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Title: THE RELATIONSHIP OF BLOOD GLUCOSE LEVELS  
TO OVULATION RATE AND EARLY EMBRYONIC  
MORTALITY IN GILTS

Abstract approved: *Redacted for Privacy*  
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A study was made of blood glucose levels and their relationship to reproduction in gilts.

Twelve blood samples, two in each of three successive weeks, before and after breeding were taken. Half of the samples were obtained from gilts in a fasting state and half after feeding. All samples were analysed for glucose content.

Feeding consisted of six pounds per gilt per day of a standard gestation ration fed once daily in a meal form.

At  $25 \pm 1$  days after breeding the gilts were slaughtered and a count was made of the corpora lutea, live embryos, and dead embryos.

Fasting and post-feeding mean blood glucose levels were described and discussed for both the pre-breeding and post-breeding

periods. Blood glucose content was higher after feeding than during fasting both before and after breeding ( $P < .001$ ). The amount of rise, as well as mean blood glucose levels before or after feeding, was not significantly different when comparing the pre-breeding and post-breeding periods.

The amount of variability in blood glucose content was also described and discussed. A significant ( $P < .01$ ) quadratic relationship was found between pre-feeding blood glucose level and the number of days in gestation, but very little variation was explained thereby ( $r^2 = .049$ ).

Regressions and correlations of the reproductive measurements on various blood glucose measurements were calculated. The magnitude of difference between the pre-feeding and post-feeding blood glucose levels in the pre-breeding period had a significant negative correlation with ovulation rate, early embryo mortality, and total embryo mortality ( $P < .05$ ,  $P < .01$ ,  $P < .05$ , respectively). No similar relationship was found in the post-breeding period or over the experimental period as a whole. No significant relationship was found between reproduction and any of the absolute blood glucose levels.

Regressions and correlations of the reproductive measurements on coefficients of variation of the blood glucose measurements indicated no significant relationships.

No relationship was found between mean blood glucose levels and failure to exhibit estrus or failure to conceive.

The Relationship of Blood Glucose Levels to  
Ovulation Rate and Early Embryonic  
Mortality in Gilts

by

Paul Edward Day

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# THE RELATIONSHIP OF BLOOD GLUCOSE LEVELS TO OVULATION RATE AND EARLY EMBRYONIC MORTALITY IN GILTS

## INTRODUCTION

In the commercial production of livestock one of the cornerstones of success is reproduction, for without sufficient offspring no amount of labor, capital, and managerial talent can result in a profitable enterprise. Swine are somewhat unique among the major livestock species in that as a multiparous animal successful reproduction is not only a qualitative matter of whether or not young are produced, but also a quantitative matter of how many are produced. In cattle, one calf per dam would be considered very adequate reproduction. Considering, however, that the return from approximately six pigs is required to produce a weaned litter, the same one-to-one ratio would be a financial disaster in swine production. Beyond the financial point of merely breaking even, litter size continues to be of importance. As the number of pigs per litter increases the fixed costs borne by each pig go down and hence the profit per pig rises.

To the geneticist and the producer of breeding stock a large litter provides the opportunity to select from a greater number of genetic recombinations when choosing the parental stock for the next generation. Thus improvement of litter size is meritorious beyond the purely economic benefits.

The size of a litter at birth is determined by three primary factors: ovulation rate, fertilization rate, and embryonic mortality. The first of these, ovulation rate, sets an absolute maximum on the litter potential of any given estrus cycle; except in the rare instance of monozygotic twinning it is impossible for the number of pigs farrowed to exceed the number of ova shed. Fertilization must occur soon after ovulation if an ovum is to survive and develop; consequently, this second limitation is imposed shortly after the first. The final limiting factor is that of the total mortality of embryos from the time of fertilization to parturition. Frequently this is sub-divided rather arbitrarily into early mortality and late mortality, referring (approximately) to the first quarter and the final three quarters of gestation. Since the causes of embryo mortality are numerous this factor in litter size is defined with considerably less precision than the preceding two.

Geneticists, nutritionists, and physiologists have expended considerable effort in attempts to increase the average number of pigs born alive. Since fertilization rates of 95-100% are commonly achieved (Squiers et al., 1950, 1952; Robertson et al., 1951a; Perry and Rowlands, 1962) with little more effort than proper timing and use of a fertile boar, it has not been necessary to apply much of the effort in this area. Furthermore little embryonic mortality takes place in the "late" portion of gestation (Squiers et al., 1950, 1952;

Perry and Rowlands, 1962), so little work has been directed toward this portion of the problem either. The remaining factors--ovulation rate and early embryonic mortality--have been the object of much of the scientific effort directed toward increasing litter size. Some of the investigation done in animal reproduction has indicated a positive relationship between energy intake and the rates of both ovulation and subsequent embryo mortality. As a step toward understanding the mechanism(s) that control these phenomena it was the goal of this project to investigate some possible relationships between blood glucose levels of gilts on a normal diet and their rates of ovulation and early embryo mortality.

## REVIEW OF LITERATURE

It has long been recognized that litter size in swine is considerably smaller at farrowing than the potential indicated by the number of ova shed. An early reference to this discrepancy is made by Hammond (1914) in an investigation of fertility in domestic animals. By studying the reproductive tracts of sows obtained from a slaughter house, he found that the average sow had 20 corpora lutea on her ovaries. At term, however, the average litter was said to be only 12 pigs for the breed involved.

In a more controlled study, Squiers et al. (1952) found that at 25 days after mating, 35 percent of the potential number of embryos were either missing or dead. His data indicated that a further loss of 11 percent could be expected by time of parturition.

Perry (1954) made a study similar to that of Hammond in which he investigated the reproductive tracts of 863 sows and gilts sent to slaughter from commercial swine herds in England. His conclusion was that a loss of at least 40 percent of the ova took place by the time of parturition, which agrees closely with previous studies. Other studies (Perry and Rowlands, 1962; Squiers et al., 1950; Robertson et al., 1951a; Haines et al., 1958) have shown basically the same results.

Several ideas have been put forth concerning the reasons for

early embryo losses and the resultant decrease of litter size. One of the earliest and probably the most thoroughly disposed of reason for mortality was that of disease. That disease is a contributor to fertility problems is not questioned; however, embryonic mortality does occur in dams which to all appearances are healthy. Hammond's (1914) attempts to isolate bacteria which might be responsible for embryonic mortality were fruitless. He felt that the absence of disease organisms was further supported by instances of healthy embryos lying side by side with dead embryos.

Corner (1923) in an extensive survey of mammalian embryo mortality also concluded that embryo deaths occur in uteri which are disease free. By removal of the corpora lutea from rabbits shortly after breeding, he demonstrated that death of the embryos could be related to causes other than disease. Removal of the corpora lutea consistently resulted in death of the embryos by the fourth day (Corner, 1928). In a subsequent experiment (Allen and Corner, 1929) in which the corpora lutea were removed as before, injected extracts of swine corpora lutea prevented death of the embryos. This result substantiates the conclusion that death was related to loss of the corpora lutea rather than to disease or to stress resulting from the operation itself.

It appeared conclusive then that litter size can be reduced by embryonic mortality not related to disease.

A second suggested cause for early embryonic mortality is concerned with intrauterine crowding and/or spacing. There has been considerable difference of opinion in this area of thought.

