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# CHANGES IN ADENOHYPOPHYSIAL CELLS AND LEVELS OF SOMATOTROPHIN AND PROLACTIN AT DIFFERENT REPRODUCTIVE STAGES IN THE PIG*

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**Summary.** Percentages of cell types in the adenohypophysis were compared with the content of STH and prolactin at different reproductive stages (e.g. immature, oestrous cycle, pregnancy, lactation and after hysterectomy) in the pig. Acidophils represent STH and prolactin activities. Chromophobes are undifferentiated, inactive or depleted cells.

Acidophils represented half (53·5%) the cell population. Percentages of STH cells were higher in 17-day-old pigs than in mature animals. This is a period of rapid growth in the young pig. During the cycle, during pregnancy and after hysterectomy, the percentages of prolactin cells were higher than those found in immature or lactating pigs.

The chromophobes represented 13·1% of the cell population. Chromophobes were the predominant cell type during lactation, but these seemingly inactive cells may have been active acidophils that were synthesizing and secreting prolactin. By rapid turnover of hormone, the cytoplasmic granules in prolactin cells lack differentiation.

The STH activity was similar in pigs during pregnancy and lactation and after hysterectomy. There were no significant correlations in the percentages of STH cells and the concentration or content of STH during pregnancy and lactation. There was a trend of increased percentages of prolactin cells in later stages of pregnancy and after hysterectomy. Exogenous oestrogen caused a marked increase in the pituitary content of STH and prolactin in hysterectomized pigs as compared with uninjected hysterectomized animals. Prolactin levels remained relatively constant during pregnancy and after hysterectomy, but declined during lactation. Exogenous oestrogen caused a consistently higher trend in prolactin activity in hysterectomized animals.

## INTRODUCTION

Adenohypophysial concentrations of gonadotrophins change markedly during the oestrous cycle, pregnancy, lactation and after hysterectomy in the pig (Parlow, Anderson & Melampy, 1964; Melampy, Henricks, Anderson, Chen & Schultz, 1966). Recent evidence indicates that serum levels of porcine LH

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remain extremely low throughout the cycle and increase for a brief period at oestrus (Niswender, Reichert & Zimmerman, 1970). Differences in the pituitary gland content and blood level of a gonadotrophin, therefore, are useful for evaluating the storage and secretion of a hormone. Histological examination of the different cell types in the adenohypophysis also gives an indication of storage of a trophic hormone. Plasma levels of somatotrophin (STH) have been determined in a few developmental stages in the pig (Kipnis, Hertelendy & Machlin, 1969) but data are unavailable for the different reproductive stages. Plasma levels of porcine prolactin also are unavailable.

We have examined the acidophils in the adenohypophysis of the pig and compared the percentages of these cell types with the content of STH and prolactin in the pituitary gland at different reproductive stages (e.g. immature, oestrous cycle, pregnancy, lactation and after hysterectomy). The STH activity was measured by epiphysial growth of the tibia in the rat and the prolactin activity was determined by stimulation of the crop-sacs in the pigeon.

## MATERIALS AND METHODS

### *Animals*

Crossbred pigs, weighing 90 to 110 kg and 5 to 7 months old, were checked daily with vasectomized boars for oestrous behaviour. Normal oestrous cycles of 18 to 23 days were completed before assigning the animals to treatment groups.

The pituitary glands of forty-one pigs were prepared for biological assay of STH and prolactin activities. Sixteen of these animals were mated and killed on Days 40, 80 or 110 of pregnancy and on Day 14 of lactation. Twenty-five others were hysterectomized on Days 8, 9 or 10 of the oestrous cycle and killed on Days 40, 80 or 110. Thirteen of these hysterectomized animals were given oestradiol benzoate (5 mg/day in corn oil, intramuscularly) for 12 days before the end of the experimental period. The CL were marked with a silk suture at hysterectomy to identify them at slaughter.

