# NEONATAL THYMECTOMY IN CALVES

THESIS FOR THE DEGREE OF M. S.

MICHIGAN STATE UNIVERSITY

DONALD L. BUELKE

1966

ABSTRACT

NEONATAL THYMECTOMY IN CALVES

by Donald L. Buelke

During the past decade the lack of immune competence produced in various species of laboratory animals by neonatal thymectomy has been repeatedly demonstrated. This experiment was conducted to explore the feasibility of such an operation in calves and to study some of the basic effects upon the animals with the possibility of using this procedure for creating animals of lowered immunologic capabilities for use in certain viral or neoplastic disease transmission studies.

Ten newborn Holstein-Friesian bull calves were alternately selected and placed into either the thymectomy or control group. Each calf was given an initial 2 liter hand feeding of colostrum which was previously obtained from several cows, mixed and frozen. Within 24 hours after birth the calves were thymectomized or subjected to a sham operation. The surgical procedure involved resectioning of the central

portion of the third left rib and bluntly dissecting the thoracic portion from its anterior mediastinal position; then placing the calf on his back and bluntly dissecting the cervical portion through a midventral cervical incision. The sham operation was identical with the exception of the extirpation of the gland.

Postsurgical care included appropriate antimicrobial and palliative treatment. The calves were fed an aseptically prepared milk replacer, a pelleted dry ration, hay, and water. During the first month of age all calves were troubled with frequent attacks of enteritis with ensuing loss of condition and anorexia. From about the fourth week until the termination of the experiment at 20 weeks of age no clinical disease entities were observed in either group.

From the weekly blood samples, no significant difference between groups for erythrocyte count, hemoglobin level, packed cell volume, total leukocyte count, and monocyte or neutrophil values of the leukocyte differential was detected. However, highly significant values ($P < 0.01$) for higher total serum protein and serum globulin, and lower serum albumin and albumin:globulin ratio were detected for the thymectomized group, as were highly significant lower weight gains. The thymectomized group also had lower relative eosinophil and lymphocyte values which approach significance at the $P < 0.05$ level.

# NEONATAL THYMECTOMY IN CALVES

by

Donald L. Buelke

A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Veterinary Surgery and Medicine

1966

ACKNOWLEDGEMENTS

The author gratefully acknowledges the encouragement and assistance given him by his wife, Teck. Special thanks are also extended to Dr. David Luhring for his assistance during surgery and throughout the project. The author also appreciates the guidance given by Dr. Gabel H. Conner and assistance given by Dr. R. W. Van Pelt and technicians in the department.

TABLE OF CONTENTS

# INTRODUCTION

In the past decade considerable emphasis has been placed on elucidating the function of the thymus gland, especially its relationship to immune mechanisms. In the past the function attributed to the thymus was that it had some obscure, or certainly controversial function; possibly concerned with lymphopoiesis, immunity, or sexual maturation; and the subject was dropped at that. Today researchers in the field generally concede that the thymus is concerned with immunologic development, although many details remain controversial.

Extensive research programs in the past five years have demonstrated the role which the thymus plays in early immunologic development in some animal species. A wasting syndrome has been demonstrated numerous times in neonatally thymectomized mice. Other species of laboratory animals have also been used to demonstrate various degrees of depression which neonatal thymectomy will produce on immunologic capacity. Recently there has also been an increase in the number of publications on the subject of agammaglobulinemia and various thymic

disorders. To date however, the effect of neonatal thymectomy in larger domestic species has not been reported.

Within the past 10 years studies have been conducted to demonstrate transmissability of the animal lymphomatoses. Early workers developed an inbred strain of leukemia-susceptable mice in order to gain better insight into the development and characteristics of the disease. However, those working with bovine and canine leukemia's are hampered in this respect because of the longer generation time and the economics involved with these species. Thus, investigative studies have been limited to spontaneous clinical cases. Since thymectomy at birth in a number of mammals has been shown to produce various degrees of immunologic incompetence, it is possible that thymectomy in newborn calves could result in an animal more susceptable to leukemia. Thymectomy would then be especially valuable in transmission studies on the bovine species.

This study was conducted to develop a suitable procedure for thymectomy in the newborn calf, explore the feasibility of such an operation, and study some of the basic effects upon the animal. The particular effects this study is designed to observe are changes or variations in: (1) erythrocyte count; (2) hemoglobin level; (3) packed cell volume; (4) leukocyte count;

(5) leukocyte differential; (6) total serum protein; (7) serum albumin and globulin; (8) weight gains; and (9) physiologic behavior.

# REVIEW OF LITERATURE

## Introduction

Review of veterinary literature reveals an absence of information on neonatal thymectomy in species other than those normally considered to be laboratory animals. The only documentation on thymectomy in calves is by Papp (1960) who performed the operation on older calves. On the other hand, there are numerous reports on the effect of thymectomy in laboratory mice, rats, hamsters, rabbits, and even dogs. It is this material which serves as a framework for the design of this study.

## Anatomy

Sisson and Grossman (1962) describe the thymus of the calf as a pale and distinctly lobulated gland which is divided into two connected parts. The thoracic part occupies most of the anterior mediastinal space. It is bordered by the pericardium posteriorly, molded by the great vessels on the right surface, bordered by the chest wall and the left apical lobe of the lung on the left, and is continuous anteriorly through the thoracic inlet with the cervical part.

The cervical portion forms the bulk of the gland. It is divided into a right and left lobe which extend from the thyroid gland to the thoracic inlet along the esophagus and trachea. The two lobes are larger and apposed at the base of the neck where they cover the esophagus, trachea, carotid arteries, and vagosympathetic trunks. Part way up they become separate entities and gradually taper off on the sides of the trachea. The description of Mcleod (1958), Frandson (1965), and Trautmann and Fiebiger (1957) coincides with that of Sisson and Grossman (1962).

Trautmann and Fiebiger (1957), Frandson (1965), and Andrew (1959) classify the thymus as an atypical endocrine gland, but many current texts on histology including Bloom and Fawcett (1962), Copenhaver (1964), and Maximow and Bloom (1957) regard the organ as being more closely allied to the lymphatic system. Trautmann and Fiebiger (1957) describe the microscopic anatomy of the thymus of domestic animals as 5 to 13 mm. pyramidal or polygonal lobules. Each lobule, on stained section, presents a dark peripheral cortex which may be partially subdivided by extensions of the surrounding connective tissue and a lighter medulla centrally. The fine meshes of the reticulum of the cortex contain vast numbers of small, compacted lymphocytes, or thymocytes, along with

a few eosinophils, plasma cells, and small stellate reticulum cells. The medullary area differs in that the lymphocytes are fewer in number and less compacted. The reticulum cells, too, are not as compacted and appear larger. Peculiar to the medulla of the thymus of all species are Hassall's bodies. These are acidophilic, concentric formations of epithelial reticulum cells which appear to be partially degenerated and hyalinized. According to Bloom and Fawcett (1962) the medullary tissue extends from one lobule to another.

Trautmann and Fiebiger (1957) state that the blood vessels are ensheathed in active lymphatic tissue. The arteries lie in the general area of the cortico-medullary junction and divide into a dense network of capillaries which in turn are drained by veins on the surface of the lobule. Lymphatics also encircle the medullary area and are drained by interlobular lymph channels. Nerves are almost exclusively vasomotor.

## Embryology

Bloom and Fawcett (1962), Andrew (1959), and Trautmann and Fiebiger (1957) describe the reticulum of the thymus in further detail. One of the main reasons some scholars will not classify the thymus as a lymphatic organ is that the thymic tissue itself, the reticulum, is of entodermal origin while lymphatic

tissue is of mesodermal origin. It arises by outgrowths from the third and fourth brachial pouches. During this histogenesis the surrounding mesodermal lymphoid primordium is incorporated into the structure, thus resulting in a mingling of two germ layers. The epithelial nature of the entodermal reticulum is more noticeable in the medullary portions of normal glands than in the cortex. It is quite pronounced in the embryo, in the irradiated thymus where the lymphocytes are depleted, and also in tumors of the thymus. Although no reference to the explicit embryology of the calf thymus could be found, Ruth (1964) mentions the fact that in the pig, chicken, and possibly in man and all mammals the thymus may have both an ectodermal and entodermal origin. This leaves the status of the embryology of the calf thymus in a state of conjecture.