In his early studies, Hammond (1914, 1921) concluded that crowding and/or spacing was not an important factor in embryo mortality. He based this opinion on the observation that while one uterine horn might be filled with a number of viable embryos, another would have only one or two dead embryos and nothing else. He also noted that some of the dead embryos were surrounded by healthy, apparently functional, membranes and that the size of an embryo did not appear to be related to the number of embryos in the uterine horn. This suggested to him that survival and ability to grow are related to the make-up of the individual embryo rather than to competition with neighboring embryos. Perry (1954) observed that litters of 20 or more at birth are not rare and concluded that "even at this high level of fecundity the number of pigs born is limited by factors other than the sow's capacity to accommodate and nourish the fetuses."

By contrast, Rathnasabapathy et al. (1956) found that there was a strong possibility that intrauterine crowding might be a major factor in embryonic death. In a group of 37 Landrace x Poland gilts a highly significant positive association ( $r = .406$ ,  $P < .01$ ) was found between litter size and length of uterus at 55 days of gestation. A

similar correlation was found ( $r=.552$ ,  $P<.01$ ) between the weights of the embryos and the uterine space occupied by them. The optimum linear space per fetus at this stage of pregnancy was calculated to be 350-450 mm. Fetal atrophy was certain to result from less space according to their data, and more space was not likely to be of benefit.

That an embryo spacing mechanism of some importance might exist was suggested by Corner (1923). It was also implied by the work of Perry and Rowlands (1962) who found that embryos in litters with low mortality tended to be spaced throughout the entire uterine horn, and only rarely overlapped.

As a part of a study conducted by Bazer et al. (1968) nine gilts were subjected to superinduction of embryos approximately 60 hours after the onset of estrus. The total number of embryos (transferred plus normally ovulated) averaged 20.2 per gilt, but at parturition the average litter was only 9.2 pigs. Since there was no differential mortality between natural embryos and transferred embryos, the uterus or some factor associated with the uterus was the limiting factor in litter size. The authors felt that their results suggested "a critical level in uterine capacity" and pointed out that this level "is not greater than that now reflected in the normal litter size of gilts."

In work done at Illinois, Dzuik (1968) made an extensive study of the effect of embryo numbers and uterine space on embryonic

survival. In addition to a control group, he set up five other groups of gilts, each having the space per embryo altered in a different manner. These included tying off of a uterine horn such that the ova from one ovary had twice as much space as those from the other ovary; cutting and tying of one oviduct so that the ova from the remaining ovary had the entire uterine capacity; pre-ovulatory removal of one ovary and the ipsolateral uterine horn which with compensatory ovulation from the remaining ovary resulted in twice the usual number of ova in the remaining horn; superovulation; and embryo transfer. Interpretation of data from 130 gilts gave little reason to believe that embryo mortality was the result of intrauterine crowding in gilts having less than 14 embryos. Since the average control gilt ovulated only 13.4 ova and suffered 26% embryo mortality it would seem that a good deal of mortality from causes other than crowding has yet to be explained.

The degree to which litter size is genetically controlled has also been well investigated. Lush and Molln (1942) used the combined records of five agricultural experiment stations and found heritability of litter size to be equal to 0.17; repeatability of litter size within a sow was also equal to approximately 0.17. These figures were based on 7415 litters from 2560 sows.

Boylan et al. (1961) pooled 1970 dam-daughter comparisons and derived a figure of  $0.03 \pm 0.07$  as a heritability estimate of first litter



size. Neither their overall figure nor any of their subgroup figures was significantly different from zero.

Similar estimates were made by Urban et al. (1966) who used the records of litters farrowed from 1944 to 1958. The estimates were  $0.09 \pm 0.04$  with  $n = 3119$  and  $0.17 \pm 0.03$  with  $n = 2033$  for heritability and repeatability respectively.

Other studies (Stewart, 1945; Squiers et al., 1950; Lasley, 1957) have also indicated that heritability and repeatability of litter size are low. It seems generally agreed that the response to selection for increased litter size will be quite small at best.

The nutritional aspects of variation in litter size have received considerable attention in recent years and appear to offer more beneficial results than have other areas of research. A relationship between nutrition and reproduction which alters the number of offspring by modification of both ovulation and embryo mortality rates has been demonstrated in both swine and sheep, and has been suggested by work done with cattle (Wiltbank et al., 1962), rabbits (Short et al., 1968), and possibly rats (Bellows et al., 1966).

That there is a reproductive response to nutritional changes has been recognized from ancient times. Clark (1934) mentions an observation by Aristotle that sheep in a favorable environment exhibited greater fertility, and another by Darwin that domesticated animals bred with greater frequency and prolificacy than did wild

members of the same species.

An early study of this phenomenon was conducted by Marshall and Potts (1921) using ewes. Flushing was carried out by feeding the treated group  $1/2$  to  $3/4$  lb. per day of a grain supplement. The flushed ewes over a period of five years averaged 18.1% more lambs born. It was also reported that treated ewes came to service earlier than did the controls. The authors mentioned that the ewes used were in relatively good condition and that they anticipated that this might lessen the effect of flushing.

Clark (1934) conducted two experiments using ewes in poor condition the first time and ewes in good condition in the second experiment. With the poorer animals he found an average increase of 0.4 ova per ewe in the flushed group. With those in good condition, the ovulation rate of the treated group was actually surpassed by the control group. Weight gains were 0.23 versus 0.02 lbs. per day for treated and control groups respectively in the first experiment. In the second trial (using ewes in good condition) the gains were 0.21 lbs. per day in the treated group and 0.04 lbs. per day for the controls. Thus the conclusions drawn were that flushing would increase the ovulation rate providing the animals were not in good condition initially, and that weight gain per se does not induce a higher ovulation rate.

In swine, some of the earliest work of this nature was done by

McKenzie (1928) in a study of swine growth and reproduction. Based on litter size at farrowing, weight gains during the two weeks prior to breeding, and weight gains during the four weeks after breeding, he concluded that flushing was of little value in swine and that increasing weights in early gestation were conducive to larger litters. The feed levels used were termed high ("fed rather generously"), medium (60-67% of high), and low (41-48% of high). Considerable doubt has been cast on these conclusions by work done in a different manner a number of years later.

Robertson et al., (1951b) conducted an experiment to determine the effects of several factors on reproduction in gilts. Among the treatments was a variation in the amount of feed given; one group was fed ad lib. and the other at 70% of this level. Half of the gilts were killed one or two days after breeding and the remainder 25 days post breeding. In the overall analysis of the ad lib. fed group shed 1.1 more ova at the second estrus ( $P < .05$ ) and had fewer normal embryos at 25 days of gestation (43% vs. 67%;  $P < .05$ ) than did the group which was limited in its feed consumption.

Christian and Nofziger (1952) reported a similar experiment in which the ad lib. group ovulated 1.7 more ova ( $P < .05$ ). The gilts in their study were allowed to go to term; the ad lib. group farrowed 2.7 more pigs per litter ( $P < .01$ ).

