# A STUDY OF THE BOVINE VAGINA AS RELATED TO REPRODUCTIVE PERFORMANCE

by

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#### A THESIS

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#### INTRODUCTION

In the last few years disorders of the reproductive tract have received much attention from both research workers and livestock breeders throughout the world. Major concern resulted from the severe economic losses that were due to problems associated with reproductive failure. The origin of the problems that lead to infertility and sterility has been considered to be both physiological and anatomical.

In order to evaluate some of the possible causes of reproductive failures, physiological as well as histological investigations have been undertaken to establish the normal variations in reproductive tissue. Considerable work has been done in this field.

A thorough study of the bovine vagina was found to be missing in the available literature. Nevertheless some who have studied other reproductive organs in the cow discussed the function of the vagina and related changes occurring in the vaginal tissue as indicative of definite periods in the estrual cycle (Woodman and Hammond 1925, Murphy 1926, Cole 1930, Hummon 1932, Brown 1944, Roark and Herman 1950). These studies however were not comprehensive in the pertinent data concerning the importance of the vagina in reproductive performance.

The present study has been undertaken to determine what changes occur in the bovine vagina during the various stages of the reproductive cycle.

Special attention was given to determine if cellular changes in the epithelium were associated with reproductive performance.

#### REVIEW OF THE LITERATURE

The vagina of the bovine is a tubular structure 8-10 inches long and 3-4 inches wide (Trotter and Lumb, 1958) extending through the pelvic canal from the vestibule to the cervix.

Nibler (1957) recorded the total length of the vagina and vestibule as 12 to 16 inches and determined that the length of the vagina varied with the size and age of the animal. Perkins et al. (1954) however, reported the average length of the vagina of 95 parous cows to be 6.9 + .72 inches.

The wall of the vagina as described by Trautmann (1957) consists of several fairly distinct layers; (1) the mucosa-submucosa lining the cavity is comprised of a high columnar secretory epithelium in the cranial part, varying from low cuboidal to stratified columnar during metestrus. There is a lamina propria forming little or no papillary body. Its connective tissue is rich in cells near the epithelium and contains lymph nodules. The submucosa is loose. (2) the tunica muscularis usually consists of a thick inner circular layer and a thin outer longitudinal layer. The latter is continued for a short distance in the wall of the uterus. (3) adventitia or cranially a serosa, with muscularis serosae, contains large vessels, nerves and ganglia.

#### Follicular Phase

During proestrum the epithelium in the anterior end is composed of several layers of large, wide mucous secreting cells with a superficial layer of tall, narrow, mucous secreting columnar cells. The epithelium is reduced to one or two layers during estrum. (Cole 1930) (Hummon 1932) (Hansel 1947). Cole (1930) indicated that there were two or three layers of polyhedral shaped epithelial cells beneath the superficial layer. According to Brown (1944) and Roark and Herman (1950), the high columnar mucoid epithelium of the superficial layer rests upon a variable number of stratified squamous cells. Roark and Herman (1950) stated that the number of layers of stratified squamous cells was minimal during estrus. However the total epithelium height was at maximum due to an accumulation of mucous in the mucoid epithelium. Brown (1944) reported that during estrus the thickening resulted from the greatly increased height of the columnar type cells of the superficial layer rather than from an increase in the number of layers of the squamous epithelium.

Cornification of the epithelial cells does not occur in the cow and this is associated with a low estrogen output (Trautmann 1957). Selye's (1948) indicated that stratification and cornification of the vaginal epithelium took place during the estrous period in the female rat. Sanger et al. (1958) reported the incidence in ewes. He stated that an increase in estrogens in the body, stimulated an increase in the number of cell layers lining the vagina, mobilized glycogen in the cells and caused

cornification of the superficial layers. Cornification of vaginal cells in the cow was reported by Murphy (1926) who noticed a distinct surface cornification beneath which were clear swollen cells above the compact multiplying germinal layer. He also reported complete desquamation in 24 to 36 hours following the onset of estrus. Burrows (1949) reported that during estrus the more superficial layer of cells became squamous and cornified.

During proestrum and active heat and the small blood vessels beneath the epithelium were congested (Cole 1930, Murphy 1926, Woodman and Hammond 1925, Hummon 1932, Roark and Herman 1950). On the 20th. day Murphy (1926) noted that lymph flowed from the mucosa through spaces in the epithelium. Woodman and Hammond (1925) reported similar observations and indicated that it was also coupled with infiltration of leucocytes at the time of heat. Infiltration of leucocytes and lymphocytes had been reported by Roark and Herman (1950) to occur in the superficial stroma during proestrous, in the deeper portions of the epithelium during estrus and in the superficial epithelium at one day postestrus. A report by Hummon (1932) stated that leucocytes were rare at all times in the anterior portion. Brown (1944) described that they are seldom seen during estrus.

