EPO) and the third with evidence indicating questionable ovulation or deficient corpus luteum function (QO/DCT). Pregnancy preceded the first menstruation in one woman in each of the breast-feeding and control groups. Of the 19 breast-feeding women who started to menstruate during the first postpartum year, five had EHSO, one had EPO, 5 had EQO/DCL and 7 had anovulatory (AO) menstruation. The corresponding figures in the 11 controls were 6, 2, 3 and 0. Pregnancy occurred before a second menstruation in one woman in both the study group and the controls. In 18 breast-feeding women observed, the second menstruation was preceded by EHSO in 7, by EPO in 3, by EQO/DCL in one and AO in 7. In 10 controls the corresponding figures were 7, 3, 0 and 0. Out of a total of 79 menstruations observed during breast-feeding the incidence of AO was 30% and of QO/DCL was 15%.

In actively breast-feeding mothers, hyperprolactinemia persisted for more than 1 yr. However, menstruation and ovulation occasionally occurred before the drop of PRL to concentrations seen during the normal menstrual cycle. In the majority of women, low  $E_2$  levels were present during lactational amenorrhea, but with occasional spikes in some. A few women maintained somewhat high values of  $E_2$  for several weeks before the resumption of menstruation. The implications of these hormonal findings to the attempts to improve on the contraceptive effect of breast-feeding are discussed.

#### INTRODUCTION

A clearer understanding of the sequence of events involved in the recovery of ovarian function during breast-feeding must be sought before the full potential of breast-feeding as an effective method of contraception can be exploited. This will also indicate ways in which this contraceptive effect can be made more reliable. McNeilly and Howie have revived interest in this area by a number of publications [1-3]. The present work is an attempt to gather more information on the physiology of lactation by the measurement, at weekly intervals, of the serum concentrations of the follicle-stimulating hormone (FSH), luteinizing hormone (LH), prolactin (PRL), estradiol  $(E_2)$  and progesterone (prog) in a group of breast-feeding women from the time of delivery and up to the resumption of regular ovulation. This is part of a study supported by Family Health International, NC, U.S.A.

The present article gives the hormonal changes during breast-feeding as compared with those in the controls. The correlation between the breast-feeding pattern and the hormonal events will be the subject of another communication.

#### MATERIAL AND METHOD

Women were recruited for the study at the time of delivery in the obstetric service of the Assiut University Hospital. They were healthy and aged 18-40 yr. They were all parous and had successfully breast-fed one or more previous child. They had normal delivery of a normal singleton fetus and were entered to the study before the 15th postpartum day. They intended to breast-feed as long as possible and not to introduce supplementary feeds for a minimum of 3 months. They breast-fed on demand day and night. The women studied gave their infants an average of seven breast-feeds during day-time and four during night-time. There was no decline in the average number of breast-feeds with passage of time in those women who continued in the study up till the end of the first postpartum year. They had regular sexual relation with husbands and none of them used any contraceptive.

The women and their babies were seen at weekly intervals. The infant-feeding practice during the previous week was obtained from a simple pictorial chart that covers the numbers of all types of feeds given to the infant during the day and night.

At each visit, 8 ml of venous blood was withdrawn from the mother 2 h or more after the last breast-feed. The serum was separated and stored frozen in small aliquots until assayed for the concen-

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tration of FSH, LH, PRL, E2 and prog. These measurements were done by radioimmunoassays using reagents obtained from the World Health Organization Special Programme for Research in Human Reproduction. The assay performance was continuously monitored by an internal quality control scheme. The follow-up was maintained up to the occurrence of one of the following end points: (1) pregnancy, (2) hormonal evidences of two successive ovulatory menstrual cycles (see below), or (3) the end of the first postpartum year. Twenty-six breast-feeding and 12 non-breast-feeding mothers completed this scheme. Some of the lactating mothers were kept in the study beyond the specified end points, whenever they agreed to, in order to gain more information about the ovarian cycles during breast-feeding.

## Normal hormone value for the population

The normal levels of the five hormones involved in this study had been previously determined for the local population in Assiut, Egypt [4]: 20 regularly menstruating non-lactating females aged between 19 and 23 yr gave daily blood samples for one complete ovulatory menstrual cycle. The results of the cycles were synchronized with one another according to the day of the LH midcycle peak which was designated as day 0 for each cycle. E<sub>2</sub> midcycle peak occurred in the day of the LH peak or in the day before (day 1). These two days were designated as the peak phase of the cycles. Days from day -2 and up to the first day of the cycle were considered to represent the follicular phase, while days from day +1 and up to the last day in the menstrual cycle represented the luteal phase. Table 1 gives the mean concentrations of the five hormones during these three phases of the menstrual cycle.