In both of the foregoing experiments the gilts had been on their

respective levels of feed intake from before puberty. To determine whether the prepuberal feed level was of importance, Self et al. (1953) fed gilts on a high or low plane of nutrition from 72 days of age to puberty. At that time half of each group was switched to the opposite feeding regimen, and all were bred at the second estrus. Half were slaughtered one or two days later and the other half at 25 days of gestation. Counts taken of corpora albicantia and corpora lutea indicated that ovulation was greater at first estrus (1.63 more ova;  $P < .05$ ) in gilts which had been on the higher energy prepuberal diet and greater at the second estrus in gilts which received the higher energy level in the interval between first and second estrus.

Of those on the low energy plane just prior to breeding the gilts which had been full fed before puberty ovulated (with the exception of one subgroup) more than the prepuberal limited-fed group. No levels of significance were given for these figures however. In regard to embryo mortality, the authors concluded that "regardless of pre-puberal feed levels, gilts full-fed from puberty to slaughter tended to have approximately 25% fewer embryos than corresponding limited-fed gilts."

In a subsequent experiment, the same workers (Self et al., 1955) used the high and low feeding levels at three different ages, i.e. pre-puberty, first estrual cycle, and the first 25 days of gestation. Feeding levels were switched from one time period to another

in appropriate experimental groups. As before, extra feed in the period just before ovulation gave a significant rise to the number of ova shed, and continued high level feeding after breeding resulted in increased embryo death. A further result of interest though was that the best rate of embryo survival was found in those which had been limited-fed through the entire program and the worst rate of survival was among those being full fed throughout the program. This suggests that there is a carry over effect of flushing on the embryo survival rate even though feeding is reduced after breeding. In terms of litter size, however, the response to flushing was great enough to override this effect. Thus the greatest litter size at 25 days gestation was found in those gilts which had been limited-fed to puberty, full-fed during the next cycle, and again limited-fed after breeding.

In sheep El-Sheikh et al. (1955) flushed a group of ewes by supplemental feeding of two pounds of grain per head per day and found (in one year of a two-year study) that the ewes on a higher plane of nutrition had a significantly lower (26.9% vs. 100.9%,  $P < .01$ ) rate of embryo survival. Although this was not the case in both years, the trend was the same in the second year; consequently, the authors concluded that their data were in harmony with the higher mortality associated with a post breeding high plane of nutrition reported in swine by Robertson et al. (1951b), Christian and Nofziger (1952), and Self et al. (1953, 1955). Others have suggested that

continued high levels of feeding might be detrimental to the embryo in sheep (Foote et al., 1959; Hulet et al., 1962; Bellows et al., 1963b), but their results have been inconclusive. None of the experiments with sheep have used the reciprocal feeding arrangement described by Self, so it cannot be determined whether the mortality is due to high level feeding before breeding, after breeding, or both.

Having established that a relationship exists between feed intake and reproduction, interest was directed toward such questions as what in the increased feed elicited the response; when, more precisely, was the time that increased feed altered reproduction; what was the site of action; and what was the mode of action.

In all of the work previously referred to concerning sheep, the flushing was accomplished by the feeding of grain supplements. In a report by Gerring (1954), however, the method used was simply to put the "treated" ewes on better pasture than the control group. The result was a consistently higher percent of twin lambs born to the pasture flushed ewes, thus it is not likely that anything peculiar to grain alone is the responsible feed component.

To investigate the role of protein in flushing, Memon et al. (1969) in a factorial experimental design fed ewes two levels of energy and two levels of protein. Weight gain, number of corpora lutea, and percent of multiple ovulations were all significantly increased by the higher energy content, but were unaffected by the

level of protein.

The swine flushing studies cited to this point relied on simple alteration of the amount of feed given without any changes in the formulation of the feed. Haines et al. (1955, 1959) reported work with gilts receiving high and low energy intakes in which the feed of the low energy group was augmented to provide an equal intake of protein, vitamins, and minerals. The usual response in the high energy group of increased ovulation and embryo mortality rates were noted although the latter was not statistically significant. In a similar project, Gosset and Sorensen (1959) found an increased ovulation rate (greater by 1.1 ova, non-significant) and a decreased embryo survival rate (57.7% vs. 74.5%,  $P < .01$ ) in the high energy group.

The effect of protein during gestation was considered by Clawson et al. (1963) who fed it at two different levels. The level of protein had no significant effect on litter size at parturition.

With evidence growing that it was the energy component of the ration which was acting on reproductive performance, consideration was given by Kirkpatrick et al. (1967a) as to what sources of energy were effective. Gilts were held on a basal ration until approximately 14 days prior to the third postpuberal estrus at which time the group was divided into three subgroups. One of these served as a control and the others were given increased energy in the form of glucose or

corn oil. Both treated rations were equal to each other in caloric content, and all rations were adequate in terms of other nutrients. The gilts were slaughtered three to five days after the third estrus and the new corpora lutea counted. Gilts on the glucose and corn oil rations had 13.0 and 13.2 corpora lutea respectively as opposed to only 11.7 in the control animals ( $P < .05$ ). These results plus those of Rigor et al. (1963) who reported a significant ovulation response to the same caloric intake of lard, led the authors to conclude that there was little difference between isocaloric quantities of glucose, corn oil, or lard as an energy source for flushing gilts.

Zimmerman et al. (1958) fed gilts at two-thirds of the normal feed intake from 154 days of age to day eight of the second estrual cycle. They then flushed half of them for the remainder of the cycle and observed a 2.1 ova increase in the treated group as compared to the controls ( $P < .01$ ) which demonstrated that high energy feeding was required for less than a full cycle to flush gilts. In a following experiment the same group (Zimmerman et al., 1960) flushed gilts for 6, 10, and 14 days prior to ovulation. There was a significant increase in ovulation for each of the flushed groups, and a trend toward greater response with a longer flushing period. In a study by Kirkpatrick et al. (1967b) flushing for eight or 12 days gave the usual response, but only four days did not. Thus it appears that as the period of flushing within the preovulatory cycle is shortened the



response is diminished. McGillivray et al. (1962) and Kirkpatrick et al. (1967a) have also reported responses to a short (ten and 14 days respectively) period of high energy feeding in gilts.

An experiment was designed by Bazer et al. (1968) to investigate the viability of embryos from flushed and non-flushed gilts. After flushing half of the gilts for 14 days all were bred.

Approximately 60 hours after the onset of estrus embryos from flushed gilts were transferred to the uteri of non-flushed gilts and vice versa. Embryonic mortality was highest in the flushed group (which received embryos from non-flushed gilts) and lowest in the non-flushed group (carrying embryos from flushed gilts). The conclusion drawn was that mortality resulting from high energy feeding prior to breeding is due to some uterine factor rather than a decreased ability of the embryo to survive.

The mechanism or mechanisms by which added energy at the appropriate times modifies the ovulation rate and the amount of embryo survival are not yet fully understood. Work has been done in several areas but the results seem inconclusive.

Effects of high energy feeding on follicular development have been shown in both sheep and swine. El-Sheikh et al. (1955) reported that there were more follicles greater than 2 mm. in diameter and that the largest follicle was larger in the ewes which had been receiving supplementary energy. Howland et al. (1966) also noted more

large follicles (greater than 4 mm. in diameter), and greater follicular fluid weight throughout the cycle in flushed ewes.

In swine Rigor et al. (1963) reported more mature follicles and greater follicular fluid weights on day one of the estrus cycle in those gilts which had been on the higher energy level. In contrast to this, Kirkpatrick et al. (1967b) found no effect of feed level on the number of follicles, average diameter of the four largest follicles, or follicular fluid weight at day 19 of the cycle. It was noted by the authors that this discrepancy with the findings of Rigor et al. (1963) might be due to time-in-cycle differences.