Pituitary glands of seventy-three pigs were prepared for histological identification of cell types during different reproductive stages. Fetal and prepubertal glands were examined from twelve animals. Pituitaries from twenty-one pigs were collected on Days 1, 4, 10, 16 or 18 of the oestrous cycle, and twenty-three mated animals were killed at three stages: early gestation (Days 12 to 40), midgestation (Days 41 to 80) and late gestation (Days 89 to 110). The pituitaries of four pigs were collected on Day 14 of lactation. Thirteen pigs were hysterectomized on Days 8, 9 or 10 of the cycle and killed on Days 24 to 26, 41 to 47 or 80 to 93.

### *Preparation of pituitary glands*

The gland was removed from the sella turcica within 15 min of killing the pig. The whole pituitary gland weight was recorded, and the anterior lobe was dissected free and fresh-frozen for biological assay of STH and prolactin activities. Whole pituitary glands for histology were immersed in fresh Susa's fixative for

TABLE 1

MORPHOLOGICAL DATA ON THE OVARY AND PITUITARY GLAND AT DIFFERENT REPRODUCTIVE STAGES IN THE PIG

| *Reproductive stage* | *No. of pigs* | *Pituitary gland fresh weight (mg)** | | *Corpora lutea** | | *Total ovarian weight* (g)* | *Ovarian follicle diameters (mm)** | | *No. of fetuses* | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | *Whole gland* | *Anterior lobe* | *No.* | *Weight (mg)* | | *< 4 mm* | *4 to 6 mm* | *Live* | *Dead* |
| Pregnancy | | | | | | | | | | |
| Day 40 | 4 | 334 | 228 | 11·0 | 425 | 12·2 | 56 | 9 | 9 | 0·8 |
| Day 80 | 4 | 342 | 261 | 11·8 | 479 | 12·6 | 53 | 11 | 9 | 0·5 |
| Day 110 | 4 | 446 | 340 | 10·8 | 519 | 12·4 | 64 | 10 | 7 | 0·5 |
| | | 374±13 | 276±9 | 11·2±0·7 | 474±36 | 12·4±0·7 | 58±5 | 10±1·6 | | |
| Lactation | | | | | | | | | | |
| Day 14 | 4 | 506 | 397 | 0 | 0 | 6·6 | 37 | 0 | | |
| Hysterectomy† | | | | | | | | | | |
| Day 40 | 4 | 314 | 234 | 11·0 | 463 | 13·6 | 46 | 5 | | |
| Day 80 | 4 | 309 | 226 | 12·8 | 571 | 15·7 | 51 | 7 | | |
| Day 110 | 4 | 386 | 247 | 11·8 | 689 | 15·1 | 34 | 11 | | |
| | | 336±13 | 235±9 | 11·8±0·7 | 574±36 | 14·8±0·7 | 43±5 | 7±1·6 | | |
| Hysterectomy† and oestradiol benzoate‡ | | | | | | | | | | |
| Day 40 | 4 | 393 | 308 | 13·5 | 449 | 9·9 | 26 | 0 | | |
| Day 80 | 5 | 377 | 262 | 12·4 | 503 | 11·4 | 36 | 0 | | |
| Day 110 | 4 | 455 | 336 | 12·5 | 552 | 14·0 | 38 | 11 | | |
| | | 408±13 | 301±9 | 12·4±0·7 | 501±36 | 11·8±0·7 | 33±5 | | | |

* Group means and standard error are listed.
† Animals were hysterectomized on Days 8 to 10 of the oestrous cycle.
‡ Oestradiol benzoate (5 mg/day in corn oil) was injected intramuscularly daily for 12 days before termination of the experimental period.

15 min, cut transversely into thirds and replaced in the fixative for 24 hr. They were washed in running tap water for 24 hr, placed in 50% ethanol and stored in 70% ethanol.