The values of LH, FSH,  $E_2$  and prog during the peak and the luteal phases were scrutinized in individual cycles to determine the levels that can indicate ovulation and adequate function of corpus luteum. It is very difficult to designate an algorithm by which one can be certain of ovulation and can characterize the adequacy of the corpus luteum by measurement of these four hormones at weekly intervals. However, three categories of ovulatory events are designated for the sake of the present study: I. Criteria highly suggestive of normal ovulation (EHSO) (if all present):

(1) a serum concentration of prog of 5 ng/ml or more in one blood sample if followed by the onset of menstruation within 14 days;

(2) an  $E_2$  value of 100 pg/ml or more in the same sample;

(3) obtaining in the blood sample obtained 1 week before the previously mentioned specimen one or more of the critical mid-cyclic hormonal events:

a-LH value of 25 IU/l or more.

b-FSH value of 6 IU/l or more.

c-E<sub>2</sub> values of 200 pg/ml or more.

- II. Criteria indicating occurrence of probable ovulation (EPO); presence of the first criterion in the previous category with absence of one or both of the other criteria.
- III. Criteria indicating questionable ovulation or deficient corpus luteum (QO/DCL): This is indicated by obtaining in one sample a serum prog value of 3-4.9 ng/ml without a previously higher value and the onset of menstruation after less than 7 days from the day of this highest prog value.

## RESULTS

# A. Menstruation during breast-feeding

Out of the 26 breast-feeding women studied, 19 (73%) started to menstruate during the first postpartum year. Table 2 gives the characteristics of the early menstrual cycles during breast-feeding as compared with the control group of women who did not breast-feed after delivery. In the former group the first menstruation occurred 73–267 days after delivery. The subsequent menstruation occurred after 22–185 days from the first one. One woman conceived before starting any menstruation and five did not menstruate up till the end of the first postpartum year.

## B. Ovulatory events

Table 3 gives the cumulative probability of menstruation and ovulation in the breast-feeding group (in this table events with EHSO and EPO are

Table 1. Serum concentrations of FSH, LH, Prl, E2 and Prog in normal ovulatory cycles (means  $\pm$  SD)

Phase of cycle	FSH	LH	Prl	E <sub>2</sub>	Prog
	(IU/l)	(IU/l)	(ng/ml)	(pg/ml)	(ng/ml)
Follicular phase* Peak phase* Luteal phase*	$4.7 \pm 1.5$ $8.7 \pm 3.9$ $3.4 \pm 1.8$	$ \begin{array}{r} 10.3 \pm 2.5 \\ 30.4 \pm 1.8 \\ 8.5 \pm 3.6 \end{array} $	$11.6 \pm 3.8$ $15.1 \pm 7.5$ $14.6 \pm 6.0$	$72 \pm 34$ 207 ± 61 103 ± 19	$0.42 \pm 0.1 \\ 1.00 \pm 0.5 \\ 5.20 \pm 3.0$

\*The three phases are defined in the text.

	Breast-fee	ding group	Nonbreast-feeding group		
	Length of cycle	Duration of period	Length of cycle	Duration of period	
Delivery-first menstruation interval	$133.8 \pm 61.0$ (n =	$2.6 \pm 0.9$ 19)*	$51.8 \pm 15.0$	$2.3 \pm 1.0$	
2nd cycle	$40.4 \pm 30.9$	$3.1 \pm 0.6$	$35.2 \pm 7.2$	$3.1 \pm 0.6$	
3rd cycle	$47.0 \pm 30.9$ ( <i>n</i> =	$3.0 \pm 0.6$	$34.0 \pm 8.5$ (n	$3.2 \pm 1.2$	
4th cycle	$34.8 \pm 8.9$ ( <i>n</i> =	$2.5 \pm 0.8$	too few subj	ects observed	

Table 2. Characteristics of the early postpartum menstrual cycles in the breast-feeding group and the controls (means  $\pm$  SD)

\*Figures in parenthesis indicate the number of cycles observed.