## Physiology

Physiologically much remains to be learned about the thymus; however, certain basic phenomena and functions have been described. Good (1964) and Hammar (1921) state that the thymus is largest in relation to the body at birth and gradually grows until puberty, but at a much lesser rate than the body in general. At puberty the gland begins to involute and all but

disappears in some species according to Fisher (1964) and Trautmann and Fiebiger (1957), while in other species, particularly dogs and cattle, remnants of the thoracic part will remain until old age.

Accidental involution is not an uncommon entity. In the early part of this century it was probably this factor more than any other which caused the great confusion over the relationship of the thymus to certain childhood disease entities. Hammar (1921) wrote an extensive treatise reviewing and discussing previous work and current concepts on the thymus. It was evident to him that many references to the "normal" thymus were based upon postmortem findings in children who had died of a debilitating disease or inanition. By showing that these factors had caused the thymus to involute, he refuted the concept of "status thymicolymphaticus"--a widely held concept of the day which implicated the thymus in many unexplainable childhood deaths based upon gross evaluation of thymus size.

Possibly due to the superficial treatment given to the study of the thymus in basic courses in veterinary medicine, accidental involution in animals is a less often reported entity. Texts on anatomy such as Sisson and Grossman (1962) and Trautmann and Fiebiger (1957); or physiology, such as Dukes (1955) mention its occurrence only in passing. Turner (1955) reported that

adrenal corticosteroids, whether from exogenous or endogenous sources, as well as androgens, estrogens, gonadotrophins, or infections and intoxications cause involution. Trautmann and Fiebiger (1957) add that other causes of accidental involution include exhaustion, malnutrition, cachectic and debilitating diseases, and radiation.

Fisher (1964) states that almost any "stress" will result in thymic involution. Radiation and corticosteroids are two common means by which thymic involution can be produced either accidentally in therapy, or for experimental purposes. Hammar (1921) recognized the marked reactability of the thymus during periods of stress in his conclusion that the thymus is not just a dormant vestige.

From the reports of Trautmann and Fiebiger (1957), Good (1964), and Hammar (1921) it appears that the thymus normally begins to involute at the time of puberty in all mammals studied; a phenomenon termed age involution. Again, early scholars were confused by the turmoil of establishing a "normal" thymus size and accidental involution. Fisher (1964) states that involution, whether accidental or age, can only be determined by microscopic and histologic means, an important criterion not considered by earlier researchers.

Hammar (1921) gave one of the first systematic descriptions of age involution of the thymus.

Hammar's (1921) description was complete and accurate and has weathered the test of time. Bloom and Fawcett (1962), Copenhaver (1964), and Maximow and Bloom (1957) describe the involutionary processes much the same as Hammar had 40 years before. Briefly, the thymus is largest in relation to total body size at birth, but continues to grow until puberty, which is between 11 to 14 years in man according to Hammar (1921), or 6 to 18 months in cattle according to Roberts (1956), after which growth ceases and it begins to involute. Lymphoctes gradually disappear from the reticular network and adipose and fibrous connective tissue slowly replaces the parenchyma. Age involution is not a dramatic and definite process, but rather the slow disappearance of lymphatic tissue with an encroaching connective tissue proliferation. This process never becomes complete and traces of thymic tissue may still be present in aged individuals according to Hammar (1921), Dukes (1955), and Trautmann and Fiebiger (1957).

The function of the thymus has been very obscure and often debatable. Good (1964) reviewed the history of the thymus and found that as early as the 17th century, literature appeared describing the thymus with some conjecture as to its function. Late in the

19th and early in the 20th centuries, because of the popular concept of status thymicolymphaticus, enthusiastic research was conducted in an attempt to delineate the function of this organ. Hammar gathered information from most of the earlier investigations and developed a format which serves as a basis for much of the current work.

In one of his most comprehensive works, Hammar (1921) established 5 major premises describing the histogenesis, anatomy, physiologic behavior, pathologic variations, and postulated cell controlling hormones. However, by disproving the concept of status thymicolymphaticus, there appeared to be no pertinent clinical need for further studies on the thymus and it became an unpopular research subject for several decades.

Current interest developed simultaneously from several sources during the 1950's. Good (1964) developed interest in the function of thymus after being confronted with a patient exhibiting both an acquired agammaglobulinemia and a thymoma. He considered these rare conditions occurring together as too significant to be passed off as chance, and investigated the problem by studying thymectomized adult rabbits. However, from his unfortunate choice of subject, in retrospect, he could conclude nothing and consequently discontinued

his experiments. More recent work has shown this relationship more clearly, such as that reported by Burnet (1962a) and Good (1964).

About this time Chang (1955) discovered the immunologic role the thymus-like bursa of Fabricius plays in the chick. In preliminary studies Glick (1955) demonstrated a bursa-testes relationship and a relationship with other endocrine glands including the pituitary, thyroid, and adrenal glands which affected the bursa's development. He then bursectomized chicks at 2 to 26 days of age; but as others had found before, he could not demonstrate any gross effects produced by its removal. Six months later, 9 birds were borrowed by Chang, a fellow graduate student, for a class demonstration in immunology. Six of the birds injected with Salmonella typhimurium antigen died immediately, while the remaining 3 failed to produce any demonstrable antibody. Bursectomy and injection of S. typhimurium antigen was repeated in other breeds of chickens and a link between the bursa of Fabricius and the immune mechanism was revealed.

Not dismayed by earlier failures, Good (1964) returned to studying thymectomized animals, this time utilizing newborn rabbits. By challenging them with bovine serum albumin at 7 to 8 weeks he was able to demonstrate a significant reduction in antibody response, which varied with the type of antigen and

was insignificant if the thymectomy was not performed during the immediate neonatal period.

Miller (1961a) is probably the first to report the phenomenal syndrome in mice following neonatal thymectomy. Prior to this time the only well-established function of the thymus was lymphopoiesis. Evidence to support endocrine and immunologic functions was far from conclusive. He found in his experiments that if thymectomy was performed after the neonatal period, at 3 weeks, no overt differences from controls were detected. However, when thymectomy was performed on newborn mice, they appeared normal until 3 to 4 weeks of age, then developed a wasting syndrome characterized by weight loss, diarrhea, lethargy, ruffled hair, and a hunched posture with death ensuing in 1 to 3 weeks.

Parrott (1962a) and Jancovic (1962) duplicated and confirmed this work. Miller (1964) reported that attempts to save these afflicted mice by high protein, high vitamin diets and the therapeutic use of broad spectrum antibiotics failed; however, by reimplanting the thymus a 70 % recovery rate was achieved. On necropsy a common finding in wasted mice was depletion of small lymphocytes from the peripheral lymph nodes and spleen. When peripheral blood was studied an analogous depletion of lymphocytes was discovered although other blood cell types were not significantly different.

After the initial work of Miller (1961a) and Metcalf (1956) two theories developed regarding the function of the thymus. Metcalf believed a hormonal "plasma lymphocyte stimulating factor" was the mechanism by which the thymus functioned in immunity. Burnet (1962b) proposed that the thymus produced potential germ cells which seed various lymphoid organs during early life, a theory which Parrott (1962b) tended to support. Although Miller (1961a, 1962a) accepted this theory he could not refute Metcalf's theory and tended to combine the two.