Increased follicular development would imply, among other things, increased production of gonadotrophins by the pituitary; this possibility has been investigated by several groups.

In swine Rigor et al. (1963) made a gonadotrophin assay of the pituitary glands of gilts on two energy levels on the first day of estrus and found no difference in follicle stimulating hormone (FSH) or luteinizing hormone (LH) content. Kirkpatrick et al. (1967b) measured pituitary FSH and LH on days 3, 7, 11, 15, and 19 of the estrus cycle. In terms of mean levels of residual gonadotrophin activity, the findings were in agreement with the results of Rigor et al. (1963), i. e. no significant difference. There was, however, a difference ( $P < .05$ ) between feed groups in quadratic regressions of gonadotrophin activity on the day of cycle. In the high-energy group both

FSH and LH activity were lower early in the cycle and higher in mid-cycle when compared with the low-energy gilts. Late in the cycle FSH activity was again lower in the high-energy group while LH activity was about equal in both energy groups. In the discussion it was these results which led to the suggestion that the effect on follicular development reported by Rigor et al. (1963) on day one of estrus might not be incompatible with the lack of follicular response noted on the days of examination in this trial.

Studies using sheep (Bellows et al., 1963a; Howland et al., 1966) have indicated no effect on pituitary FSH concentration arising in conjunction with different feed levels. Both, however, noted an increase in pituitary weight in those ewes receiving greater amounts of energy, which would result in greater total FSH content. The results concerning LH were not in agreement. Bellows et al. (1963a) found no difference in LH concentration of the pituitary glands whereas Howland et al. (1966) reported increased LH concentrations at all stages of the cycle in those which had been subjected to the high energy treatment. In either case though the increased pituitary weight would result in greater total content of LH in the high energy ewes.

As a reason behind increased follicular development an alternative to pituitary involvement would be the possibility of greater sensitivity of ovarian tissue to the same amount of gonadotrophin

production. Bellows et al. (1963a) and Rigor et al. (1963), using sheep and swine respectively, have cast doubt on this idea. In both instances, the procedure was to block the release of endogenous pituitary gonadotrophins by injection of exogenous progesterone. This was followed by injections of exogenous gonadotrophin (pregnant mare serum--PMS) and measurement of the ovarian response. Both reported a response to the PMS, but neither found a differential response due to level of feed intake.

In a post-breeding study of the effects of feed level on pituitary gonadotrophin content Haines and Warnick (1959) found no significant differences between treatment groups. The contribution of this result to an understanding of the nutrition-embryo mortality relationship is somewhat questionable though since there was only a non-significant difference of 0.2 embryos per litter at 25 days gestation between the high energy and low energy groups.

Work with luteal characteristics as modified by energy intake has been reported by Howland et al. (1966) with ewes and by Kirkpatrick et al. (1967b) with gilts. The results are in conflict, with the former showing no differences in progesterone concentration or other luteal characteristics while the latter found a significant rise in both the progesterone concentration and the size of the average corpus luteum (hence greater total progesterone production) in the high energy group.

Goode et al. (1965) cited previous work concerning the relationship of alkaline phosphatase and acid phosphatase to reproduction, and reported an experiment which dealt with two levels of energy intake, embryo mortality, and phosphatase activity in the endometrium of gilts. A significant negative correlation ( $-.35$ ,  $P < .05$ ) was found between endometrial alkaline phosphatase and the number of viable embryos at 25 days of gestation. The regressions of endometrial acid phosphatase on embryos surviving to the 25th day after breeding was positive but non-significant. While there was no significant feed effect on the phosphatase activity, the regressions just noted were most pronounced in low energy feeding groups.

O'Bannon et al. (1966) reported a similar experiment in which there was no clear relationship between the phosphatase enzymes and reproduction except in various dietary sub-groups. It was hypothesized that this indicated a possible effect of energy on embryonic well being which was mediated directly or indirectly through the phosphatases, but the authors decline to speculate on how this took place citing the need for further research.

✓ Turning from the primarily nutritional and endocrinological aspects of the energy-reproduction complex it would seem logical to consider what happens to the feed after it is ingested. In most swine rations the bulk of the energy is provided by starch from various grains which is broken down in the digestive tract of swine to glucose.

The glucose is readily absorbed into the bloodstream and transported to the various tissues of the body for either immediate use as an energy source or for conversion and storage as glycogen. Some of those who have investigated the response to added energy intake have as a part of their work measured the blood glucose content.

Such an experiment was reported by Zimmerman et al. (1958, 1960) in which blood samples were taken just prior to and approximately one hour after feeding. The treated group (which was receiving extra energy in the form of glucose at the rate of one percent of their body weight per day) exhibited a rise in blood sugar from the fasting to post-feeding sampling time that was greater by an average of 9.4 mg. % ( $P > .05$ ,  $< .10$ ). The same group of gilts ovulated an average of 2.1 more ova ( $P < .01$ ). There was no indication of any difference in fasting blood glucose levels.

In the study conducted by Kirkpatrick et al. (1967a) using isocaloric quantities of glucose or corn oil as a source of supplemental energy, blood samples were taken at 2, 8, and 16 hours post-feeding. The average of the blood glucose levels was affected ( $P < .06$ ) by feed with the levels being 87.5 mg. %, 97.4 mg. %, and 108.0 mg. % for the basal ration, basal ration plus glucose, and basal ration plus corn oil respectively. Both of the augmented rations resulted in an increased ovulation rate. A regression of corpora lutea number on blood glucose level was reported to be only .002 ova/mg. %.

This regression was not significant. It was also noted that the maximum rise in blood glucose occurred later in the corn oil group.

In an experiment with pregnant ewes in which flushing did not take place Wright et al. (1962) measured blood glucose levels and commented that the "energy content of the ration appeared to directly affect blood glucose level."

An increase of 8.4 mg. % ( $P < .01$ ) between the plasma glucose levels of flushed and control ewes was reported by Howland et al. (1966). Ovulation rate, follicular fluid weight, number of large follicles, weight and dry matter percent of the anterior pituitary, and LH activity were also significantly higher in the flushed ewes ( $P < .01$  for each item).

In summary, the means by which the dietary energy level helps to regulate reproductive capacity is still obscure; evidence, however, suggests an involvement between high energy intake and greater follicular development which is brought about by increased production of pituitary gonadotrophins. A relationship between energy and progesterone production may or may not exist. Such is also the case with energy and phosphatase activity in reproductive tissues. Blood glucose levels appear to be raised by inclusion of larger amounts of energy in the diet and may be related to some of the reproductive parameters which have been discussed.

The objective of the present study was to determine blood

glucose levels as a characteristic of non-flushed individual gilts and to ascertain whether differences among individuals in blood glucose levels were related to ovulation rate and embryo mortality.



## METHODS AND MATERIALS

The data presented in this study were gathered in an experiment conducted at Oregon State University during the winter of 1968-1969. In its original form the study was used to determine what effects, if any, 800 mg. per head per day of the anthelmintic dichlorvos (2, 2-dichlorovinyl dimethyl phosphate) would have on reproduction and on blood glucose levels in gilts. The gilts were grouped in a  $2^2$  factorial design for purposes of feeding dichlorvos both before and after breeding. Analysis of the blood glucose data indicated that dichlorvos treatment had no effect on blood sugar level during the treatment period (England et al., 1969). For the purpose of evaluating the relationship of blood glucose and reproduction, therefore, the gilts were considered to be of one group all receiving equal treatment.