According to Cole (1930) the epithelium starts as one layer in the anterior portion of the vagina and the layers increase in number posteriorly. The epithelium in the posterior portion increases in height during the follicular phase (Hummon 1932). Lymphocytes and leucocytes were prevalent in the posterior end at this time (Hummon 1932, Cole 1930, Murphy 1926).

#### Luteal Phase

Two days postestrum the epithelium in the anterior portion consists of several layers and the superficial layer is variable in form and only slightly differentiated from layers beneath it (Cole 1930). From the second to the fourth day, Hummon (1932) reported that a great number of the cells rupture and often the entire portion of the cell disappears together with the mucous content of the cells. Often the nuclei are not present. From the fifth to the eleventh day a gradual increase in size of these cells is noted. According to Cole (1930), the epithelium appeared vacuolar and partially degenerate in character from the eighth to the eleventh day. In general the epithelium reverts to a low stratified cuboidal or columnar epithelium in metestrus (Trautmann 1957).

Following ovulation, a gradual decrease in vascular congestion has been noted and the least congestion is observed during 7 to 10 days postestrum (Roark 1950, Hummon 1932). Leucocyteic migration and infiltration have been reported by Murphy (1926) following ovulation and 3 days after.

In the posterior end, mucous secreting cells appear for the first time 2 days postestrum whereas, they are present during proestrum in the anterior portion (Cole 1930). A drop in thickness of the epithelium occurs in the third and fourth days postestrum due to a great reduction in number and size of the surface squamous cells in the posterior portion. No change is noticed from the eleventh day to the fourteenth day at which time a

gradual increase begins and continues until 12 hours following the beginning of heat. Congestion of blood vessels in the subepithelium is noted in postestrum and continues until the fifth day postestrum (Hummon 1932). Intense infiltration of leucocytes occurs two to five days after estrus according to Brown (1944), eight to eleven days postestrum according to Cole (1930) and on third and fourth days metestrum according to Hummon (1932) who also stated, that the least infiltration takes place in epithelium and subepithelium, during the eleventh day.

#### Diestrum

During diestrum, the mucoid epithelium changes from columnar to cubical in type in the anterior portion (Roark and Herman 1950). Congestion of the vessels is noted in the posterior portion during the eighteenth day of the estrous cycle. (Hummon 1932).

### Pregnancy

The changes occurring during pregnancy in the vaginal wall have been reported by Hummon (1932) who stated that the thickness of the epithelium increases from 21st. day to the 152nd. day of gestation. From that time until 263 days a slight decrease in thickness of epithelium is observed. The cells contain large, granular, lightly stained nuclei. By the 253rd. day mucous and desquamated cells are seen on the surface of the epithelium. During late pregnancy the cells in the anterior portion of the

vagina are columnar. The mid and posterior portions of the vagina contain a stratified squamous epithelium. The changes in the anterior portion during pregnancy are less marked than in other regions of the vagina (Cole 1930). In most cases the epithelium is low and usually flattened. Lymphocytes are present but leucocytes are few or absent. Burrows (1949) stated that in pregnancy and pseudopregnancy the superficial layer of cells become rounded, swollen and ultimately columnar in shape and are loaded with mucous-like material.

#### Post Partum

Roark and Herman (1950) reported vascular congestion in the bovine vagina following parturition which decreased within 20 to 30 days after calving.

#### MATERIALS AND METHODS

Vaginal tissues were obtained from the dairy cows, with known reproductive histories, eliminated from the Kansas State University dairy herd, supplemented by tissues from cows of unknown histories slaughtered at the Armour Packing Company plant in Kansas City, Missouri. The cows from the Kansas State herd represented all stages of reproductive cycles from normal pregnancies to known pathological conditions. The tissues obtained from the packing plant were selected from clinically normal animals also represented various stages of the reproductive cycle. The stage of gestation of the pregnant cows was determined by fetus

size; and the stage of the estrous cycle of the non-pregnant cows was established by ovarian structures.