Profound success stimulated research initiative and it appeared that the field was just beginning to open. Archer (1961) and Glick (1956) had already demonstrated diminished antibody response in other species, and Miller (1961a, 1962b) was able to demonstrate complete survival of skin homo-grafts on thymectomized mice. He discovered that as the time lapse before thymectomy increased, a poorer degree of homograft survival occurred, up to a point of about 3 weeks, after which homograft immunity was apparently normal. This work and variations of it were soon replicated by Martinez (1962a, 1962b, 1964a, 1964b). The theory that the thymus was the seat of immunologic development became more widely accepted, as exemplified in a report by Miller (1964b), although the exact mechanisms still needed clarification.

An interesting sidelight now appeared unpredictedly. For some time it had been known that thymectomy at 1 to 2 months of age prevented development of leukemia in inbred susceptable strains of mice as well as its development from usual carcinogenic methods. Miller (1961b) using mice and later Kunii (1965) using rats attempted to induce leukemia with viruses in neonatally thymectomized animals but met with failure. Miller offered a simple explanation by attributing the resistance to leukemia to an absence of leukemia-precipitating factors within the thymus. This is further supported by Siegler (1964) in view of the fact that lymphoma of mice is primarily of thymic origin. Furth (1964) has performed extensive research on the thymic lymphoma factor and its relationship to viruses known to induce leukemia.

Although Hammar (1921) and Metcalf (1956) proposed that humoral factors were produced by the thymus it remained a very controversial subject until recently. Lymphopoiesis was one of the earliest established functions of the thymus, but Miller (1962b), in light of recent developments, explored the mechanisms of this function in further detail. By thymectomizing mice shortly after birth and later reimplanting a donor thymus he found the mice did not suffer from wasting disease and

the thymus graft and the host's lymphopoietic system appeared to be functioning normally.

However, auxiliary information from these studies opened a new realm of thymic function. Lymphocytes of the implanted thymus as well as lymphocytes multiplying in the host's lymphoid organs were of host rather than donor origin. Secondly, transplanting a thymus from a young mouse into an old mouse whose thymus was deteriorating resulted in a rapid rate of lymphopoiesis as would be expected in the thymus of a young animal. This evidence tended to support the contention that a humoral factor may be secreted by the thymic reticulum which stimulated lymphopoiesis in dormant lymphoid organs of a neonatally thymectomized host. Metcalf (1956) originally reported a hormone produced by the thymus and found in high levels in human leukemia patients. Levey (1963) set out to further investigate the stimulating influence of the thymus. By implanting a donor thymus enclosed within a Millipore filter into a neonatally thymectomized mouse, he was able to produce results similar to implanting an intact thymus directly. Furthermore, he demonstrated the immunologic competence of these mice, which were previously incompetent; thus demonstrating conclusively the presence of a non-cellular lymphocyte stimulating factor. Osoba (1963, 1965)

was able to confirm this reaction in mice, and Trench (1966) produced similar results in rabbits.

To date much of the basic information has been obtained from mice. The obvious question is whether similar results can be obtained for all species, or is the mouse the only animal which is capable of such a dramatic response. After the initial discovery of the wasting syndrome, new discoveries led to questioning the theory shared by Miller (1961a) and most others that the wasting syndrome was due to a form of self-immunity. McIntire (1964) and Wilson (1964) concluded that a latent infectious agent was responsible for the wasting and death of mice, because they were unable to demonstrate such a syndrome in germ-free animals. This in part would explain the relative absence of a wasting syndrome in other species.

Results in other species have been variable. Sherman (1963) reported a fatal wasting syndrome in hamsters occurring only in the males; however, females would undergo the wasting syndrome if oophorectomized and treated with testosterone. Balner (1966) recently reported a tendency for a more pronounced wasting in male mice than in females. Defendi (1964) demonstrated an early, atypical wasting syndrome in male and female hamsters in which losses and symptoms occurred only during the first 3 post-thymectomy weeks, after which

survivors appeared normal. One cannot help but wonder if the more extensive trauma of neonatal thymectomy as compared to the sham operation is responsible for these results.

Wasting syndrome in rats is rarely reported although the neonatally thymectomized rat will undergo some degree of immunologic depression of the delayed or cellular type, and a loss in ability to form humoral antibodies according to Arnason (1962a, 1962b, 1964) and Defendi (1964). Associated with this is a deficiency of small lymphocytes in the peripheral circulation and in the remaining lymphoid tissues. An analogous condition in the rabbit is reported by Archer (1962, 1964).

Thymectomy of newborn dogs was reported by Tilney (1965) to produce a classical wasting syndrome including loss of condition, lymphoid hypoplasia and reduction in gamma globulins. Although VandeWater (1964) observed no such syndrome, Tilney suggested that those who met with failure in attaining similar results perhaps did not remove the entire gland or left fragments behind which regenerated.

Chickens have been used almost as extensively as mice in thymus studies, and have played an important role in first developing the link between thymus-like organs and immunity. The chicken has both a cervical thymus and a thymic analog, the bursa of Fabricius.

This latter organ was reputed to function as everything from a storehouse for semen, secondary sex gland, or egg reservoir to an auxiliary urinary bladder, cloacal lubricator, or third cecum as Glick (1964) mentioned in his review. It was not until he and his associate Chang (1955) discovered immunologic suppression in neonatally bursectomized chickens that any plausible function of the bursa could be substantiated. Meyer (1959) performed hormonal thymectomies by injecting testosterone into the incubating egg on the fifth day and Mueller (1964) found the effect of this method comparable to postnatal surgical thymectomy. However, Warner (1962) found that by thymectomizing and bursectomizing the chick he was able to demonstrate a homograft immunity impairment which bursectomy alone did not alter.

No attempt to study neonatal thymectomy in calves has been reported. Papp (1960) reported a significant lymphopenia on calves thymectomized at approximately 3 weeks of age. Coleman (1964)* performed neonatal thymectomies in calves, but to date no followup report has been published.

---

*Coleman, Gerald L.; Personal communication. Pesticide Regulation Division, USDA, ARS, Washington, D.C. 20250

From data accumulated over the past few years current reviews such as Miller's (1965, 1966) and Good's (1966) favor the theory that the thymus is essential in the prenatal or neonatal period to produce small lymphocytes as precursors to form immunologic competent lymphoid centers throughout the body, and that lymphopoiesis, especially when related to immuno-capable forms, is regulated by a thymus reticular hormone throughout life. But as Good (1964) suggests there may be other functions of this long-mysterious gland.

Furth (1964) reported work implicating the thymus hormone directly in leukemogenesis, but further research in this aspect is needed. A more recent development has been Rieke's (1966) work reporting that lymphocytes in neonatally thymectomized rats contain a defective ribonucleic acid (RNA). In still another aspect of thymus function Pansky (1965) has reported finding an insulin-like substance produced by the thymus which he feels is responsible for post-thymectomy hypoglycemia.

Another particular line of research involving thymectomized animals has been in transmission studies of viral neoplasms. As previously cited there is a decrease in the incidence of leukemia following thymectomy; however, this decrease is attributed to a lack of thymus <u>per se</u> in mice, which is apparently necessary

as a primary nidus for infection. This has recently been substantiated by Haran-Ghera (1966) who found foci of potentially neoplastic cells in the thymus only one week following the direct injection of a leukemogenic virus into the gland, whereas no effect was encountered when the agent was injected into other lymphoid organs. After discovering the impairment of immunity following thymectomy Miller (1963, 1964a) indicated this tool may have other potential. Perry (1963) and Vandeputte (1963) were able to demonstrate a significant increase in tumors in thymectomized rats. Defendi (1964) observed a similar decrease in resistance to the polyoma virus in hamsters and mice. Grant (1965) reported an increase in sarcomata in mice resulting from injected carcinogens. Law (1966) found the thymus necessary in oncogenic immunity.

## MATERIALS AND METHODS

Ten newborn, colostrum-deprived Holstein bull calves were obtained from area dairy farms and were alternately selected and placed into either the thymectomy or control groups. Upon arrival at the clinic each calf was fed 2 quarts of a "standardized" colostrum and given a physical examination. The "standardized" colostrum was previously obtained from several cows, mixed, and frozen as Allen (1944), Hansen (1947), Jacobson (1951), and Morrison (1957) have recommended. The physical examination included: a rectal temperature reading, auscultation of the heart and lungs, observation for anomalies or physical defects, assessing the general condition, and examination of the mucous membranes. Following feeding and the physical examination, 10,000 units procaine penicillin G per pound of body weight was injected intramuscularly as a general prophylactic measure.