The trial began with 80 gilts of either Yorkshire or Yorkshire x Berkshire breeding from the Oregon State University 1968 spring farrow. The gilts were housed in pens which had slotted floors over the entire area except for concrete floored individual feeding stalls along one side. Cup type automatic waterers were provided. At the beginning of the experimental period the gilts were approximately nine to ten months of age and, as a group, had been observed to be expressing estrus. After initiation of the experiment, as each gilt exhibited estrus, the date was recorded and she was placed in the

experiment. At the next estrus each gilt was bred and  $25 \pm 1$  days later (the first day of observed estrus was termed day one) was sacrificed for examination of the reproductive tract.

### Feeding Procedure

The gilts were fed once daily at the rate of six pounds of feed per head per day. To eliminate competitive feeding between fast and slow gilts the feed was placed in the end of a concrete floored individual feeding stall. The feed was fed as a dry meal, the composition of which is presented in Table 1.

Table 1. Experimental ration.

Ingredients	Percent of Ration
Wheat	65.0
Alfalfa	25.0
Fishmeal	3.0
Soybean Oil Meal	3.0
Tankage or Meat and Bone Scraps	3.0
Salt	0.5
Ground Limestone or Oyster Shell Flour	0.5
Zinc Sulphate	12 oz./ton
Irradiated Yeast	.05 lb./ton

### Breeding Procedure

After the gilts had been put on trial they were checked daily for estrus either visually for enlargement and increased vascularization of the vulva or by use of a boar. When a gilt entered the next estrus phase of her cycle she was bred on two consecutive days (with rare exception). To eliminate any differential in the ability of the embryos to survive due to heterosis, gilts of the Yorkshire breed were bred to boars of Yorkshire x Berkshire breeding, and Yorkshire x Berkshire gilts were bred by Yorkshire boars. Following breeding, daily inspection was continued in order to detect any reoccurrence of estrus before the scheduled slaughter date.

### Bleeding Procedure

During the course of the experiment 12 blood samples were scheduled to be collected from each gilt. They were arranged such that during the cycle prior to the estrus at which a gilt was to be bred, three samples would be taken while the gilt was in a fasting state (i.e. just prior to that day's feeding) and each of these would be followed by a sampling on the same day 90 or more minutes after the gilt had been fed. It was attempted to space the three bleeding days somewhat evenly throughout the cycle although no rigid schedule was maintained to this end. A similar bleeding program was followed

after the gilts were bred. Thus a full set of blood samples would consist of three pre-feeding pre-breeding samples, three post-feeding pre-breeding samples, three pre-feeding post-breeding samples, and three post-feeding post-breeding samples.

On any given day, the gilts which were due to be bled were sorted out and confined in a temporary holding pen. From this they were individually driven through a chute into a steel restraining crate in which they were held while the bleeding was accomplished. They were then released into a second holding pen and kept there as a group until the pre-feeding sample had been drawn from all gilts for that day. After being returned to their pens, all were fed at the same time in the manner previously described. Beginning 90 minutes after feeding time, the collection procedure was repeated to obtain the post-feeding samples.

Blood was collected from prominent ear veins using a 20 gauge needle and a vacuum tube with a 5 ml. (approximately) draw. Each tube contained 10 mg. of potassium oxalate to inhibit clotting and 10 mg. of sodium fluoride to prevent glycolysis. The tubes were tilted and agitated during collection to insure thorough mixing of these reagents with the blood.

As soon as the blood collection was completed the blood samples were transported to a laboratory for processing.

### Blood Glucose Determination

Determination of the blood glucose content was made by the method of Nelson (1944). Due to the large number of samples to be processed the procedure was modified somewhat in that after the initial step of deproteinization, the glucose-containing filtrate was frozen and stored at  $-19^{\circ}\text{C}$ . in stoppered glass test tubes. This was considered essential as a precaution against glycolysis by any micro-organisms which might have contaminated the sample during collection and/or processing up to this point.

### Collection of Reproductive Data

At  $25 \pm 1$  days of gestation the gilts were slaughtered at the Oregon State University Meats Laboratory. The reproductive tract was removed as soon as a gilt was killed and the uterine horns cut open for inspection of the embryos. A count of live and dead embryos was made, but no attempt was made to determine the age at death for dead ones. A visible heart beat was the criterion applied in determining that the embryo was live or dead. The ovaries were dissected from the surrounding tissues and the corpora lutea were counted to determine the rate of ovulation.

### Statistical Procedure

Differences between mean values in the data of this experiment were tested for significance using Student's t test. The least squares method of regression analysis was used to determine the relationship of variability of reproductive traits to the variability of blood glucose measurements (Mendenhall, 1967).

## RESULTS

### General

Of the gilts originally placed in the experiment 52 were found to be pregnant at  $25 \pm 1$  days after breeding. The remainder of those discussed herein provided blood data in the pre-breeding portion of the experiment and either failed to breed, or mated and were found to be not pregnant when slaughtered. Some animals which were bred and returned to estrus prior to slaughter were bred a second time; the original pre-breeding blood data were treated as a separate body of data provided by a gilt which bred but did not conceive.

### Blood Glucose Data

Since it was not possible to obtain the post-feeding samples at precisely the same time after feeding with each gilt, a record was kept of the elapsed time from feeding to collection of blood samples. This made it possible to correct the post-feeding blood glucose values to a constant time after feeding in order to eliminate the variation in blood glucose level due to the amount of time available for the feed to be digested and absorbed into the bloodstream. Linear regression was used as the method of adjustment. Regressions were calculated separately for the pre-breeding and post-breeding periods as

there was a difference of 11 minutes in the mean elapsed times arising from the reduced number of pigs to be bled in the post-breeding period. In the pre-breeding period the regression of blood glucose on minutes elapsed was .157 mg. % with a mean elapsed time of 139 minutes. The corresponding figures for the post-breeding period were .203 mg. % and 128 minutes respectively. The blood glucose values ranged from 23.6 mg. % to 164.8 mg. % in the fasting state and from 33.9 mg. % to 164.5 mg. % after feeding. The mean blood glucose values, actual and adjusted, are presented in Table 2.

Table 2. Blood glucose levels of gilts in mg. % (mean  $\pm$  s. d. ).

Sampling Period	Pre-feeding	Actual Post-feeding	Adjusted Post-feeding	Adjusted Post-feeding Minus Pre-feeding
Pre-breeding	63.8 $\pm$ 19.0	68.3 $\pm$ 19.8	69.2 $\pm$ 19.5	5.4 P < .005
Post-breeding	61.0 $\pm$ 13.4	68.6 $\pm$ 16.7	68.9 $\pm$ 17.1	7.9 P < .001

Blood glucose content increased after feeding for both non-bred and gestating gilts. The mean amount of rise was 5.4 mg. % and 7.9 mg. % for the pre-breeding and post-breeding periods respectively. Mean post-feeding glucose values during both periods were found to be significantly greater than the mean fasting values. The amount of increase in the pre-breeding period was not significantly different from the amount of increase in the post-breeding period.