Vaginal tissue was removed from anterior, middle and posterior portions of the vagina, as soon as possible after slaughter. In certain instances it was not possible to secure tissues from all three sites mentioned, because of damage to the reproductive tract occurring when the tract was excised. The material collected was immediately cut into 10 by 10 millimeter sections, to ensure efficient fixation; then placed in a 10 percent buffered formalin. They were left in formalin for 24 to 36 hours and dehydrated in series of isopropyl alcohol.

The tissues were transferred to a 5 percent xylol solution in alcohol and placed in an oven maintained at 50°C for 45 to 60 minutes to ensure good infiltration of the paraffin. After the tissues had attained oven temperature, paraffin was added to the solution.

Vaporization of the alcohol and xylol, required 16 to 18 hours in the 50°C oven. After this preliminary infiltration, the tissues were transferred to a 57°C oven, and infiltration was completed with three changes of 54° - 56°C paraffin over a 36 hour period. The tissues were then embedded in fresh paraffin. Tissues that hardened excessively during infiltration were soaked in 50 percent alcohol over-night prior to sectioning. The tissues were sectioned at 8 microns and were mounted with Mayer's albumen

<sup>&</sup>lt;sup>1</sup>Mayer, "Journal of the Royal Microscopical Society-London Vol. 4:317 (1884).

on glass slides. They were stained with a modified iron haematoxylin solution. This required the sections to be placed in a 1 percent iron alum solution for 15 to 30 minutes, then in a .06 percent haematoxylin for 5 minutes and finally in a solution of .025 percent acid fuchsin and .1 percent orange G for five minutes. Some of the sections were processed through the Periodic Acid Schiff's (PAS) reaction to ascertain if the epithelial cells contained glycogen or glycoproteins.

The prepared sections were examined microscopically and the following observations recorded: The type of epithelium present, the morphological appearance of the cells which make up the epithelium, the extent of vascularization of the vaginal wall, the presence of cornification and finally the extent of leucocytic infiltration present. These observations were made on tissues from the anterior portion, the middle portion and the posterior region of the vagina which represented various stages of the reproductive cycle. Appropriate microphotographs were taken for each phase of the reproductive cycle.

#### RESULTS AND DISCUSSION

During the follicular phase of the estrous cycle, the vaginal epithelium in the anterior portion was comprised of a basal layer of polyhedral cells one or two cells thick and a superficial layer of tall, wide, secretory cells (Plate I, Fig. 1). The secretory function of the high columnar cells was demonstrated by using the Periodic Acid Schiff's (PAS) reaction (Plate III, Fig. 3).

The glycogen content of the mucoid secretion of the cells responded to the treatment with Periodic Acid Schiff's reagent rendering the superficial layer bright red. The nuclei of the cells of this layer were compressed, oval in shape with tapering They were darkly stained. The cytoplasm was finely shredded (granulated). The basal layer varied from one to three cells thick. The initiation of secretory function in the superficial columnar cells and likewise their increase in height may be attributed to the stimulation by estrogen which is secreted by the developing Graafian follicle. Even though an increase in the height of the superficial layer of the epithelium did occur during the follicular phase, a decrease in the number of polyhedral cells was observed during this high estrogen production period. Considerable variation had been observed in the height of the vaginal epithelium of cows representing the same stage of the estrous The average height of the epithelium in the anterior portion during the follicular stage was 22.7 microns with a range between 19.8 to 25.6 microns. Lymphocytes and leucocytes were rare yet the subepithelium was rich in blood vessels.

The change from the anterior portion to the mid portion was characterized by an increase in the number of polyhedral cells below the secretory columnar layer during the follicular phase of the cycle. The average height of the epithelium in this region was 33.9 microns with an average range of 31.4 to 36.4 microns. No abnormal number of leucocytes was observed.

In the posterior portion of the vagina the epithelium was

also stratified with a superficial layer of secretory columnar cells, a basal layer and an intermediate zone of polyhedral-shaped cells which varied considerably in thickness. The nuclei of the polyhedral-shaped cells were large, oval, granulated and lightly stained. There were two, sometimes three or more, darkly stained nucleoli in each nucleus. The superficial layer of epithelium in this region was not as high as in the anterior end and a number of the cells were dark staining and non-secretory (Plate I, Fig. 2). The average height of the epithelium was 36.3 microns with an average range of 33.6 to 39 microns. The increased height was due to the highly stratified polyhedral cells. Lymphocytes, leucocytes and occasional erythrocytes were evident in the epithelial and subepithelial tissue.