Table 3. Cumulative probability of menstruation and ovulation during breast-feeding

Time (weeks)	Percentage of all cases $(n = 26)$		
Menstruation			
At 12 weeks	7 (26.9%)		
At 24 weeks	14 (53.8%)		
At 36 weeks	19 (73.0%)		
At 48 weeks	20 (76.9%)		
At 60 weeks	21 (80.7%)		
Ovulation			
At 12 weeks	3 (11.5%)		
At 24 weeks	9 (34.6%)		
At 36 weeks	17 (65.4%)		
At 48 weeks	19 (73.0%)		
At 60 weeks	20 (76.9%)		

grouped together). The ovulatory events that occurred in the breast-feeding group are given in Table 4. It seemed that the woman who got pregnant during lactation amenorrhea conceived during the 14th postpartum week. By that time she was giving an average of 10 breast-feeds per day; but she gave also an average of 3.4 supplementary formula feeds and 2 feeds of other fluids. An appreciable drop in her serum prolactin concentration preceded the ovulation (from 70 ng/ml at the eleventh postpartum week to 31 ng/ml at the 13th week).

Of the 19 women who started to menstruate during the first year of breast-feeding, 5 had EHSO, one EPO, 5 QO/DCL and 7 had anovulatory (AO) menstruation. The corresponding figures in the controls were 6, 2, 3 and 0. Pregnancy occurred before the second menstruation in one woman in each of the study group and the controls. In 18 breast-feeding women observed through the second postpartum menstruation, there were EHSO in 7, EPO in 3, QO/DCL in one, and 7 had AO menstruation. In 10 controls the second menstruation was preceded by EHSO in 7 and by EPO in 3.

## C. Changes in hormonal levels during breast-feeding

Figure 1 shows the means of serum concentrations of prolactin, FSH, LH,  $E_2$  and prog during breast-

feeding. One week after delivery the mean basal concentration of prolactin was  $85.2 \pm 44.7$  ng/ml which was significantly higher (P < 0.001) than the mean value observed in the controls ( $37.3 \pm 12.4$  ng/ml). By the second week of lactation, the mean prolactin concentration was  $63.4 \pm 43.4$  ng/ml, and it was  $53.6 \pm 24.6$  ng/ml at the 12th week,  $32.2 \pm 16.0$  ng/ml at the 24th and  $27.0 \pm 12.8$  ng/ml at the 48th.

In the breast-feeding group the mean serum  $E_2$  remained below 100 pg/ml with a small SD up till the end of the 5th postpartum month. Thereafter, fluctuations were observed but the mean levels were still below 100 pg/ml in the majority of weeks up to the end of 53rd postpartum week.

## Hormonal changes preceding resumption of menstruation

Table 5 presents the means of the changes in the concentrations of PRL, E2, prog, FSH, LH during the weeks preceding the first postpartum menstruation. The week of onset of menstruation is designated as week 0 and the preceding weeks are given minus signs, e.g. week -1 and week -4. The paired t-test was used to assess the significance of the change. The week of onset of menstruation showed significant drop in  $E_2$  and prog concentrations and an insignificant drop in PRL concentration relative to the preceding week. When week -1 was compared with week -2 there was not significant change in hormonal levels except in FSH that showed a significant decrease during week -2 (P < 0.05). When week -2 was compared with week -4, the former showed higher concentrations of  $E_2$  and prog than the latter, but there were no significant differences in the other hormones.

Examination of the breast-feeding pattern and hormonal levels in individual cases has shown the following observations:

(1) Rapid decline in the number of breast-feeds per 24 h was associated with rapid return of menstruation and ovulation (Fig. 2).

	Total	Pregnancy	EHSO*	EPO*	EQO*	AO*	Results not complete
Before the first period	20	1 (5%)	5 (25%)	1 (5%)	5 (25%)	7 (35%)	1 (5%)
2nd cycle	19	(5.2%)	7 (36.8%)	3 (15.8%)	1 (5.3%)	7	( )
3rd cycle	13	(7.7%)	(23.0%)	2 (15.4%)	3	(30.9%)	
4th cycle	8	2 (25%)	(25.670)	(131,1,0) 1 (21,5%)	(2370)	(12,5%)	
5th cycle	6	(2070)	(83.3%)	(21.570)		1	
6th cycle	2		(05.5%)			(10.0%) 1 (50%)	
7th cycle	2		(50%)			1	
8th cycle	2		(3070)		1	(50%)	
9th cycle	1				(30%)	(30%) 1 (100%)	
10th cycle					1	(100%)	
11th cycle	1				(100%)		
12th cycle	1		1		(100%)		
13th cycle	1		(100%) 1 (100%)				
14th cycle	1		(100%)				
15th cycle	1		(100%) 1 (100%)				
Total	79	5 (6.5%)	30 (40.0%)	7 (8.90%)	12 (15.2%)	24 (30,4%)	1 (1.3%)

Table 4. Incidences of ovulation and pregnancy in women who resumed menstruation during breastfeeding

\*EHSO—Evidence highly suggestive of ovulation.