*Biological assay of somatotrophic hormone activity*

The bioassay consisted of the increase in width of the rat epiphysis in response to standard hormone and test preparations. A standard growth hormone preparation (NIH-GH-B-7, bovine), which contained 0·88 units/mg, was compared with three levels of the test preparation. The test and standard hormone preparations were diluted in saline and injected intraperitoneally once daily for 4 days into female rats, hypophysectomized at 28 to 30 days of age. The rats were killed the day after the last treatment, and both tibiae were dissected and prepared for measurement of epiphysial width. Ten rats were assigned to each treatment level.

*Biological assay of prolactin activity*

The glandular proliferation of the crop-sacs of young pigeons was used to measure prolactin activity in standard hormone and test preparations. The test preparations were diluted in saline and compared with responses of the standard prolactin (NIH-P-B1, bovine). The test and standard preparations were injected intradermally over the crop-sacs once daily for 4 days. Each pigeon was injected at two different sites, and the birds were killed the day after the last injections. The crop-sacs were removed and examined by transmitted light, and the stimulated area was measured. Ten pigeons were assigned to each treatment level.

*Histological sections of pituitary glands*

After dehydration and embedding in paraffin, the tissues were cut at 4 to 5 $\mu$m and stained by a modification of the aldehyde–thionin–PAS (periodic acid–Schiff) technique as described by Ezrin & Murray (1963). Before staining these sections with orange G, haematoxylin was included to differentiate acidophils. Every tenth section was mounted, and every sixth section was counted; therefore, sections were counted at intervals of 240 $\mu$m. For counting the cells, a starting point was chosen randomly and, thereafter, every tenth field across and down the section was counted.

*Statistical analyses*

The results of the biological assays for STH and prolactin activities were calculated by the method described by Finney (1952) for parallel line assays. The relative potencies of these two hormones were expressed as the geometric means and their confidence limits. Mean concentrations and contents of these hormones are presented (Table 2). Morphological data on the ovary and pituitary gland were analysed by analysis of variance.

Statistical analyses of the counts of the cell types within the adenohypophysis at different reproductive stages were determined by the Scheffe test (Walker & Lev, 1969).

TABLE 2

ANTERIOR PITUITARY SOMATOTROPHIC HORMONE AND PROLACTIN ACTIVITIES DURING DIFFERENT REPRODUCTIVE STAGES IN THE PIG

| *Reproductive stage* | *No. of pigs* | *Somatotrophic hormone activity* | | | *Prolactin activity* | | |
|---|---|---|---|---|---|---|---|
| | | *Adenohypophysial** | | | *Adenohypophysial§* | | |
| | | *Concentration (μg/mg)* | *95% Confidence interval* | *Content (μg)* | *Concentration (i.u./mg)* | *95% Confidence interval* | *Content (i.u.)* |
| Pregnancy | | | | | | | |
| Day 40 | 4 | 20·0 | 6·5 to 61·3 | 4560 | 0·099 | 0·073 to 0·135 | 22·6 |
| Day 80 | 4 | 30·0 | 13·8 to 65·3 | 7830 | 0·111 | 0·078 to 0·159 | 29·0 |
| Day 110 | 4 | 13·2 | 3·3 to 53·8 | 4488 | 0·090 | 0·074 to 0·120 | 30·6 |
| Lactation | | | | | | | |
| Day 14 | 4 | 12·2 | 5·0 to 31·0 | 4843 | 0·054 | 0·035 to 0·077 | 21·4 |
| Hysterectomy† | | | | | | | |
| Day 40 | 4 | 31·6 | 11·0 to 90·0 | 7394 | 0·091 | 0·078 to 0·129 | 21·3 |
| Day 80 | 4 | 19·7 | 10·5 to 36·3 | 4452 | 0·094 | 0·077 to 0·151 | 21·2 |
| Day 110 | 4 | 35·0 | 17·5 to 70·0 | 8645 | 0·077 | 0·063 to 0·095 | 21·8 |
| Hysterectomy† and oestradiol benzoate‡ | | | | | | | |
| Day 40 | 4 | 31·6 | 16·3 to 60·8 | 9733 | 0·070 | 0·047 to 0·106 | 21·6 |
| Day 80 | 5 | 37·2 | 15·8 to 88·5 | 8746 | 0·111 | 0·077 to 0·189 | 29·1 |
| Day 110 | 4 | 35·0 | 17·0 to 71·3 | 11760 | 0·086 | 0·063 to 0·118 | 28·9 |