The observations reported by Cole (1930) and Hansel (1947) who stated that the epithelium in the anterior end of the vagina consisted of one cell layer during estrum, do not agree with the observations of this study, as it was observed that the epithelium in the anterior region was comprised of at least two layers during the high follicular phase. However, this work is in agreement with these workers as well as Roark and Herman (1950) who reported the existence of a stratified epithelium in the vagina proper during this phase of the cycle. Hummon (1932) reported that the epithelium in the anterior portion of the vagina during the follicular phase of the cycle was pseudostratified which is in contrast with the observations made in this study. Roark and Herman (1950) observed the low basal layers of cells and described the epithelium

during proestrous as "not true stratified squamous in character." Several investigators (Murphy 1926, Cole 1930, Hummon 1932, Brown 1944, Hansel 1947, Roark and Herman 1950, Trautmann 1957 and Sanger et. al. 1958) reported that the epithelium became high in the anterior portion during this period. However this statement can only be applied to the superficial columnar layer. The observations of this study support those reported by Cole (1930), Brown (19/1) and Roark and Herman (1950) that no increase occurred in the epithelial layers during the follicular stage. However in this study it was observed that the epithelial height is reduced due to a decreased number of underlying polyhedral cells. Trautmann (1957) and Sanger et. al. (1958) disagree with these observations when they reported an increase in the epithelial layers. Vascular congestion in the anterior portion during the follicular phase did occur as was previously described by Woodman and Hammond (1925), Cole (1930) and Roark and Herman (1950). Infiltration of leucocytes and lymphocytes was reported by Woodman and Hammond (1925), Hummon (1932) indicated that these cells were rare at all times in the anterior portion. This was also observed in all clinically normal cows which were included in this study.

Shortly after ovulation a decrease in the height of the superficial cell layer of the epithelium of the anterior end of the vagina was observed. The cells decrease in width and secretory activity (Plate I, Fig. 3). This may be due to a drop in estrogen secretion by the ovaries. An increase in the stratification of polyhedral cells was noted. Considerable variation in the height

of the epithelium and the secretory activity of the superficial layer was observed. The average height was 28.6 microns, ranging from 25.9 to 31.3 microns. There was no congestion of blood vessels of the underlying tissue during this phase nor a heavy leucocytic invasion.

The epithelium in the mid portion two days after ovulation phase was comprised of a low cubical compared to a low columnar superficial layer in the anterior region. Several layers made up the epithelium with an average height of 49.6 microns. The average range for this was from 44.9 to 54.8 microns. The zone of polyhedral-shaped cells described in the posterior end of the vagina during the follicular phase was evident in this area. A few more leucocytes were observed in this area compared to the anterior portion.

The posterior region of the bovine vagina during this period contained of a low stratified epithelium with a cubical to a squamous superficial layer. Slight cornification was observed. The average height of the epithelium was 37.6 microns with an average range of 35.2 to 40 microns.

Murphy (1926), Hummon (1932), Brown (1944) and Trautmann (1957) had reported a low columnar epithelium in the anterior portion of the vagina during the luteal phase of the estrous cycle. However this statement should only apply to the superficial layer, which was observed in this study to be low columnar or cubical (Plate I, Fig. 4). Cole (1930) stated that vacuoles were present in the epithelium of the anterior portion during this

period. However it is doubtful if this would represent the average condition. The observations on some of the material presented here comply with this statement. Roark and Herman (1950) agreed with the results of this study in respect to the non-occurrence of vascular congestion in the underlying tissue. The leucocytic infiltration observed by Hummon (1932) and Brown (1944) as occurring two to five days postestrum was only observed in one case among the material studied here, and the high number of leucocytes was probably not due to the stage of the estrous cycle.

During the height of the luteal phase of the estrous cycle the epithelium through the vagina appeared considerably less active. In the anterior region the cells of the superficial layer were reduced in height and some of the cells appeared to have ruptured (Plate I, Fig. 5). The height of the superficial layer was observed to be progressively lower posteriorly and there was a highly stratified layer of polyhedral cells throughout the vaginal epithelium (Plate I, Fig. 6).

A high number of leucocytes was also evident in the posterior region of the vagina. The average height of the epithelium during this period in the anterior portion was 42.5 microns with a range from 38.4 to 46.6 microns. In the posterior region the epithelium ranged from 37.2 to 50.2 microns in height with an average of 43.7 microns.

It was observed that during early pregnancy the superficial layer of the epithelium of the anterior portion was low cubical or squamous and non-secretory. There were two or three layers of

polyhedral cells under the superficial layer (Plate II, Fig. 1). Few leucocytes were observed in the epithelium during early pregnancy.