EPO-Evidence of probable ovulation.

EQO-Evidence of questionable ovulation and/or luteal phase deficiency.

AO-Anovulatory menstruation.

	Week of menstruation vs the preceding week (week -1)	Week -1 vs week -2	Week -2 vs week -4
1. Prolactin	$-2.0 \pm 3.56$	$-3.428 \pm 3.101$	$-1.88 \pm 2.54$
	(n = 16)	(n = 14)	(n = 18)
2. Estradiol	$-37.11 \pm 11.58^{**}$	$0.2 \pm 65.21$	$50.3 \pm 13.53^{**}$
	(n = 18)	(n = 15)	(n = 18)
3. Progesterone	$-2.98 \pm 0.72^{**}$	$1.58 \pm 0.81$	$1.95 \pm 0.57^{**}$
U	(n = 18)	(n = 15)	(n = 19)
4. FSH	$0.88 \pm 0.812$	$-3.0 \pm 0.866*$	$0.88 \pm 1.55$
	(n = 18)	(n = 15)	(n = 17)
5. LH	$-0.61 \pm 1.29$	$-2.86 \pm 2.26$	$0.611 \pm 2.568$
	(n = 18)	(n = 15)	(n = 18)

Table 5. Mean change ± SEM in the hormonal levels during the weeks preceding menstruation

Figures in parenthesis indicate number of subjects in whom the comparison was made. \*Significantly different, with P < 0.05. \*Significantly different, with P < 0.01.



Fig. 1. Serum concentrations (mean ± SD) of prolactin, FSH, LH, estradiol and progesteron during breast-feeding.

(2) Continuation of frequent breast-feeding in spite of substantial supplementation maintained the hyperprolactinemia and prolonged the ovarian suppression (Figs 3 and 4).

(3) Ovulation occasionally occurred in spite of high serum prolactin concentration (Fig. 5). However, a rapid decline of prolactin concentration even if not to levels seen in non-lactating women may herald the return of menstruation and ovulation (Fig. 2).

(4) There was some variability in estradiol levels during lactational amenorrhea. Some women showed consistently low  $E_2$  levels up to the 2 or 3 weeks preceding ovulation (Fig. 3). A second group maintained a low  $E_2$  level but with occasional spikes (Fig. 6). A third group maintained a somewhat high

 $E_2$  level (near 100 pg/ml or above) for several weeks before starting to menstruate (Fig. 4).

(5) In four instances in the 26 breast-feeding women hormonal changes that can suggest ovulation were observed during lactational amenorrhea, i.e. without being following by menstruation; two examples are depicted in Figs 6 and 7.

## DISCUSSION

#### Menstruation and ovulation during breast-feeding

The duration of lactational amenorrhea was on the average  $19 \pm 8$  weeks. This mean would have been higher had the women who completed the first postpartum year without resuming menstruation



Fig. 2. Infant feeding pattern and hormonal levels: the rapid decline in the number of breast-feeding episodes is followed by return of ovulation. (G.E.—gastroenteritis of infant; VON—volunteer order number.)

been included. The early menstrual cycles during lactation tended to be long and irregular.

The cumulative probability of menstruation in the present study was higher than that reported by Rivera *et al.*[5] for a similar number of subjects in rural Mexico, but the cumulative probability of

ovulation in our study was not higher than in the Mexican one. The latter study monitored the subjects by measurement of the overnight urinary excretion of pregnandiol at weekly intervals. This may mean that more of the menstruations observed in the present study were anovulatory. Another



Fig. 3. Infant feeding pattern and hormonal levels: continuation of frequent putting of the baby to the breast in spite of supplementary formula maintained ovarian suppression.



Fig. 4. Infant feeding pattern and hormonal levels during the later part of the first postpartum year: continuation of frequent putting of the baby to the breast in spite of giving supplementary solids prolonged suppression of ovulation and menstruation. However, estradiol values were elevated, indicating follicular growth.