Surgical procedure

Several hours before commencing surgery, each animal was injected with a 0.01 mg./kg. intramuscular dose of 9-alpha-fluoroprednisolone acetate* as a prophylactic measure to allay surgical shock. Atropine was administered at a dose rate of 0.01 mg./kg. by intravenous injection to inhibit secretory activity and to suppress vagal activity since the surgery necessitated handling this nerve trunk. The left thoracic wall was clipped from the mid-abdominal region anteriorly over the shoulder, and from the sternum to the spinal column--using a fine clipper head.** The ventral half of the neck, from the sternum to the intermandibular space, was also clipped.

Calves were anesthetized while standing, using a mask on an open circuit gas anesthetic apparatus employing methoxyflurane*** and placed on a table in lateral recumbency. The mask was removed and the animal was intubated with a size 40, cuffed endotracheal tube. The surgical site was prepared for aseptic surgery. In

---

* Predef 2X, Upjohn, Kalamazoo, Michigan

** Oster Small Animal Clippers, Size 40 head, Milwaukee, Wisconsin

***Metofane, Pitman-Moore, Indianapolis, Indiana

securing the animal suitably for the surgical procedure, the left foreleg was extended over the animal's head, thus exposing the entire left thoracic wall. The other limbs were secured in their normal extended position.

The surgical procedure employed was a modification of Coleman's (1964).* The left thoracic area was covered with a sterile drape which permitted longitudinal exposure of the third rib. After controlling capillary bleeding in the incised skin and fascia, the incision was continued through all muscle layers down to the lateral surface of the rib. A 12 cm. segment of the third rib was then resected, from the costochondral junction dorsally, and entrance was made into the thoracic cavity. Using a Balfour retractor the opening was stretched transversly; thus, the anterior portion of the thoracic cavity was exposed from approximately the anterior aspect of the heart to a point just posterior to the thoracic inlet.

The left apical lobe of the lung was manipulated posteriorly thus exposing the thoracic portion of the thymus in the anterior mediastinum. A portion of the gland was picked up and by blunt dissection, freed from surrounding structures. Extreme care was necessary

---

*Coleman, Gerald L.; Personal communication. Pesticide Regulation Division, USDA, ARS, Washington, D.C. 20250

throughout the tedious dissection, especially when working next to the heart so as not to disrupt the pericardium, and when removing the more dorsal portions of the gland through which the vagosympathetic and phrenic nerve trunks coursed. Blood vessels of the gland, branches of the internal thoracic vessels and the anterior vena cava, were ligated. Blunt dissection was continued until the entire thoracic portion of the gland was removed.

Closure was effected with a continuous lock-stitch pattern using No. 1 chromic catgut. The stitch pattern did not encircle the periosteum of the resected rib, but rather passed through the doubled thickness of the anterior and posterior halves of the periosteum thus apposing the original incised edges. The suturing was started from each end of the incision and just prior to closure the lungs were inflated and the pleural cavity was evacuated. Muscle layers were closed individually with a similar lock-stitch pattern, and the skin was closed with simple interrupted sutures employing a non-capillary, synthetic suture material*.

An assistant now repositioned the animal in dorsal recumbency and prepared the ventral cervical surgical site. The area was again shrouded and a bold incision

---

*Vetafil, 0.4 mm., Bengen, West Germany, Dr. S. Jackson, Bethesda, Maryland 20014

was made on the midline from the mandibular rami to the thoracic inlet. Following incision of the underlying fascia the ventral muscles of the neck were bluntly separated. This exposed the complete cervical portion of the thymus. As with the thoracic portion of the thymus, the cervical portion was removed by blunt dissection. Difficulty was experienced in removing the small portions of the gland located dorsolaterally to the trachea adjacent to the submaxillary salivary glands because of the proximity to the thyroid gland and the carotid arteries, and because of its poorly accessible location and a strong fibrous attachment. Excised tissue from this area was later confirmed to be thymus by histologic examination. Muscles and fascia were apposed with a single continuous lock-stitch pattern and the skin was closed using material and techniques previously described. The sham operation on control animals included all of the procedures used in thymectomy with the exception of extirpation of the gland. Surgical procedure for removal of the entire thymus lasted from 3 to 3-1/2 hours.

Postsurgical care

Postsurgical care included treatment for surgical shock and hypothermia. Two hundred fifty milliliters of 5 % dextrose in 0.85 % physiological saline warmed to

body temperature and administered intravenously. For severely depressed individuals amphetamine sulfate* was injected subcutaneously and intravenously. Following return to a pen, the calf was covered, warmed with hot water bottles, and observed until able to stand unassisted, a period of time usually not exceeding 1 hour.

As a routine regimen, 10,000 units procaine penicillin G per pound of body weight was injected intramuscularly daily for a minimum of 10 days. Diarrhea, which frequently became a problem in both groups, was controlled by the oral administration of a neomycin-anticholinergic preparation.** Fever during the first few postoperative days was controlled with the aid of aspirin; 60 gr. b.i.d. the first day and 30 gr. b.i.d. the second day, administered per os. After the initial 10 day period, treatment was allayed unless absolutely necessary, in which case appropriate measures were taken.

---

* Amfetasul, Pitman-Moore, Indianapolis, Indiana

**Biosol-M, Upjohn, Kalamazoo, Michigan

All calves were fed a synthetic milk diet* following their first and only meal of colostrum. As they learned to eat a pelleted calf meal the diet was gradually changed to a pure skim milk milk-replacer. The initial feeding rate was 1/2 liter of reconstituted milk-replacer per 10 kg. body weight and an attempt was made to increase this level to 1/2 liter per 5 kg. body weight by 1 week of age. However, the initial level of feeding was reverted to whenever the first sign of enteric disorder appeared.

The calves were maintained on a skim milk diet until they consumed the pelleted feed at a rate of 1/2 kg. per day. Thereafter, according to the manufacturer's directions,** the calves were gradually phased off of the liquid diet and placed on a complete dry ration consisting of pellets and a ground grain mixture. Fresh, second-cutting hay and water were available at all times. At approximately 3 months of age the pellets were completely deleted from the grain ration.

---

* V-2 Vealer, Mutual Products Inc., Minneapolis, Minnesota

**Calf Manna, Albers Milling Company, Kansas City, Missouri

Hematologic determinations

At birth, and at weekly intervals for 20 weeks, 2 blood samples were taken from the 5 animals in each group. One 7 ml. sample was collected in a tube containing 0.05 ml. of 15 % tripotassium ethylene-diamine-eteracetic acid (EDTA) anticoagulant to be used for cell studies. The other 7 ml. sample was permitted to clot and the serum was separated and frozen at -7 C. for subsequent protein analyses.

Total erythrocyte and total leukocyte counts were performed on the Coulter Counter* using duplicate samples. Duplicate determinations of the packed cell volume (PCV) were performed using the microhematocrit method. Hemoglobin (Hb) was determined by the cyan-methemoglobin spectrophotometric method. Weight gains were assessed by biweekly weighings.

Animals were authanatized and necropsied at 20 weeks of age. The thymectomized animals were examined carefully for remnants or regeneration of thymus tissue. Any tissue which could possibly be considered thymus was submitted for histological examination. Specimens of liver, kidney, lung, spleen and various lymph nodes were

---

*Coulter Electronics, Model b, Hialeah, Florida

submitted for histologic examination along with any specific tissue showing gross lesions.

Total serum protein and serum albumin were determined by the "Improved" Biuret Method, Ferro-Ham modification (1955)* as discussed by Bray (1957). The albumin value was subtracted from the total protein value to determine the globulin value. The albumin: globulin (A:G) ratio was determined from these values.