The mid portion was similar in structure to the anterior portion. The posterior region showed more desquamation of the superficial layer which was definitely squamous. (Plate II, Fig. 2).

The superficial layer of the epithelium in the anterior region increased in height as pregnancy progressed and was low columnar by mid pregnancy (Plate II, Fig. 3). In the posterior region it had also increased but was not as high as in the anterior region (Plate II, Fig. 4). Occasional leucocytes were observed throughout the vagina.

The increase in height of the superficial layer of the epithelium progressed so that it reached its highest point at 5 months and remained high until parturition. It was high columnar and secretory in the anterior portion during late pregnancy (Plate II, Fig. 5). This increase in height and function of the superficial layer during late pregnancy may be due to the increased amount of estrogen being secreted by the placenta. The average height of the epithelium in the anterior region was 28.6 microns with an average range of 26 to 31.2 microns. The basal layer was comprised of flat cells. Few leucocytes were observed in the anterior portion. The secretory function was also evident in mid region and posterior region. Leucocytes were more abundant, however it was evident that part of the superficial layer in the posterior region was non-secretory. The average height of the

epithelium in the mid portion was 27.7 microns with an average range of 25 to 30.4 microns. In the posterior portion the average height was 31.5 microns with an average range of 27.7 to 35.2 microns.

The increase in epithelial height that was reported by Hummon (1932) during early pregnancy was not observed in this study. Furthermore, there was no indication in our observations of a squamous epithelium in the mid and posterior portions which Hummon (1932) reported. However the results of this study are in agreement with Burrow's (1949) statement that described the cells of the superficial layer of the epithelium as swollen, secretory and columnar in shape. Our results indicate definite changes occurring in the epithelium of the anterior portion of the vagina during pregnancy which is in disagreement with the work of Cole (1930) who reported no specific changes during pregnancy compared to other regions of the bovine vagina.

Shortly after parturition the vaginal epithelium appeared to lose its secretory function. The superficial layer in the anterior portion at 10 days post-partum was low cuboidal and resting on 2 to 3 layers of polyhedral cells (Plate III, Fig. 5). The epithelium and subepithelium were heavily infiltered with leucocytes. This condition was probably associated with post-partum regression of the uterus.

During the interval from parturition to first estrus the vaginal epithelium appeared now functional throughout (Plate III, Fig. 1 and 2). In the posterior portion the superficial layer

of cells was found to be cornified and flaking off from the surface (Plate III, Fig. 2). This may be due to a prolonged low level of estrogens. The influence of low estrogenic levels was also observed in vaginal tissues from cows which failed to exhibit normal estrous cycles. These tissues which were often associated with embryonic mortality contained extremely high numbers of leucocytes throughout the vaginal wall (Plate III, Fig. 4). The presence of white blood cells appeared to be associated much more closely with pathological conditions than with the stage of the estrous cycle.

#### SUMMARY AND CONCLUSIONS

A thorough knowledge of the histological changes which occur in the bovine vagina is essential if the importance of this organ to successful reproductive performance is to be determined. This study was undertaken to determine the cellular changes that occur in the bovine vagina during the various phases of the estrous cycle and pregnancy and whether these changes have any significance in reproductive performance.

It was noted that there was some variation in epithelial changes occurring during the reproductive cycle between the anterior and posterior regions of the vagina.

During both the follicular phase of the estrous cycle and the stage of gestation when the placenta is secreting a relatively high level of estrogen, the superficial layer of the vaginal epithelium consisted of highly secretory columnar cells and a basal layer of polyhedral cells which varied considerably in thickness. Generally the secretory activity of the superficial layer decreased and the thickness of the polyhedral layer of the cells increased posteriorly. During the luteal phase of estrus cycle and early in pregnancy, the superficial layer of the vaginal epithelium varied from a non-secretory, low cuboidal layer in the anterior end of the vagina to a flat squamous layer of cells in the posterior end. The layer of underlying polyhedral cells was considerably thicker in all parts of the vagina, however it was at its highest level in the posterior end. During the extended period of low estrogen activity following parturition the vaginal

epithelium was more highly stratified and the superficial layer appeared cornified and was flaking off into the vaginal lumen.

The presence of abnormal numbers of leucocytes appeared to be more closely associated with pathological conditions than with any phase of the normal reproductive cycle.

The results of this study would indicate that the presence of abnormally high numbers of leucocytes in the vaginal epithelium might be indicative of bacterial contamination which could cause reproductive failures.