Fig. 5. Infant feeding pattern and hormonal levels: resumption of ovulation and menstruation in spite of high prolactin level.



Fig. 6. Infant feeding pattern and hormonal levels during the later part of the first postpartum year: occasional spikes of estradiol and progesterone during lactational amenorrhea.



Fig. 7. Infant feeding pattern and hormonal levels: hormonal pattern suggestive of ovulation during lactational amenorrhea.

possible explanation of this difference between the two studies may be that the criterion adopted as indicative of ovulation by Rivera *et al.* is less stringent than that used in the present study. To our knowledge, this is the first study that documents a longitudinal assessment of the blood levels of ovarian hormones and the related pituitary hormones during breast-feeding. Previous studies have measured the urinary output of estrogen and progesterone metabolites either in 24-hr [6] or overnight specimens [5]. Urinary pregnandiol output will not always offer a reliable assessment of endogenous production of progesterone, and this view has been cogently argued by Klopper and Billewicz[7].

The weekly measurement of FSH, LH, E<sub>2</sub> and prog in blood in the present study has offered more opportunity to diagnose and characterize ovulatory events during lactation. Pregnancy preceded the return of menstruation in only one of the 26 women studied. However, ovulation as suggested by obtaining a serum progesterone concentration of 5 ng/ml or more was observed in six other subjects, giving an incidence of ovulation before the resumption of menstruation of 23%. Gross and Eastman [8] have, in a prospective study of breast-feeding Australian mothers, found that 30% had hormonal confirmation of ovulation before the first menstruation. Brown et al.[6] have found in another group of Australian breast-feeding mothers that normal ovulation preceded the first bleed in 19%. Our results are in close agreement with those reported in the previous study of Howie et al.[2], but contrast with the high incidence of 78% reported in Chilean mothers [9]. However, it has to be noted that in the latter study there was great discrepancy between this rate of ovulation and a low rate of pregnancy of 7% in the same women. With the rate of ovulation of 23% observed in the present study, and taking the currently accepted figure that unrestricted intercourse during normal ovulatory cycles would result in pregnancy in 25% of them, the overall conception rate expected for a couple who are trying to avoid pregnancy up to the time of the first postpartum menstruation would be approximately 5.7%. This is near the approximate figure which has been given by Short[10].

The incidence of ovulation markedly increased after the first menstruation during lactation, emphasizing the need to use a more reliable contraceptive after this occurrence.

## Inadequate ovarian cycles during breast-feeding

Out of 79 ovarian cycles ending in menstruation observed in the breast-feeding mothers, 15% showed evidence suggestive of imperfect ovarian cycles. These were taken as a rise of progesterone concentration to a level above 3 ng but less than  $5 \mu g/ml$ , usually without a concomitant rise in estradiol. In some of these inadequate cycles this small rise of progesterone was shortly followed by onset of menstruation suggestive of shortened luteal phase. These features can equally suggest either luteinization of unruptured follicle (LUF) or deficient corpus luteum function. It is probable that some of the cycles that showed evidence of probable ovulation may be representing more incidents of LUF or luteal phase deficiency. This phenomenon has previously been observed in breast-feeding women [1, 11] and has been suggested as one of the mechanisms by which fertility is reduced during lactation. The association of hyperprolactinemia of physiological or pathological origin with inadequate luteal function has been previously documented [12– 15].

## Hormonal levels during lactational amenorrhea

Hyperprolactinemia is the denominator of active breast-feeding and of the concomitant suppression of ovarian functions. The present study has demonstrated the persistence of hyperprolactinemia for more than 12 months in actively breast-feeding mothers. However, it has also shown that menstruation and ovulation ocassionally occurred in spite of serum prolactin values much higher than the normal values observed in non-lactating women. It seems that the range of prolactin against which ovariant function can be resumed during breastfeeding is wide enough to preclude defining a certain critical level of prolactin above which the mother is reasonably protected from pregnancy. This will make it difficult to use the concentration of prolactin in blood as an indicator of return of fertility during lactational amenorrhea as suggested by Gross and Eastman[14].

The present study has demonstrated some variability in  $E_2$  levels during lactational amenorrhea. This has its practical implications: The group of women who consistently have suppressed  $E_2$  levels will be good candidates for the use of cervicovaginal mucus observation methods for natural family planning (NFP) as a means of augmenting the contraceptive effectiveness of breast-feeding. The group that showed occasional  $E_2$  spikes may experience false alarms during the use of such methods during lactation. The situation will be most difficult with the third group, who maintain an elevated  $E_2$  level during lactational amenorrhea.