The data obtained were processed employing trend analysis.

---

*Lab-trol Manual of Clinical Methods (1963) Dade Reagents, Inc., Miami, Florida

# RESULTS

Following the extensive procedure of surgical removal of the thymus, a state of shock ensued as evidenced by marked hypothermia (as low as 35 C.), rapid, weak pulse, and a comatose condition. Since operative hemorrhage was minimal, shock was judged to be the result of a long period of general anesthesia (up to 5 hours) and trauma inherent to removal of tissue from a large area. Postoperative recovery was marked by hyperthermia, often as high as 43 C., which persisted for at least 2 weeks despite antibiotic therapy.

Calves in both the thymectomy and control groups were frequently troubled with an enteritis during the initial 4 to 6 weeks during which time their diet was primarily liquid. At the first indication of an enteric disorder, the amount of liquid diet was decreased by one-half and appropriate antimicrobial and palliative measures were instituted. Of the bacterial organisms isolated from fecal specimens, Escherichia coli was the only one that could have been incriminated as an etiological agent, with the exception of the isolation

of Salmonella sp. from one calf. Rarely was the antibiotic of choice (as indicated by sensitivity tests) effective in controlling diarrhea.

Between the ages of 4 to 6 weeks the calves began to gain weight rapidly and did not exhibit any further clinical illnesses.

## Hematologic findings

The data were processed employing trend analysis. When the thymectomized calves were compared with the non-thymectomized control calves, no significant differences were noted for total erythrocyte counts (Appendix, Table 1), the hemoglobin values (Appendix, Table 2), the packed cell volumes (Appendix, Table 3), or the total leukocyte counts (Appendix, Table 4).

### Differential leukocyte counts

Lymphocytes. No significant difference was noted between the 2 groups for absolute lymphocyte counts. However, the thymectomized group had a considerably lower relative lymphocyte count. Analysis showed that the mean values for the 2 groups throughout the 19 week period (factor B) approached a significant difference, with an approximate probability of F statistic (i.e. the

probability this situation would occur by chance) of 0.06. (Appendix, Tables 5a and b.)

Neutrophils. No significant difference was noted between the 2 groups for either absolute or relative neutrophil counts. (Appendix, Tables 6a and b.)

Moncytes. No significant difference was noted between the 2 groups for either absolute or relative monocyte counts. (Appendix, Tables 7a and b.)

Eosinophils. No significant difference was noted between the 2 groups for absolute eosinophil count. However, the thymectomized group exhibited a relative eosinophilia. The means averaged throughout the 19 week period (factor B) approached a significant difference with an approximate probability of F statistic of 0.08. (Appendix, Tables 8a and b.)

Basophils. A significant basophilia, both absolute and relative was detected for calves in the thymectomized group. The average mean values throughout the 19 week period (factor B) were significantly different at the $P < 0.01$ level with an approximate probability of F statistic of 0.00 for the absolute values and 0.01 for the relative values. (Appendix, Tables 9a and b.)

The following hematologic determinations were performed on the serum collected at weekly intervals:

Total protein

Thymectomized calves had a higher total serum protein level throughout the 19 week period (factor B) which was significant at the $P < 0.01$ level. The approximate probability of F statistic was 0.00. (Appendix, Table 10.)

Serum albumin

Thymectomized calves had a lower serum albumin level throughout the 19 week period (factor B) which was significant at the $P < 0.01$ level. The approximate probability of F statistic was 0.00. (Appendix, Table 11.)

Serum globulin

Thymectomized calves had a higher serum globulin level throughout the 19 week period (factor B) which was significant at the $P < 0.01$ level. The approximate probability of F statistic was 0.00. (Appendix, Table 12.)

Albumin:globulin ratio

Thymectomized calves had a lower albumin:globulin ratio throughout the 19 week period (factor B) which was

significant at the $P < 0.01$ level. The approximate probability of F statistic was 0.00. (Appendix, Table 13.)

Weight gains

Weight gains were assessed by a biweekly weighing. Cumulative gains, using birth weight as base zero were employed to equilibrate gains regardless of initial weight.

Thymectomized calves gained significantly less weight throughout the 19 week period (factor B) which was significant at the $P < 0.01$ level. The approximate probability of F statistic was 0.00. (Appendix, Table 14.) The control calves had a mean weight of 27.6 kg. or 1.8 kg. more than the thymectomized calves which had a mean weight of 25.8 kg.

Due to lack of foresight the calves were all weighed at two week intervals rather than weighing each calf every other week of his life starting from birth. This resulted in an alternate week arrangement of data for the different calves so that in any given week of their lives actual weights were available for only some of the individuals. Therefore, when the data were selected for analysis they were based upon a two week weighing period and the figures presented in Table 14

would not be the actual average weight gains for every other week of all calves' lives. However, a sum of weights which was projected by the analysis with a mean net gain of 64.4 kg. for the thymectomized group and 69.0 kg. for the control group compared almost exactly with a mean of the actual final weights.

All calves with the exception of T4 and C1 had weight losses as great as 5 kg. during the first few weeks after birth. At about 4 to 6 weeks of age all calves began a rapid growth period.

Necropsy: Gross and histologic observations

Gross pathological findings were minimal. The terminal portion of one ureter in thymectomized calf T3 was involved with a chronic inflamatory process. In 2 of the thymectomized calves, T1 and T4, small remnants of tissue were removed from the postmandibular area which were later histologically confirmed as thymus tissue. In all other thymectomized calves careful examination revealed no traces of thymus tissue.

Histologically the lungs showed the most frequent, although minor, pathologic involvement. Calves C2, C3, C4, T3, T4, and T5 had various pneumonic involvements.

Renal disorders included glomerular atrophy in calf T2, glomerulitis in calf C5, and accumulations of

lymphocytes in various areas of the kidneys in calves C1, C4, C5, and T1.

Splenic involvement included decreased lymphoblastic activity in calf T1, and neutrophil infiltration of the red pulp in calves C3, T1, T3, T4, and T5.

The liver in calf T4 had neutrophilic infiltration in the hepatic sinusoids and lymphocytic infiltration in the portal triads.

Lymph node involvement was minor and varied coincidental with the pathology mentioned above. However, of pertinent, general interest was the mild reactivity of the lymph nodes in calves T1 and T5; the hypoplasia of the germinal centers of the lymph nodes of calf T2; the eosinophil infiltration of the lymph nodes of calves C2, T3, and T4; and the neutrophil infiltration of the lymph nodes of calves C1 and T3.

# DISCUSSION

The problem of rearing calves, subjected to extensive surgery within 24 hours after birth, and deprived of colostrum except for an initial hand feeding is difficult. Numerous experiments have been conducted which point out the paramount necessity of feeding colostrum. Smith (1948), Morrison (1957), and Ingram (1958) state that colostrum is absolutely necessary for proper antibody protection as well as for nutrients necessary for growth. Davis (1962) adds that the calf has practically no antibodies during the first 48 hours of life. Smith (1922) was probably one of the first to run a controlled experiment on colostrum feeding. He found that two-thirds of the group deprived of colostrum died within the first few weeks of life while all the colostrum fed control animals lived. Hansen (1947) demonstrated immediate increases in serum globulins following colostrum ingestion. Miles' (1948) work supported Smith's earlier study. He also noted the runted condition of colostrum deprived calves which survived. Recent work by Jacobson (1951) and Gay (1965), further support the importance of colostrum during the first few days of neonatal life.

Sweat (1965) derived calves by hysterectomy and raised them in isolation without colostrum. His initial experiment failed when the animals died of a coliform septicemia after changing from a sterile diet to milk replacer on the third day. Further attempts, maintaining the animals on sterile diets, did prove more successful.