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APPENDIX

# Explanation of Plate Abbreviations

ep. -- Epithelium

ves. -- Blood Vessel

leu. -- Leucocytes

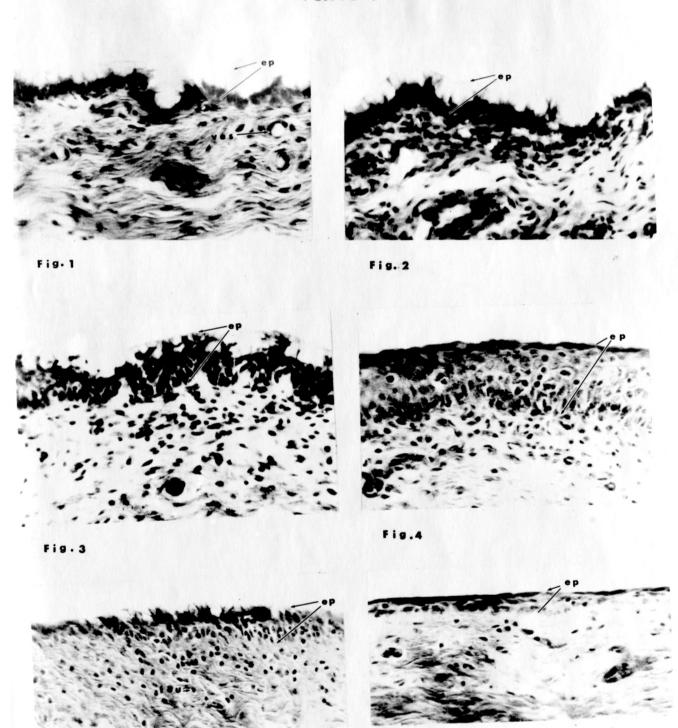
#### EXPLANATION OF PLATE I

- Fig. 1. A section from the anterior portion of the vaginal epithelium during the follicular phase of the estrous cycle showing the high columnar secretory superficial layer and one or two basal layers. Vascularity of the subepithelium is evident too. Cow. No. 382 (X125)
- Fig. 2. A section from the posterior end of the vaginal epithelium during the follicular phase showing the columnar secretory superficial layer also present in that region, and an increased number of basal cells underneath can be noted. Cow No. 382 (X125).
- Fig. 3. A section from the anterior end of the vaginal epithelium at two days post-ovulation, showing a partially secretory columnar superficial layer with a highly stratified layer of flat and polyhedral cells underneath.

  Cow No. 383 (X125).
- Fig. 4. A section from the posterior end of the vagina of the same cow as Fig. 3. The epithelium consists of a non-secretory low cuboidal superficial layer and several layers of polyhedral cells underneath. Leucocytes are evident in epithelium and subepithelium (X125).
- Fig. 5. A section from anterior portion of vaginal epithelium at 14 days past ovulation showing a low cuboidal superficial layer with some ruptured cells and a layer of polyhedral cells of variable thickness. Leucocytes are prevalent. Cow No. 376 (X125).

Fig. 6. A section from posterior portion of the vaginal epithelium of same cow as Fig. 5 showing a squamous superficial layer with polyhedral shaped cells underneath (X125).