These varying patterns of estrogen secretion during lactational amenorrhea explain the clinical observation of the varying extent of uterine involution during this period. This is commonly observed at the measurement of the uterine cavity before inserting an IUCD in women with lactational amenorrhea. In some women the uterine cavity is of normal length and in others it is so compromised as to necessitate special care during the insertion. Devising simplified methods for measuring the estrogen level in body fluids like urine or saliva can augment the reliability of breast-feeding as a contraceptive, particularly if these can be brought to the practicability of do-it-yourself kits.

The ovarian production of progesterone is not always suppressed during lactational amenorrhea; levels suggestive of ovulation with concomitant rise in  $E_2$  were occasionally seen. This indicates that factors other than resumption of ovarian activity may be contributing to lactational infertility and amenorrhea. Lack of responsiveness of the endometrium and may be also the cervix may be contributing to this effect.

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#### REFERENCES

- McNeilly A. S., Howie P. W., Houston M. J.: Relationship of feeding patterns, prolactin and resumption of ovulation postpartum. In *Research Frontiers in Fertility Regulation* (Edited by G. I. Zatuchni, M. H. Labbock and J. J. Sciarra). Harper & Row, New York, Ch. 12 (1980) pp. 102-116.
- Howie P. W., McNeilly A. S., Houston M. J., Cook A. and Boyle H.: Fertility after childbirth: infant feeding patterns, basal prolactin levels and postpartum ovulation, *Clin. endocr.* 17 (1982a) 315-322.
- Howie P. W., McNeilly A. S., Houston M. J., Cook A. and Boyle H.: Fertility after childbirth: postpartum ovulation and menstruation in bottle and breast-feeding mother. *Clin. endocr.* 17 (1982b) 323-332.
- Shaaban M. M., Hammad W. A., Chaneimah S. A., Salem H. T. and Gomaan A. A.: Serum prolactin concentration during ovulatory menstrual cycle. J. Egypt Soc. obstet. Gynec. 2 (1980) 19-27.
- 5. Rivera R., Ortiz E., Barrera M., Kennedy K. and

Bhiwandiwala P.: Preliminary observations on the return of ovarian function among breast-feeding and postpartum non-breast-feeding women in a rural area of Mexico. J. biosoc. Sci. Suppl. 9 (1985) 127-136.

- Brown J. B., Harrisson P. and Smith M. A.: A study of returning fertility after childbirth and during lactation by measurement of urinary estrogen and pregnanediol excretion and cervical mucus production. J. biosoc. Sci. Suppl. 9 (1985) 5-23.
- 7. Klopper A. and Billiwicz W. J.: Obstet. Gynec. Br. Commonw. 70 (1963) 1024.
- 8. Gross B. A. and Eastman C. J.: Effect of breastfeeding status on prolactin secretion and resumption of menstruation. *Med. J. Aust.* **1** (1983) 313.
- 9. Perez A., Vela P., Masnick G. S. and Potter R. G.: First ovulation after childbirth: the effect of breast-feeding. *Am. J. obstet. Gynec.* **114** (1972) 1041–1047.
- Short R. V.: The biological basis for the contraceptive effect of breast-feeding. In Advances in International Maternal and Child Health. (Edited by Jelliffe and Jelliffe). Oxford University Press, Oxford, Vol. 3 (1983).
- Gross B. A. and Eastman C. J.: Prolactin and the return of ovulation in breast-feeding women. J. biosoc. Sci. Suppl. 9 (1985) 25-42.
- Del Pozo E., Wyss H., Lancranjan I., Obolensky W. and Varaga L.: Prolactin-induced luteal insufficiency and its treatment with bromocriptine: preliminary results. In *Ovulation in the human* (Edited by P. G. Crosignani). Academic Press, London (1976).
- Seppala M., Hirvonen E. and Ranta T.: Hyperprolactinaemia and luteal insufficiency. *Lancet* i (1976) 229.
- Corenblum B., Pairaudeau M. and Shewchuk A. B.: Prolactin hypersecretion and short luteal phase defects. Obstet. Gynec. 47 (1976) 486.
- Tyson J. E.: Nursing and prolactin secretion: principal determinants in the mediation of puerperal infertility. In *Prolactin and Human Reproduction* (Edited by P. G. Crosignami and C. Rabyn) (1977).