* Expressed as μg-equivalents of NIH-GH-B7/mg fresh weight of the anterior pituitary gland.
† Animals were hysterectomized on Days 8 to 10 of the oestrous cycle.
‡ Oestradiol benzoate (5 mg/day in corn oil) was injected intramuscularly daily for 12 days before termination of the experimental period.
§ Expressed as i.u.-equivalents of NIH-P-B1/mg fresh weight of the anterior pituitary gland.

## RESULTS

### *Morphological changes in pituitary and ovarian function*

Morphological data on ovarian function at different reproductive stages are presented in Table 1. The injection of oestrogen in hysterectomized pigs caused an increase ($P<0·01$) in the weight of the whole pituitary gland. Within 14 days after parturition, there was an increase ($P<0·01$) in whole gland weight when compared with any stages of pregnancy. Within the three stages (Days 40, 80 and 110) during pregnancy, after hysterectomy and after hysterectomy plus oestrogen treatment, there was a linear increase ($P<0·01$) in the weight of the pituitary gland. The same significant increases in weight of the adenohypophysis were found for these reproductive stages.

The number of CL was similar ($P>0·05$) within and between reproductive stages, with the exception of the absence of CL in lactating animals. The weight of CL was similar ($P>0·05$) between experimental groups, but there was a linear increase ($P<0·01$) in luteal tissue weight from Days 40 to 110 within the three experimental groups.

The total ovarian weight was similar ($P>0·05$) in pregnant and hysterectomized pigs, but a marked decline ($P<0·001$) occurred after parturition. After hysterectomy, ovarian weight declined ($P<0·01$) when hysterectomized pigs were given injections of oestrogen.

After parturition, the numbers of ovarian follicles of <4 mm in diameter declined ($P<0{\cdot}01$). There was a greater number ($P<0{\cdot}01$) of these small follicles in the pregnant compared to the hysterectomized animals. The injection of oestrogen did not reduce significantly the number of small follicles in hysterectomized pigs. Within reproductive stages, there was no significant linear increase in the number of these follicles. Because of considerable variation in the numbers of larger follicles (4 to 6 mm) in pregnant and hysterectomized pigs, there were no significant differences between these groups. Soon after parturition, the larger follicles were absent. Furthermore, exogenous oestrogen in hysterectomized pigs reduced ($P<0{\cdot}01$) the number of these follicles. The numbers of live and dead fetuses are shown in Table 1.

*Adenohypophysial cell types*

In our investigation, the acidophils were characterized by a positive staining reaction to orange G and were further differentiated by yellow and orange granules in the cytoplasm. These acidophils represented STH (STH cell) and prolactin (prolactin cell) activities in the porcine adenohypophysis. The chromophobes lacked staining reaction in their cytoplasm and may represent undifferentiated, inactive or depleted adenohypophysial cells.

The number of cells representing each acidophilic cell type was expressed in percentages of the total cell count for each pituitary gland. The overall mean for each group was expressed as the mean of all animals in each reproductive stage. A typical range of the total cell count for each gland was 2500 to 4500. Analysis of variance indicated a difference ($P<0{\cdot}01$) between the reproductive stages in the numbers of the yellow (STH cell) and orange (prolactin cell) acidophils, and the chromophobes (Table 3). With the mean values for the different reproductive stages (Table 3), the Scheffe test for multiple populations was used to determine differences between reproductive stages for individual cell types.