Despite the new, clean facilities in which these experimental animals were housed and the sanitary technique used in food handling, every calf suffered repeated attacks of enteritis and consequential systemic sequelae. Antibiotics, anticholinergic drugs, and intravenous fluids were employed as needed to maintain the calves, but these measures seemed to be of only temporary value. Enteritis would soon reappear following cessation of medication. It is the opinion of this author that without the antibodies which a calf can apparently obtain only from ingestion of colostrum early in life, antimicrobial and symptomatic treatment are of only palliative value.

From the analyses of data it is apparent that thymectomy of neonatal calves has no effect on their erythropoietic capabilities.

Likewise, total leukocyte counts did not differ significantly between groups. A mean of 9,169 leukocytes/cmm., falls well within the normal ranges

established for cattle, although no specific references were available for Holstein cattle during early life.

An analysis of the leukocyte differential does show a statistically significant difference between groups. Lymphopenia has been one of the most characteristic findings of the wasting syndrome as Miller (1961a) and others have demonstrated. In this experiment, however, no significant differences were noted between groups although the thymectomized calves had a considerably lower relative percentage of lymphocytes which approached statistical significance at the $P < 0.05$ level. In the concept of trend analysis, had more animals or a longer period of time been employed, a significant difference between the 2 groups may have been noted.

No significant difference was noted between the 2 groups for either absolute or relative neutrophil counts. Both groups had extreme variance which ranged from a low of 180 neutrophils/cmm. which constituted 5 % of the leukocytes for one calf to a high of 29,050 neutrophils/cmm. which constituted 89 % of the leukocytes for another calf. Since individuals of both groups displayed similar variation which could be roughly correlated to attacks of enteritis, the author concludes that the variability seen is proportional to the challenge presented to the individual.

The reason for a basophilia in the thymectomized group is not readily apparent. Schalm (1961) mentions that the function of the basophil is very obscure. It is believed they are related to tissue mast cells since the granules of both contain heparin which indicated an anticoagulant activity. Guyton (1962) comments that basophil numbers increase slightly during the healing phase of inflammation as well as during chronic inflammatory processes. Benjamin (1964) states that a basophilia often occurs concurrently with an eosinophilia.

The normal mean value for basophil count for cattle is 0.5 % or 40 cells/cmm., according to Schalm (1961). These values coincide with those presented by Benjamin (1964). The mean value for the control group is considerably above this at 0.9 % or 74 cells/cmm., and the thymectomized group had a mean value significantly higher than this at 1.2 % or 101 cells/cmm. No reports were found in which other research workers found a basophilia following thymectomy.

Serum protein values were significantly different between the 2 groups. Because of an extremely small amount of variation among calves from week to week, the thymectomized group has a significantly higher total protein level at 5.25 gm./100 ml. vs. 5.23 gm./100 ml. for the control group; a lower albumin level of 3.18 gm./100 ml. vs. 3.25 gm./100 ml.; a higher globulin level

of 2.07 gm./100 ml. vs. 1.98 gm./100 ml.; and a lower A:G ratio of 1.61 vs. 1.74. The finding of hypoglobulinemia in some thymectomized species was not observed in this work. Hypoalbuminemia and a low A:G ratio, on the other hand, is typical of debilitated or cachechtic animals.

These values for serum proteins are considerably different from those put forth by Dukes (1955). The values he gives for cattle are: 7.60 gm./100 ml. total protein, 3.63 gm./100 ml. albumin, 3.97 gm./100 ml. globulin, and 0.91 A:G ratio. None of the available literature gave age, sex, or environmental differences for serum protein values in cattle.

The initial fluctuations in the weight of the calves were judged to be a result of colostrum deprivation and the neonatal surgery. Calves in both groups experienced similar weight losses which persisted up to 3 weeks in some instances. Since no abrupt changes were made in the diet and each calf was individually phased off the liquid diet as he consumed more of the dry ration, the lack of substantial weight gains prior to 5 or 6 weeks of age was judged to be in part due to factors other than nutrition alone.

Smith (1965) and Ingram (1965) state that depending on the type of antigen, calves can develop few

antibodies prior to 1 month of age. Prior to this age the calves in this experiment frequently had enteritis and suffered from sequential anorexia. However, at the age of 4 to 5 weeks these calves ceased having enteric disorders, and appetites and gains increased concomitantly. In the judgement of this author the inability of these calves to combat infectious diseases during the first month of life was responsible in part for the lack of good general health and weight gains.

Of particular interest in necropsy findings was the fact that 2 of the thymectomized calves retained small amounts of thymus tissue. The amount present was judged to be minimal considering the mass of thymus tissue removed from these calves. Whether remnants of thymus tissue can profoundly affect the results of this experiment cannot be accurately assessed. Tilney (1965) suggested that subtotal thymectomy in dogs essentially negated the entire effect; however, other workers have been unable to duplicate Tilney's result despite scrutinous extirpation of the thymus of dogs. Although accurate statistical analysis is not feasible, observation of the data does not reveal any noticeable difference between calves T1 and T4 and the rest of the thymectomized group.

Histopathologic findings suggest no difference between the 2 groups. Both appeared to be equally

afflicted with various pneumonic or renal pathologic processes. However, of interest is the preponderance of reticuloendothelial disorders in the thymectomized calves. Here again the diversity of abnormalities observed obviates drawing any specific conclusion. Since Miller (1961a) and others have observed depletion of lymphocytes from lymphoid organs in thymectomized animals, the author feels it is pertinent to point out only that these abnormalities are almost exclusively limited to the thymectomized group.

Immunologic studies were beyond the scope of this experiment and empirical observations of the general health of the calves can give only a crude estimate of the immunologic capabilities of thymectomized calves. The lower relative lymphocyte and eosinophil values could be of importance, however, when their place in current immunologic theories is observed.

Whether thymectomized calves will be of value in virus or neoplasm transmission studies remains to be explored. Theilen (1965) and Dungworth (1964) reported thymus involvement in the calf form of bovine leukemia. This could mean the thymus contains necessary factors in the evolution of this disease as Miller (1961b) pointed out in his work with mice. On the other hand, Siegler (1964) reported that the mouse form of leukemia

is primarily of thymic origin, whereas, Dungworth (1964) and Theilen (1965) did not find thymus involvement in all cases of leukemia in calves.

# CONCLUSIONS

Neonatal thymectomy in calves is a feasible surgical operation. Raising calves with only a small initial feeding of colostrum is difficult and is probably the main factor resulting in frequent enteritis and general unthriftiness.

Neonatal thymectomy has no observable effect on the erythropoietic system.

While not producing an observable effect on the total leukocyte count, neonatal thymectomy altered the leukocyte differential. A highly significant basophilia, both relative and absolute, occurred in the thymectomized animals. A tendency toward an eosinophilia and a lymphopenia was also observed in the thymectomized group.

Neonatal thymectomy produced a highly significant difference in serum proteins. The thymectomized group had higher total proteins and serum globulin level, and a lower serum albumin level and A:G ratio.

Neonatal thymectomy resulted in reduced weight gains which were highly significant.

## SUMMARY

During the past decade the lack of immune competence produced in various species of laboratory animals by neonatal thymectomy has been repeatedly demonstrated. This experiment was conducted to explore the feasibility of such an operation in calves and to study some of the basic effects upon the animals with the possibility of using this procedure for creating animals of lowered immunologic capabilities for use in certain viral or neoplastic disease transmission studies.

Ten newborn Holstein-Friesian bull calves were alternately selected and placed into either the thymectomy or control group. Each calf was given an initial 2 liter hand feeding of colostrum which was previously obtained from several cows, mixed and frozen. Within 24 hours after birth the calves were thymectomized or subjected to a sham operation. The surgical procedure involved resectioning of the central portion of the third left rib and bluntly dissecting the thoracic portion from its anterior mediastinal position; then placing the calf on his back and bluntly dissecting the cervical portion through a midventral

cervical incision. The sham operation was identical with the exception of the extirpation of the gland.