The average fasting blood glucose level of the pre-breeding



period was not significantly different from the average fasting blood glucose level of the post-breeding period. The same is true of the corresponding post-feeding blood glucose values.

With the exception of the pre-feeding post-breeding values, no significant effect of day in cycle or day in gestation on blood glucose level was found by using linear or quadratic regression analysis. Analysis of the pre-feeding post-breeding blood glucose values indicated a significant ( $P < .01$ ) quadratic relationship when blood glucose content was regressed on the number of days into gestation. The regression formula derived was:

$$\hat{y} = 48.28 + (2.3576)(x) + (-.0894)(x^2)$$

where  $\hat{y}$  = the predicted value in mg. % of the pre-feeding blood glucose content in a pregnant gilt, and  
 $x$  = the number of days into gestation.

The coefficient of determination ( $r^2$ ) was .0490, and the regression was calculated with 134 degrees of freedom. Mean values by day of cycle and day of gestation are listed in Tables 3 and 4 respectively.

Since the differences between pre- and post-breeding periods were not statistically significant, the pre- and post-breeding data for each gilt were pooled in several ways to develop blood glucose profiles. These profiles were designed to characterize the gilts, as individuals, in various states (i.e., fasting, fed, gestating, etc.)

Table 3. Blood glucose levels of gilts in mg. % on various days of the estrus cycle.

Day	Pre-feeding			Post-feeding		
	Mean	Standard Deviation	n	Mean	Standard Deviation	n
2	82.5	34.88	3	68.9	15.53	3
3	62.0	19.67	13	63.8	15.33	13
4	64.5	17.09	12	65.4	7.29	12
5	52.1	8.29	4	71.2	11.57	4
9	53.9	7.56	5	55.3	11.64	5
10	66.1	27.18	9	61.0	12.17	9
11	60.8	14.43	13	68.8	22.85	13
12	65.9	15.71	3	76.0	4.46	3
13	60.7	9.76	2	71.2	5.73	2
15	57.2	9.00	4	64.0	22.71	4
16	64.6	13.18	12	71.9	26.89	12
17	77.3	39.86	7	72.0	28.00	7
18	54.8	16.37	5	63.6	16.94	5
19	64.4	7.32	5	59.5	4.03	5

Table 4. Blood glucose levels of gilts in mg. % on various days of gestation.

Day	Pre-feeding			Post-feeding		
	Mean	Standard Deviation	n	Mean	Standard Deviation	n
3	55.9	9.25	6	75.3	19.22	6
4	63.7	24.57	6	71.8	30.03	6
5	56.5	2.50	4	66.4	6.98	4
6	61.1	5.08	3	69.6	7.73	3
7	52.0	4.90	5	67.0	11.34	5
8	51.5	8.63	5	57.3	8.14	5
9	52.7	13.58	2	54.9	10.96	2
10	64.4	15.83	5	73.6	29.28	5
11	69.5	16.88	6	60.8	7.32	6
12	65.5	19.45	5	68.1	15.27	5
13	86.2	26.60	3	80.8	24.75	3
14	54.8	9.71	6	61.6	10.92	6
15	79.2	29.63	4	70.1	7.03	4
16	60.2	0.29	3	54.9	9.97	3
17	59.2	8.05	6	66.4	18.85	6
18	67.8	25.74	3	74.6	11.55	3
19	59.3	6.49	6	66.8	12.51	6
20	59.9	5.31	7	66.0	6.38	7
21	63.3	2.90	3	78.6	18.81	3
22	56.4	8.72	8	60.8	8.59	8

and were later used in regression analysis to determine association with reproductive data. The following profiles were developed for each gilt:

- a. Pre-feeding average. The sum of all pre-feeding blood glucose values divided by the number of samples.
- b. Post-feeding average. The sum of all post-feeding blood glucose values divided by the number of samples.
- c. Pre-breeding average. The sum of all pre-breeding blood glucose values divided by the number of samples.
- d. Post-breeding average. The sum of all post-breeding blood glucose values divided by the number of samples.
- e. Overall average. The sum of all blood glucose values divided by the number of samples.
- f. Average pre-breeding difference. The difference of the post-feeding blood glucose value minus the pre-feeding blood glucose value summed for all sets of blood glucose values in the pre-breeding period and divided by the number of sets.
- g. Average post-breeding difference. The difference of the post-feeding blood glucose value minus the pre-feeding blood glucose value summed for all sets of blood glucose values in the post-breeding period and divided by the number of sets.
- h. Average overall difference. The difference of the post-feeding blood glucose value minus the pre-feeding blood glucose value

summed for all sets of blood glucose values and divided by the number of sets.

From these individual mean values a mean of means has been calculated for each profile to characterize the entire group of gilts. These means are tabulated in Table 5.

Table 5. Population blood glucose values for fasting and fed gilts in pre-breeding and bred statuses (mean  $\pm$  s.d. ).

Profile	Mean	Standard Deviation
a. Pre-feeding average	63.02	7.84
b. Post-feeding average	69.93	9.55
c. Pre-breeding average	65.80	10.95
d. Post-breeding average	65.07	8.33
e. Overall average	67.84	7.87
f. Average pre-breeding difference*	5.32	12.46
g. Average post-breeding difference*	6.99	11.62
h. Average overall difference*	6.37	9.72

\*These data include both positive and negative quantities which result in standard deviations which are larger than the means.

Considerable variation was found to exist in individual gilts within both the fasting and fed states. As a means of expressing this variability several coefficients of variation were determined for each gilt using the same format as was used in construction of the previously mentioned profiles. Coefficients of variation were not calculated for the pre-breeding average, post-breeding average, and overall average profiles, since they contain some artificially induced

variation which results from the pooling of pre-feeding and post-feeding data. The average pre-breeding difference and average post-breeding difference profiles were also omitted as it was felt that they contained too few samples ( $n=3$ ) to be of meaningful use. On an individual basis these coefficients of variation are used in regression analysis to determine if variation in blood glucose content is associated with reproduction. The population means of the coefficients of variation are presented in Table 6 for the appropriate profile groups.

Table 6, Population means of the individual gilt coefficients of variation of blood glucose values in fasting and fed states.

Profile	Mean	Standard Deviation
a. Pre-feeding average	24.13	12.25
b. Post-feeding average	22.02	9.65
h. Average overall difference	27.10	16.69

A regression of the difference between pre-feeding and post-feeding blood glucose levels (fed minus fasting) on the before feeding level was calculated to determine if the amount of change in blood glucose content is related to the fasting level. The regression indicated that smaller difference values are significantly ( $P < .01$ ) associated with high pre-feeding blood sugar levels. The prediction formula developed was:

$$\hat{y} = 40.94 + (-.5490)(x)$$

where  $\hat{y}$  = the predicted change in blood glucose content after feeding in mg. %, and

$x$  = the pre-feeding blood glucose content in mg. %.

The coefficient of determination ( $r^2$ ) was .2307, and the regression was calculated with 357 degrees of freedom.

### Reproductive Data

At  $25 \pm 1$  days after breeding the gilts were killed in the University's meats laboratory. The uteri were removed and examined immediately to determine the number of live and dead embryos. Observation of a visible heart beat was the criterion applied in determining if the embryo was viable. The ovaries were removed to a laboratory for a count of the corpora lutea.