*Acidophils*

When considering all reproductive stages, the acidophils (STH cell and prolactin cell) represented half (53·5%) the cell population in the adenohypophysis. Notable exceptions were the low percentages of these cells in fetuses and during lactation (Table 3). In fetuses, the percentages of STH cells were low ($P<0{\cdot}05$) when compared with both groups of prepubertal pigs (Groups 2 and 3; Table 3). Percentages of orange acidophils (prolactin cell) were similar ($P>0{\cdot}05$) in these three groups of immature pigs. The proportion of STH cells was higher ($P<0{\cdot}05$) in 17-day-old pigs than in any reproductive stages in mature animals. This is a period of rapid growth in the young pig.

During the different days of the oestrous cycle, the percentages of yellow acidophils (STH cell) remained constant ($P>0{\cdot}05$). Furthermore, the proportions of these STH cells were similar ($P>0{\cdot}05$) to those found in pregnant (Groups 9 to 12), lactating (Group 13) and hysterectomized (Groups 14 to 17) animals.

The percentages of orange acidophils (prolactin cell) were higher ($P<0{\cdot}05$) during stages of the cycle (Groups 4 to 8), during pregnancy (Groups 9 to 12) and after hysterectomy (Groups 14 to 17) than those found in immature

(Groups 1 to 3) or lactating (Group 13) pigs. The marked drop in the proportion of prolactin cells in the lactating pigs may indicate actively secreting cells and, thus, little accumulation of cytoplasmic granules that retain prolactin. The high percentages of prolactin cells during late pregnancy (Groups 11 and 12) and the late stage in hysterectomized animals (Group 17) suggested an accumulation of cytoplasmic granules that retain prolactin activity. Within the five stages of the oestrous cycle, the prolactin cell population remained relatively constant ($P>0{\cdot}05$).

TABLE 3

ADENOHYPOPHYSIAL CELL TYPES AT DIFFERENT REPRODUCTIVE STAGES IN THE PIG

| *Reproductive stage* | *No. of pigs* | *Mean percentage of cell type** | | |
|---|---|---|---|---|
| | | *Acidophils* | | *Chromophobes clear* |
| | | *STH cells* | *Prolactin cells* | |
| Immature | | | | |
| 1 Fetal | 7 | 8·9 | 7·1 | 14·0 |
| 2 Prepuberal (17 days old) | 2 | 43·7 | 2·6 | 22·3 |
| 3 Prepuberal (105 to 118 days old) | 3 | 25·6 | 26·0 | 9·5 |
| Oestrous cycle | | | | |
| 4 Day 1 | 4 | 14·6 | 34·1 | 13·0 |
| 5 Day 4 | 4 | 20·4 | 39·9 | 5·8 |
| 6 Day 10 | 4 | 12·8 | 46·0 | 16·5 |
| 7 Day 16 | 5 | 20·2 | 40·5 | 14·1 |
| 8 Day 18 | 4 | 19·2 | 42·7 | 5·4 |
| Pregnancy | | | | |
| 9 Days 12 to 40 | 8 | 11·8 | 42·7 | 11·4 |
| 10 Days 41 to 80 | 4 | 13·3 | 46·8 | 8·4 |
| 11 Days 80 to 110 | 8 | 8·8 | 55·0 | 8·9 |
| 12 After unilateral ovariectomy | 3 | 7·2 | 66·1 | 14·0 |
| Lactation | | | | |
| 13 Day 14 | 4 | 10·8 | 15·8 | 36·4 |
| After hysterectomy | | | | |
| 14 Days 24 to 26 | 4 | 14·0 | 36·9 | 15·5 |
| 15 Days 41 to 47 | 2 | 15·4 | 38·6 | 9·3 |
| 16 Days 80 to 92 | 4 | 12·7 | 42·4 | 10·6 |
| 17 Days 120 to 150 | 3 | 8·1 | 60·1 | 8·3 |
| Analysis of variance F value/cell type† | | 14·98 | 30·83 | 14·58 |

* Criteria for distinguishing cell types: Acidophils: Somatotroph, orange G positive, yellow; Prolactin cell, orange G positive, orange. Chromophobes: (undifferentiated, inactive or depleted cells).