Postsurgical care included appropriate antimicrobial and palliative treatment. The calves were fed an aseptically prepared milk replacer, a pelleted dry ration, hay, and water. During the first month of age all calves were troubled with frequent attacks of enteritis with ensuing loss of condition and anorexia. From about the fourth week until the termination of the experiment at 20 weeks of age no clinical disease entities were observed in either group.

From the weekly blood samples, no significant difference between groups for erythrocyte count, hemoglobin level, packed cell volume, total leukocyte count, and monocyte or neutrophil values of the leukocyte differential was detected. However, highly significant values ($P < 0.01$) for higher total serum protein and serum globulin, and lower serum albumin and albumin:globulin ratio were detected for the thymectomized group, as were highly significant lower weight gains. The thymectomized group also had lower relative eosinophil and lymphocyte values which approach significance at the $P < 0.05$ level.

## LITERATURE CITED


1. Allen, N.N.: The Use of Stored Colostrum to Replace Marketable Milk for Calf Feeding. J. Dairy Sci., 27, (August, 1944): 652-653.

2. Andrew, Warren: Textbook of Comparative Histology. Oxford University Press, New York, N.Y., 1959, 349-350.

3. Archer, O. and Pierce, J.C.: The Role of the Thymus in Development of the Immune Response. Fed. Proc., 20, (1961): 26.

4. Archer, O., Pierce, J.C., Papermaster, B.W., and Good, R.A.: Reduced AB Responses in Thymectomized Rabbits. Nature, 195, (July, 1962): 191-193.

5. Archer, O., Papermaster, B.W., and Good, R.A.: Thymectomy of Rabbit and Mouse; Consideration of Time of Lymphoid Peripheralization. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 414-435.

6. Arnason, B.G., Jancović, B.D., Waksman, B.H., and Wennersten, C.: Role of the Thymus in Immune Reactions in Rats: II. Supressive Effect of Thymectomy at Birth on Reactions of Delayed (Cellular) Hypersensitivity and the Circulating Lymphocyte. J. Exptl. Med., 116, (1962a): 177.

7. Arnason, B.G., Jancović, B.D., and Waksman, B.H.: A Survey of the Thymus and Its Relation to Lymphocytes and Immune Reactions. Blood, 20, (November, 1962b): 617-628.

8. Arnason, B.G., Jancović, B.D., and Waksman, B.H.: The Role of the Thymus in Immune Reactions in Rats. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 492-503.

9. Balner, H. and Dersjant, H.: Sex Difference for Immune Depression and Runting in Neonatally Thymectomized Mice. Nature, 209, (February, 1966): 815-816.

10. Benjamin, M.M.: Veterinary Clinical Pathology. 2nd ed. The Iowa State University Press, Ames, Iowa, 1964.

11. Bloom, W. and Fawcett, D.W.: A Textbook of Histology. 8th ed. W.B. Saunders Company, Philadelphia, Pa., 1962, 317-323.

12. Bray, W.E.: Clinical Laboratory Methods. 5th ed. The C.V. Mosby Company, St. Louis, Mo., 1957, 325-327.

13. Burnet, F.M. and Holmes, M.C.: Thymic Lesions in Auto-Immune Disease of Mice. Nature, 194, (1962a): 146.

14. Burnet, Sir Macfarlane: Role of the Thymus and Related Organs in Immunity. Brit. Med. J., 2, (September, 1962b): 807-811.

15. Chang, T.S., Glick, B., and Winter, A.R.: The Significance of the Bursa of Fabricius of Chickens in Antibody Production. Poultry Sci., 34, (1955): 1187.

16. Copenhaver, W.M.: Bailey's Textbook of Histology. 15th ed. The Williams and Wilkins Co., Baltimore, Md., 1964, 294-298.

17. Davis, R.F.: Modern Dairy Cattle Management. Prentice-Hall Inc., Englewood Cliffs, New Jersey, 1962, 104.

18. Defendi, V. and Roosa, R.A.: Effect of Thymectomy on Tumor Induction by Polyoma Virus and Polyoma-Induced Tumor Cells. Fed. Proc., 23, (1964): 393.

19. Dukes, H.H.: The Physiology of Domestic Animals. 7th ed. Comstock Publishing Associates, Ithaca, N.Y., 1955, 50 and 87.

20. Dungworth, D.L., Theilen, G.H., and Lengyel, J.: Bovine Lymphosarcoma in California: II The Thymic Form. Path. Vet., 1, (1964): 323-350.

21. Fisher, E.R.: Pathology of the Thumus and Its Relation to Human Disease. The Thymus in Immunobiology. Edited by Good Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 676-730.

22. Frandson, R.D.: Anatomy and Physiology of Farm Animals. Lea and Febiger, Philadelphia, Pa., 1965, 435.

23. Furth, J., Okani, H., and Kunii, A.: Role of the Thymus in Leukemogenesis. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 595-622.

24. Gay, C.C., Anderson, N., Fisher, E.W., and McEvan, A.D.: Gamma Globulin Levels and Neonatal Mortality in Market Calves. Vet. Rec., 77, (1965): 148-149.

25. Glick, B.: Growth and Function of the Bursa of Fabricius. Poultry Sci., 34, (1955): 1196.

26. Glick, B., Chang, T.S., and Jaap, R.G.: The Bursa of Fabricius and Antibody Production. Poultry Sci., 35, (1956): 224.

27. Glick, B.: The Bursa of Fabricius and the Development of Immunologic Competence. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 343-358.

28. Good, R.A., Martinez, C., and Gabrielsen, A.E.: Clinical Considerations of the Thymus in Immunology. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 1-22.

29. Good, R.A., Cooper, M.D., Peterson, R.D.A., Kellum, M.J., Sutherland, D.E.R., and Gabrielsen, A.E.: The Role of the Thymus in Immune Process. Ann. N.Y. Acad. Sci., 135, (1966): 451-478.

30. Grant, G.A. and Miller, J.F.A.P.: Effect of Neonatal Thymectomy on the Induction of Sarcomata in C57BL Mice. Nature, 205, (March, 1965): 1124-1125.

31. Guyton, A.C.: Textbook of Medical Physiology. 2nd ed. W.B. Saunders Company, Philadelphia, Pa., 1962, 160-173.

32. Hammar, J.A.: The New Views as to the Morphology of the Thymus Gland and Their Bearing on the Problem of the Function of the Thymus. Endocrinol., 5, (1921): 543-572 and 731-760.

33. Hansen, R.G. and Phillips, P.H.: Studies on Proteins From Bovine Colostrum. I. Electrophoretic Studies on the Blood Serum Proteins of Colostrum-free Calves and of Calves Fed Colostrum at Various Ages. J. of Biol. Chem., 171, (November, 1947): 223.

34. Haran-Ghera, Nechama, Lieberman, Miriam, and Kaplan, Henry S.: Direct Action of a Leukemogenic Virus on the Thymus. Cancer Res., 26, (March, 1966): 438-441.

35. Ingram, P.L., Shillam, K.W.G., Hawkins, G.M., and Roy, J.H.B.: The Nutritive Value of Colostrum for the Calf. Brit. J. Nutrition, 12, (1958): 203-215.

36. Ingram, D.G. and Smith, A.W.: Immunologic Responses of Young Animals. Can. Vet. J., 6, (1965): 194-204.

37. Jacobson, W.C., Converse, H.T., Wiseman, H.G., and Moore, L.A.: The Effect of Substituting Colostrum for Whole Milk in the Ration of Dairy Calves. J. Dairy Sci., 34, (1951): 905-910.

38. Jancović, B.D., Waksman, B.H., and Arnason, B.G.: Role of the Thymus in Immune Reactions in Rats. J. Exptl. Med., 116, (1962): 159-206.

39. Kunii, A., Cali, A., and Furth, J.: Effect of Neonatal Thymectomy on Induction of Leukemia by Virus in Rats. Proc. Soc. Biol. and Med., 118, (1965): 815-818.

40. Law, Lloyd W. Studies of Thymic Function with Emphasis on the Role of the Thymus in Oncogenesis. Cancer Res., 26, (April, 1966): 551-574.