The overall percent embryo mortality was defined as:

$$\frac{\text{no. corpora lutea} - \text{no. live embryos}}{\text{no. corpora lutea}} \times 100$$

In order to more closely study embryo mortality, all visible but dead embryos were termed "late" mortality and the difference between the total count of embryos (live plus dead) and the number of corpora lutea was termed "early" mortality. More precisely they were defined as follows:

$$\% \text{ late mortality} = \frac{\text{no. dead embryos}}{\text{total embryos}} \times 100$$

$$\% \text{ early mortality} = \frac{\text{no. corpora lutea} - \text{total embryos}}{\text{no. corpora lutea}} \times 100$$

Table 7 contains the means and standard deviations of the data collected from the reproductive tracts.

Table 7. Average reproductive performance of 52 gilts.

Item	Mean	Standard Deviation
Corpora Lutea	15.25	2.29
% Early Mortality	25.08	19.54
% Late Mortality *	6.27	9.27
% Total Mortality	29.83	19.32

\* The definition of this term (see above) prevents the sum of early mortality and late mortality from being equal to total mortality.

### Regression of Reproductive Data on Blood Glucose Level

To investigate possible relationships between blood glucose and reproduction a number of regressions were calculated using the individual gilt blood glucose profiles as the independent variable and the reproductive measurements as the dependent variable. The resultant regressions, correlations, and coefficients of determination are tabulated in Tables 8, 9, 10, and 11, for the number of corpora lutea, percent early mortality, percent late mortality, and percent total mortality respectively.



Table 8. Correlations and regressions of corpora lutea number on blood glucose profiles in gilts.

Profile	d. f.	r	$r^2$	Linear Regression Coefficient
a. Pre-feeding average	38	.1303	.0170	.0362
b. Post-feeding average	36	-.1962	.0385	-.0535
c. Pre-breeding average	39	-.0748	.0056	-.0146
d. Post-breeding average	39	.0529	.0028	.0176
e. Overall average	28	.0608	.0037	.0179
f. Average pre-breeding difference	40	-.3159*	.0998	-.0499
g. Average post-breeding difference	41	-.0282	.0008	-.0060
h. Average overall difference	30	-.1603	.0257	-.0423

\*P &lt; .05

Table 9. Correlations and regressions of percent early mortality of embryos on blood glucose profiles in gilts.

Profile	d. f.	r	$r^2$	Linear Regression Coefficient
a. Pre-feeding average	38	.1200	.0144	.2920
b. Post-feeding average	36	-.0741	.0055	-.1708
c. Pre-breeding average	39	-.0574	.0033	-.0932
d. Post-breeding average	39	.1549	.0240	.4386
e. Overall average	28	-.0264	.0007	-.0760
f. Average pre-breeding difference	40	-.4057**	.1646	-.5393
g. Average post-breeding difference	41	.0721	.0052	.1290
h. Average overall difference	30	-.2090	.0437	-.5178

\*\*P &lt; .01

Table 10. Correlations and regressions of percent late mortality of embryos on blood glucose profiles in gilts.

Profile	d. f.	r	$r^2$	Linear Regression Coefficient
a. Pre-feeding average	38	-.0100	.0001	-.0122
b. Post-feeding average	36	-.1053	.0111	-.1162
c. Pre-breeding average	39	.0754	.0057	.0533
d. Post-breeding average	39	.1024	.0105	.1432
e. Overall average	28	-.0223	.0005	-.0274
f. Average pre-breeding difference	40	.0632	.0040	.0357
g. Average post-breeding difference	41	.0489	.0024	.0429
h. Average overall difference	30	-.0032	.00001	-.0038

Table 11. Correlations and regressions of percent total mortality of embryos on blood glucose profiles in gilts.

Profile	d. f.	r	$r^2$	Linear Regression Coefficient
a. Pre-feeding average	38	.1220	.0149	.2864
b. Post-feeding average	36	-.1081	.0117	-.2394
c. Pre-breeding average	39	-.0264	.0007	-.0429
d. Post-breeding average	39	.1679	.0282	.4680
e. Overall average	28	-.0264	.0007	-.0720
f. Average pre-breeding difference	40	-.3774 <sup>*1</sup>	.1425	-.4830
g. Average post-breeding difference	41	.0692	.0048	.1215
h. Average overall difference	30	-.2017	.0407	-.4773

\*P &lt; .05

<sup>1</sup>This value approaches significance at the .01 level of probability where  $r = .393$  with 40 d. f.

The regressions of number of corpora lutea (Table 8), percent early embryo mortality (Table 9), and percent total embryo mortality (Table 11), on the average pre-breeding difference profile were significant ( $P < .05$ ,  $P < .01$ ,  $P < .05$ , respectively).

#### Regression of Reproductive Data on Blood Glucose Coefficients of Variation

To determine whether the variability of blood glucose content within the individual gilt is related to reproduction, regressions were calculated using the coefficients of variation as the independent variable and the reproductive measurements as the dependent variable. No significant relationships were indicated by linear or quadratic solutions.

#### Reproductive Failure and Blood Glucose Level

In a number of instances in this experiment there was a total failure of reproduction in that some gilts did not exhibit estrus a second time, or having bred at the second estrus failed to conceive and returned to estrus a third time. To determine if blood glucose levels were related to these phenomena, mean blood glucose values for breeders vs. non-breeders and conceiving gilts vs. non-conceiving gilts were compared. These comparisons are summarized in Table 12, and Table 13. No significant differences in mean blood

Table 12. Population blood glucose values in mg. % and occurrence of estrus in gilts.

Status of Gilts	Breeders		Non-breeders		Difference in Means
	Mean	Standard Deviation	Mean	Standard Deviation	
Pre-breeding, Fasting	64.3	19.5	62.9	18.7	1.4
Pre-breeding, Post-feeding	70.5	22.4	66.4	15.1	4.1
Pre-breeding, Post-feeding Minus Fasting	6.9	21.2	4.2	18.4	2.7

Table 13. Population blood glucose values in mg. % and occurrence of conception in gilts.

Status of Gilts	Gilts That Conceived		Gilts That Did Not Conceive		Difference in Means
	Mean	Standard Deviation	Mean	Standard Deviation	
Pre-breeding, Fasting	63.2	20.0	66.0	17.5	2.8
Pre-breeding, Post-feeding	69.9	22.5	72.5	23.6	2.6
Pre-breeding, Post-feeding Minus Fasting	7.5	22.0	5.8	20.4	1.7
Post-breeding, Fasting	59.4	12.8	62.0	15.9	2.6
Post-breeding, Post-feeding	66.8	16.2	74.4	25.2	7.6
Post-breeding, Post-feeding Minus Fasting	7.0	16.7	12.5	21.7	5.5

glucose values were found between gilts that exhibited estrus and were bred and those that did not exhibit estrus subsequent to the cycle at which they were included in the experiment (Table 12). Likewise, no significant differences in mean blood glucose values were found for gilts that conceived and those which mated but did not conceive (Table 13).