† Statistical significance: F 0·05, 2·14; F 0·01, 2·91.

*Chromophobes*

The chromophobes represented 13·1 % of the cell population. Their distribution changes ($P<0{\cdot}01$) during the reproductive stages (Table 3). In immature pigs, during the oestrous cycle or pregnancy and after hysterectomy, only minor changes ($P>0{\cdot}05$) were evident in the percentages of chromophobes. A marked increase ($P<0{\cdot}01$) was found, however, in the percentage of chromophobes in lactating animals (Table 3). Chromophobes were the predominant cell type

during lactation (Group 13), but these seemingly inactive cells may have been active acidophils that were synthesizing and releasing prolactin. By rapid turnover of hormone, the cytoplasmic granules in prolactin cells would lack differentiation and appear as inactive cells. A considerable proportion of the cell population in 17-day-old pigs (Group 2) consisted of chromophobes, though this percentage was not higher ($P>0{\cdot}05$) than that found in other reproductive stages (Table 3). A rather large proportion of undifferentiated cells may be expected in the young pig.

*Somatotrophic hormone activity*

The concentrations and total contents of STH activity remained similar ($P>0{\cdot}05$) during the three stages of pregnancy and during lactation (Table 2). Levels of STH activity tended to decline during late pregnancy (Day 110) and in lactating animals. The total content of STH did not reflect this trend since pituitary gland weight increased during pregnancy and after parturition (Table 1).

After hysterectomy, the concentrations of STH activity tended to increase, but these shifts were not different ($P>0{\cdot}05$) from those in pregnant or lactating pigs (Table 2). The total content of STH was greater ($P<0{\cdot}05$) in hysterectomized than in pregnant pigs by Day 110. Again, this marked increase in STH during later reproductive stages was caused primarily by increased weight of the pituitary gland.

When oestradiol benzoate was given to hysterectomized pigs, there was a consistently high level of STH activity, though it was not greater ($P>0{\cdot}05$) than the level in uninjected hysterectomized animals. The pituitary gland content of STH in the oestrogen-treated pigs was greater ($P<0{\cdot}05$) than that found during late pregnancy or after parturition.

*Prolactin activity*

During gestation, prolactin concentrations and contents remained similar ($P>0{\cdot}05$; Table 2). By Day 14 of lactation, there was a significant decline ($P<0{\cdot}05$) in the level but not in the content of prolactin.

In hysterectomized pigs, the levels of prolactin remained constant ($P>0{\cdot}05$) and were similar to those found during the three stages of pregnancy. Exogenous oestradiol benzoate caused a consistently higher trend in prolactin activity in hysterectomized animals. The total adenohypophysial content of prolactin was higher ($P<0{\cdot}05$) at Days 80 and 110 after injection of oestrogen in hysterectomized pigs than that found in uninjected hysterectomized animals. This increase in the total content of prolactin activity was related to the marked increase in weight of the pituitary gland after the injection of oestrogen.

## DISCUSSION

Somatotrophin and prolactin-producing cells represent the classical acidophils of the adenohypophysis. These two hormones have been identified by immunofluorescent staining procedures as two distinct cell types (Emmart, Spicer & Bates, 1963; Baker, Midgley, Gerster & Yu, 1969). In the porcine pituitary

gland, cells producing STH were characterized as yellow acidophils and those producing prolactin as orange stained cells.