# PLATE I



... 5

Fig. 6

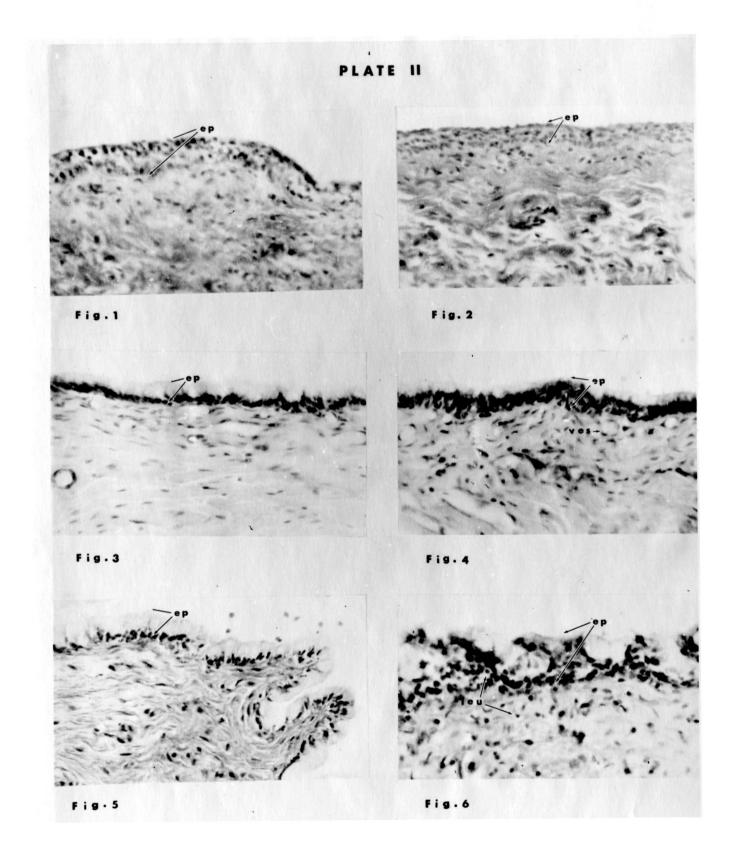
#### EXPLANATION OF PLATE II

- Fig. 1. A section from the anterior end of the vaginal epithelium during early pregnancy (74 days) showing a low cuboidal non-secretory superficial layer and several layers of polyhedral cells underneath. Cow No. 377 (X125).
- Fig. 2. A section from the posterior portion of the vaginal epithelium of same cow as (Fig. 1 above) showing a squamous superficial layer with no noticable change in the thickness of the layer of polyhedral cells underneath (X125).
- Fig. 3. A section from the anterior end of the vaginal epithelium during mid pregnancy (129 days) showing an increase in height of the superficial cellular layer and a decrease in the number of polyhedral cells underneath. The cells of the superficial layer exhibit secretory activity. Cow No. 381 (X125).
- Fig. 4. A section from the posterior portion of the vaginal epithelium of same cow as Fig. 3 also showing some increase in the height of the superficial layer and some indication of secretory activity. The underlying layer of polyhedral cells is thicker than it is in the anterior region (X125).
- Fig. 5. A section from the anterior portion of the vaginal epithelium during late pregnancy (152 days) showing a high columnar secretory superficial layer. The underlying

- polyhedral cells are arranged in only one or two cell layers. Cow No. 375 (X125).
- Fig. 6. A section from the posterior region of the vaginal epithelium of the same cow as Fig. 5 showing that the secretory function is also present in this region.

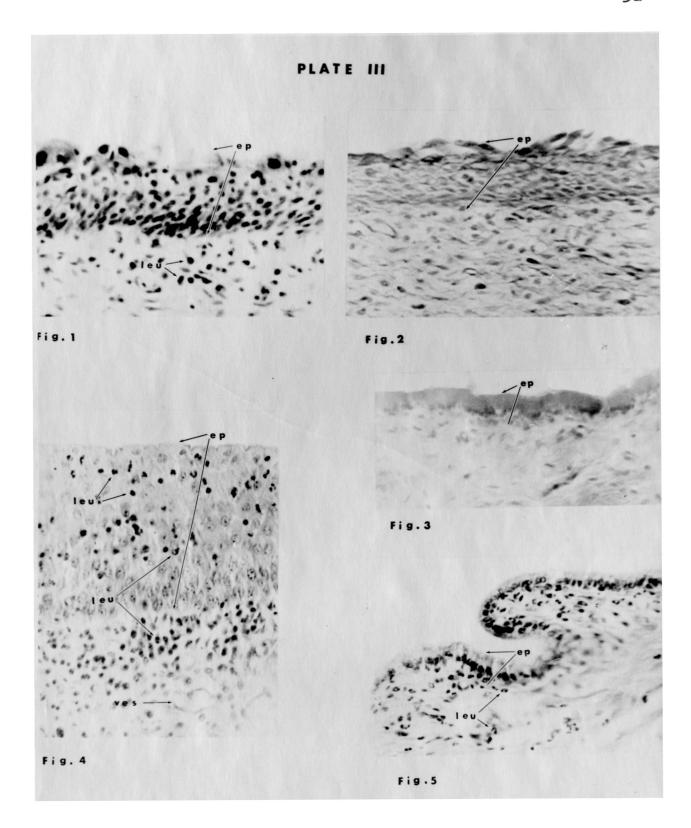
  Leucocytes are more evident than in the anterior end.

  (X125).