## DISCUSSION

There seems to be considerable variation in swine in blood glucose levels. Eveleth (1933) mentioned reported ranges of 38 to 500 mg. % and found levels in excess of 300 mg. % in his own work. The average fasting value in his control group of pigs was 49.2 mg. %. In fasting miniature swine Sowers et al. (1969) reported a mean blood glucose level of  $105.3 \pm 27.5$  mg. %. Consequently, while quite variable, the blood glucose levels cited in Table 2 appear to be compatible with other reports. Regression analysis shows no relationship of blood glucose level to day in estrus cycle (Table 3) or to day in gestation (Table 4) with the one exception of the pre-feeding, post-breeding glucose levels. A highly significant relationship was found to exist between these variables, yet the coefficient of determination (.0490) indicates that very little of the variability in blood glucose level is due to this relationship.

Some of the variability is probably explained by the work of Bunding, Davenport, and Schooley (1956), who found that one pig in eight has a "diabetic" type response to glucose loading of the blood.

The individuality of pigs in their tolerance of glucose is also brought out by the results of Sowers et al. (1969) which show a range of 0.92%/min. to 3.64%/min. in the ability of 41 pigs to clear injected glucose from their blood. Using the data in Table 1 of their

report, a coefficient of variation of 26.12 can be calculated for the fasting blood sugar level which is quite similar to the corresponding value (24.13) in Table 6 of this study.

Another factor in the variability of blood glucose content quite probably was that of excitation of the gilts during the bleeding process. Excitement has been shown by Meyer et al. (1962) to raise blood glucose levels sharply. Varying degrees of resistance were displayed by the gilts in this experiment while blood samples were being drawn.

That the mean number of corpora lutea (Table 7) observed in this experiment is slightly higher than would normally be expected in gilts can be explained by the fact that the gilts used were nine to ten months of age at the beginning of the experimental period. The amount of embryo mortality is similar to that reported by others in previous work (Squiers et al., 1952; Perry, 1954; Perry and Rowlands, 1962).

In comparing the various blood glucose profiles of each gilt to her reproductive performance (Tables 8, 9, 10, and 11), the absolute blood glucose level was not found to be significantly related to ovulation rate or embryo mortality rate. Among the three difference profiles however, the average pre-breeding difference was found to have a significant negative relationship to the ovulation rate and a significant negative relationship to the early embryo mortality rate, and the total (to 25 days) embryo mortality rate. The remaining

difference profiles bore no relationship to reproduction.

Little work has been reported comparing blood glucose measurements to reproduction in swine. Of the information that is available one report is in agreement and one in apparent disagreement with this study. Kirkpatrick et al. (1967a) regressed ovulation rate on blood glucose level and found no relationship between the two. The same results were found in this study.

Zimmerman et al. (1958) in their study of high energy feeding and reproduction noted a 9.4 mg. % greater difference (post-feeding minus pre-feeding) in the blood glucose response to feeding with the group of gilts that shed more ova. This would seem to be contrary to the results of this study where a larger difference between the two blood glucose values was associated with a smaller ovulation rate. The techniques employed in the two experiments were not the same and it is possible that this may have a bearing on the conflicting results. Outstanding among the differences is the fact that the post-feeding blood sample in the work of Zimmerman was taken one hour after feeding while the present study used post-feeding values adjusted to more than two hours after feeding. Considering the differing ability of pigs to clear glucose from the blood (Sowers et al., 1969) it does not necessarily follow that gilts having the highest blood glucose at an hour post-feeding would still have the highest levels at two hours post-feeding. More elaborate characterizations



of the blood glucose response of individual gilts to feed intake (such as repeated blood sampling over time after feeding) would be necessary to determine if these results are actually in conflict.

The gilts in Zimmerman's study were not bred, consequently there were no embryo mortality data to be considered as there were in this study.

Relating embryo mortality to a pre-breeding factor is not novel. As was discussed in the literature review (see page 13) Self et al. (1955) described an apparent effect of pre-breeding energy intake on embryo mortality in gilts.

The existence of a significant correlation between two variables does not prove that there is a cause and effect relationship between them. Therefore, in considering the relationships between blood glucose and reproduction described in this work it would seem advisable to consider what mechanism(s) might be involved if indeed there is a cause and effect relationship. Toward this end it is interesting to note that, using heifers, Lynn et al. (1965) have reported an increased production of progesterone in response to both in vivo and in vitro glucose treatment. In their experiment they gave glucose intravenously one half hour before removal of the corpus luteum on day 14 of the estrus cycle. The excised luteal tissue was incubated in a medium containing a progesterone precursor (pregnenolone) and analysed for progesterone. Those treated in vivo produced more

progesterone ( $P < .05$ ). Tissue incubated in a medium that also contained glucose (200 mg. %) produced more progesterone ( $P < .01$ ) regardless of whether or not there had been in vivo treatment. In the instance of in vivo plus in vitro treatment there was no interaction between the two treatments; thus it was suggested that the two glucose treatments were additive in their effect on progesterone production. The authors concluded "that the glycemic state of the animal may be involved in determining the relative level of steroidogenesis."

Previous work using similar in vitro techniques demonstrated the ability of swine corpora lutea tissue slices to produce progesterone with glucose and pregnenolone in the incubating medium (Duncan et al., 1960). In vivo experimentation of the nature described by Lynn et al. (1965) in heifers apparently has not been undertaken in swine, so it is impossible to say just how closely the gilt would parallel the heifer in response to glucose treatment. If, however, the corpora lutea of the gilt are responsive to changes in blood glucose content, and considering the intricate interrelationships of the reproductive hormones, then it would seem to be a reasonable possibility that the correlations between blood glucose measurements and reproductive measurements reported here are more than a statistical curiosity. To resolve this question would require considerable further research involving blood glucose content and variation;

production, release, transport, and action of the various reproductive hormones; and interrelationships of the two areas of study.

## SUMMARY

A number of comparisons were made between the blood glucose levels of gilts and their reproductive performance from early in the estrus cycle to  $25 \pm 1$  days of gestation.

A significant ( $P < .01$ ) quadratic relationship was found to exist when the pre-feeding, post-breeding blood sugar level was regressed on the day of gestation. The relationship, however, explained only a minor (4.9%) portion of the variation in the blood sugar level. With this exception, day in cycle or gestation was not found to be related to pre-feeding or post-feeding blood glucose levels.

For each gilt several blood glucose profiles were derived to characterize the animal both before and after feeding during the normal estrus cycle and during early gestation. Using these profiles as independent variables, linear and quadratic regressions were calculated with the rate of ovulation, percent early embryo mortality, percent late embryo mortality, and percent total embryo mortality being used alternately as dependent variables. No relationship was found between any of the absolute blood glucose levels and any of the reproductive parameters. Significant linear correlations and regressions were found to exist between the amount of change in blood glucose level resulting from feeding (post-feeding minus pre-feeding) in the pre-breeding period and ovulation rate ( $P < .05$ ),

percent early embryo mortality ( $P < .01$ ), and percent total embryo mortality ( $P < .05$ ). The corresponding values in the post-breeding period were not significantly related to reproduction, nor were the overall values which were an average of the pre-breeding and post-breeding values.

The reproductive measurements were also regressed on the coefficients of variation derived from the profiles for each gilt dealing with pre-feeding blood glucose level, post-feeding blood glucose level, and amount of change in blood sugar resulting from feeding. No significant linear or quadratic relationships were found.

Comparison of mean blood glucose levels revealed no significant differences between gilts which bred and those which did not breed. The same was true, among those which did breed, in comparing the gilts that conceived to those which did not.

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