In immature animals (Groups 1 to 3), acidophils, particularly STH cells, increased markedly during a period of rapid growth. In mature animals (Groups 4 to 12), the predominant cell type was the orange acidophil (prolactin cell) with percentages ranging from 34 to 66%. The prolactin cells were moderately granulated throughout the cycle, whereas during the first 80 days of pregnancy and after hysterectomy, these cells were heavily granulated. In late pregnancy, these cells were partly degranulated with indistinct outlines. After parturition, the percentage of orange acidophils declined to 16% with a corresponding increase in the chromophobes.

The chromophobes in these lactating pigs were mostly large with abundant cytoplasm and large nuclei. It is probable that the chromophobes were originally degranulated prolactin cells. Similar shifts in cell type have also been observed during reproductive stages in the guinea-pig (Kirkman, 1937), rabbit (Allanson, Cameron & Foster, 1966) and human (Swanson & Ezrin, 1960). Farquhar & Wellings (1957) suggested that chromophobes may secrete corticotrophin and Herlant (1964) proposed a third type of acidophil with small granules as a secreting cell type for corticotrophin.

The finding in the young pig (17 days old; Table 3) of the highest percentage of STH cells correlated with the extraordinarily high basal plasma growth hormone levels (50 to 200 ng/ml) in newborn and 21-day-old pigs (Kipnis *et al.*, 1969).

Between Days 1 and 18 of the oestrous cycle, no significant changes in the percentages of STH cells were evident (Table 3). In the adult pig, basal levels of plasma growth hormone were consistently low (5 to 7 ng/ml) and increased only by severe insults such as fasting, stress and exercise (Machlin, Horino, Hertelendy & Kipnis, 1968). Kipnis *et al.* (1969) concluded that growth hormone does not play an essential or vital rôle in maintaining metabolic homeostasis in the adult.

There were no significant correlations in the percentages of STH cells in the porcine adenohypophysis and the concentration or content of STH during different stages of pregnancy and lactation (Tables 2 and 3). Earlier work indicated that concentrations of STH activity were similar in cycling, ovariectomized and hysterectomized pigs (Anderson & Melampy, 1966).

In hysterectomized pigs, the percentages of STH cells and the concentrations of STH tended to be higher than those found during similar stages during pregnancy. Exogenous oestrogen caused a marked increase in the pituitary content of STH activity by Day 110 in hysterectomized pigs as compared with uninjected hysterectomized animals (Table 2). Diethylstilboestrol increased the weight of the adenohypophysis, the concentration of STH activity in the pituitary gland (Struempler & Burroughs, 1959) and the plasma level of growth hormone in cattle (Trenkle, 1970).

During pregnancy and after hysterectomy, the percentages of prolactin cells and the concentrations of prolactin in the adenohypophysis indicated no significant changes. There was a trend to increased percentages of these orange acidophils in the later stages of pregnancy and after hysterectomy, but this

trend was not evident in the levels of prolactin. Exogenous prolactin was essential for maintaining CL after hypophysectomizing the pig at Day 70 of pregnancy (du Buisson & Denamur, 1969), but ineffective in maintaining luteal function during the oestrous cycle (Sammelwitz & Nalbandov, 1958). The tendency toward increased percentages of prolactin cells in animals with older CL agrees with the concept that the need for prolactin for luteal maintenance in the pig is a function of the age of the CL (du Buisson & Denamur, 1969).

Exogenous oestrogen caused a marked increase in prolactin content in the adenohypophysis of hysterectomized pigs by Days 80 and 110. These results suggest a rôle of prolactin in maintaining luteal function for prolonged periods in the pig. Earlier work by Gardner, First & Casida (1963) demonstrated the luteotrophic action of exogenous oestrogen in pigs.

In the lactating pig, the marked decline in the percentage of prolactin cells correlated with the low concentration and content of prolactin in the adenohypophysis (Tables 2 and 3). Since milk production is maximal soon after parturition, these parameters indicate a hyperactive synthesis and secretion of prolactin, with almost complete absence of storage of this hormone in the cytoplasm of the adenohypophysis.

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