#### EXPLANATION OF PLATE III

- Fig. 1. A section from the anterior end of the vaginal epithelium, 15 days post-partum showing the low cuboidal superficial layer and the increased number of layers of the polyhedral cells underneath. Leucocytes are abundant in both epithelium and subepithelium. Cow No. 384 (X125).
- Fig. 2. A section from the posterior portion of the vaginal epithelium of the same cow as Fig. 1 showing the cornified superficial layer and many layers of polyhedral cells underneath (X125).
- Fig. 3. A section from the anterior portion of the vaginal epithelium during the follicular phase showing an intense response of the superficial layer to the Periodic Acid Schiff's reaction indicating the presence of glycogen in the mucoid secretion of these cells. Cow No. 382 (X125).
- Fig. 4. A section from the mid portion of the vaginal epithelium 25 days after abortion showing a highly stratified epithelium. Numerous leucocytes are present in the epithelium and a high concentration of these cells is evident in the subepithelium. Cow No. 118 (X125).
- Fig. 5. A section from the anterior end of the vaginal epithelium, 10 days post partum showing the low columnar superficial layer and the presence of leucocytes in epithelium and subepithelium. Cow No. 239 (X125).



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A thorough knowledge of the histological changes which occur in the bovine vagina is essential if the importance of this organ to successful reproductive performance is to be determined. The present study was undertaken to determine the cellular changes that occur in the bovine vagina during the various phases of the estrous cycle and pregnancy and whether these changes have any significance in reproductive performance.

Vaginal tissues were obtained from dairy cows, with known reproductive histories, eliminated from the Kansas State University dairy herd, supplemented by tissues from cows of unknown histories slaughtered at the Armour Packing Company plant, in Kansas City, Missouri. The cows from the Kansas State herd represented all stages of reproductive cycles from normal pregnancies to known pathological conditions. The tissues obtained from the packing plant were selected from clinically normal animals also representing various stages of the reproductive cycle. The stage of gestation of the pregnant cows was determined by fetus size; and the stage of the estrous cycle of the non-pregnant cows was established by ovarian structures.

Vaginal tissue was removed from anterior, middle and posterior portions of the vagina, as soon as possible after slaughter. After being secured they were immediately placed in 10% buffered formalin. Normal histological techniques were employed in preparing the tissues for study. The height of the epithelium was measured and cellular characteristics were recorded. Photomicrographs of the prepared sections representing the various phases of the reproductive

cycle were taken.

During the follicular phase of the estrous cycle the epithelium in the anterior portion of the vagina consisted of a superficial layer of highly secretory columnar cells and a basal layer
of polyhedral cells ranging from one to three cells in thickness.
The average height of the epithelium was 22.7 microns. In the
mid portion of the vagina during this stage of the cycle, the
superficial layer of columnar cells was very similar to that found
in the anterior portion. However, the underlying layer of polyhedral cells increased slightly in thickness. The epithelium in
the posterior region during the follicular phase of the estrous
cycle varied considerably between animals. In some cows the
superficial layer was highly secretory while in others only a few
secretory cells were noted. The underlying polyhedral layer of
cells was found to be highly stratified. The average height of
the epithelium in this region was 36.3 microns.

Shortly after ovulation the superficial layer of the vaginal epithelium began to lose its secretory characteristics. In the anterior region of the vagina, the columnar cells had become cuboidal and there was an increase in the number of polyhedral cells. These changes were more noticeable distal to the cervix and only slight secretory activity was noted in the posterior vaginal epithelium. Throughout the luteal phase of the cycle, the superficial layer of the epithelium exhibited very little secretory function. This may have been due to the absence of estrogenic stimulation. Clinically normal cows exhibited no abnormal

numbers of leucocytes, however, a few more white blood cells were evident in the posterior region of the vagina than in the anterior end.

During early pregnancy the vaginal epithelium consisted of a non-secretory superficial layer of squamous cells and a stratified layer of polyhedral cells. By mid pregnancy it had changed to a highly secretory epithelium characterized by a secretory, high columnar superficial layer and a reduced number of polyhedral cells. The changes during pregnancy might have been due to the absence of estrogenic stimulation during the early stages and the high level of estrogen being secreted by the placenta during the late stages of pregnancy.

During a prolonged period of low estrogen activity, the secretory layer of the epithelium appeared cornified and cells were sloughing off. This was characteristic of vaginal epithelium obtained from post partum cows. During early post partum, the epithelium contained many leucocytes which was probably due to the processes involved in uterine regression.

This study would indicate that it might be possible to determine whether or not the reproductive tract was clinically normal by the presence of an abnormal number of leucocytes in the vaginal wall.