41.

42.

43

4

4

41. Levey, R.H., Trainin, N., and Law, L.W.: Evidence for Function of Thymic Tissue in Diffusion Chambers Implanted in Neonatally Thymectomized Mice. Preliminary Report. J. Nat. Cancer Inst., 3, (1963): 199-217.

42. Martinez, C., Dalmasso, A.P., and Good, R.A.: Acceptance of Tumor Homografts by Thymectomized Mice. Nature, 194, (1962a): 1289-1290.

43. Martinez, C., Kersey, J., Papermaster, B.W., and Good, R.A.: Skin Homograft Survival in Thymectomized Mice. Proc. Soc. Exptl. Biol. and Med., 109, (1962b): 193.

44. Martinez, C., Dalmasso, A.P., and Good, R.A.: Effect of Thymectomy on Development of Immunological Competence in Mice. Ann. N.Y. Acad. Sci., 113, (1964a): 933-945.

45. Martinez, C., Dalmasso, A.P., and Good, R.A.: Homotransplantation of Normal and Neoplastic Tissue in Thymectomized Mice. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964b, 465-477.

46. Maximow, A.A. and Bloom, W.: A Textbook of Histology. 7th ed. W.B. Saunders Company, Philadelphia, Pa., 1957.

47. McIntire, K.R., Sell, S. and Miller, J.F.A.P.: Pathogenesis of the Post-neonatal Thymectomy Wasting Syndrome. Nature, 204, (1964): 151-155.

48. Mcleod, W.M.: Bovine Anatomy. 2nd ed. Burgess Publishing Co., Minneapolis, Minn., 1958, 99.

49. Metcalf, D.: The Thymic Origin of the Plasma Lymphocyte Stimulating Factor. Brit. J. Cancer, 10, (1956): 442-457.

50. Meyer, R.K., Rao, M.A., and Aspinall, R.L.: Inhibition of the Development of the Bursa of Fabricius in Embryos of the Common Fowl by 19-nortestosterone. Endocrinol., 64, (1959): 890.

51. Miles, J.T., Hinton, S.A., and Patrick, H.: Raising Dairy Calves Without Colostrum. J. Dairy Sci., 31, (August, 1948): 67.

52.

53.

54.

55

5

5

52. Miller, J.F.A.P.: Immunological Function of the Thymus. Lancet, 2, (1961a): 748-749.

53. Miller, J.F.A.P.: Analysis of the Thymus Influence in Leukaemogensis. Nature, 191, (1961b): 248.

54. Miller, J.F.A.P.: Immunological Significance of the Thymus of the Adult Mouse. Nature, 195, (September, 1962a): 1318.

55. Miller, J.F.A.P.: Role of the Thymus in Transplantation Immunity. Ann. N.Y. Acad. Sci., 99, (October, 1962b): 340-354.

56. Miller, J.F.A.P.: Role of Thymus Immunity. Brit. Med. J., 3, (August, 1963): 459-461.

57. Miller, J.F.A.P.: Effect of Thymic Ablation and Replacement. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964a, 436-464.

58. Miller, J.F.A.P.: The Thymus and the Development of Immunologic Responsiveness. Science, 144, (1964b): 1544-1551.

59. Miller, J.F.A.P.: The Thymus and Transplantation Immunity. Brit. Med. Bull., 21, (1965): 111-117.

60. Miller, J.F.A.P.: Immunity in the Foetus and the New-born. Brit. Med. Bull., 22, (1966): 21-36.

61. Morrison, F.B.: Feeds and Feeding. Morrison Publishing Co., Ithaca, N.Y., 1957, 174 and 665.

62. Mueller, A.P., Wolfe, H.R. and Cole, W.P.: Antibody Studies in Hormonally and Surgically Bursectomized Chickens. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 359-375.

63. Osoba, D. and Miller, J.F.A.P.: Evidence of a Humoral Thymus Factor for the Maturation of Immuno Faculty. Nature, 199, (August, 1963): 653.

64. Osoba, D.: The Effects of Thymus and Other Lymphoid Organs Enclosed in Millipore Diffusion Chambers on Neonatally Thymectomized Mice. J. Exptl. Med., 122, (1965): 633-650.

65.

66.

67.

68

69

7

65. Pansky, B., House, E.L., and Cone, L.A.: An Insulin-like Thymic Factor. Diabetes, 14, (1965): 325-332.

66. Papp, Eugene: Effects of Thymectomy in Rats and Calves. Ph.D. Thesis. Ohio State University, Columbus, Ohio, 1960.

67. Parrott, Delphine M.V.: Strain Variation Mortality and Runt Disease in Mice Thymectomized at Birth. Trans. Bull., 29, (1962a): 102.

68. Parrott, Delphine M.V. and East, June: Role of Thymus in Neonatal Life. Nature, 195, (July, 1962b): 347-348.

69. Perry, G.C., Faulk, M., Shapiro, E., Mellors, J., and Money, W.L.: Function of the Thymus and Growth of Tumor Homograft. Nature, 200, (December, 1963): 1294.

70. Rieke, William O.: Lymphocytes from Thymectomized Rats: Immunologic, Proliferative and Metabolic Properties. Science, 152, (April, 1966): 535-538.

71. Roberts, S.J.: Veterinary Obstetrics and Genital Diseases. Stephen J. Roberts, Publishers, Ithaca, N.Y., 1956, 253.

72. Ruth, R.F., Allen, C.P., and Wolfe, H.R.: The Effect of Thymus on Lymphoid Tissue. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 183-206.

73. Schalm, O.W.: Veterinary Hematology. Lea and Febiger, Philadelphia, Pa., 1961.

74. Sherman, J.D. and Dameshek, W.: "Wasting Disease" Following Thymectomy in the Hamster. Nature, 197, (1963): 469-471.

75. Sherman, J.D. and Dameshek, W.: Post Thymectomy Wasting Disease of the Golden Hamster. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 542-550.

76. Siegler, R.: The Morphology of the Thymuses and Their Relation to Leukemia. The Thymus in Immunobiology. Edited by Good and Gabrielsen. Hoeber Medical Div'n, Harper and Row, Publishers, New York, N.Y., 1964, 623-655.

77. Sisson, S. and Grossman, J.D.: The Anatomy of the Domestic Animals. 4th ed. W.B. Saunders Company, Philadelphia, Pa., 1962, 554-555.

78. Smith, A.N. and Ingram, D.G.: Immunological Responses of Young Animals: II Antibody Production in Calves. Can. Vet. J., 6, (1965): 226-232.

79. Smith, E.L.: The Isolation and Properties of the Immune Proteins of Bovine Milk and Colostrum and Their Role in Immunity: A Review. J. Dairy Sci., 31, (February, 1948): 127.

80. Smith, Theobald and Little, Ralph B.: Significance of Colostrum to the New-born Calf. J. Exptl. Med., 36, (August, 1922).

81. Sweat, R.L. and Dunn, J.W.: Hysterectomy-derived, Colostrum-deprived, Isolation-raised Calves for Disease Research. Am. J. Vet. Res., 26, (November, 1965): 1309.

82. Theilen, G.H. and Dungworth, D.L.: Bovine Lymphosarcoma in California. III The Calf Form. Am. J. Vet. Res., 26, (May, 1965): 696-709.

83. Tilney, N.L., Beatie, E.J. Jr., and Economou, S.G.: The Effect of Neonatal Thymectomy in the Dog. J. Surg. Res., 5, (1965): 23-30.

84. Trautmann, A. and Fiebiger, J.: Fundamentals of the Histology of Domestic Animals. Comstock Publishing Associates, Ithaca, N.Y., 1957, 141-142.

85. Trench, C.A.H., Watson, J.W., Walker, F.C., Gardner, P.S., Green, C.A.: Evidence for a Humoral Thymic Factor in Rabbits. Immunol., 10, (February, 1966): 187-191.

86. Turner, C.D.: General Endocrinology. 2nd ed. W.B. Saunders Co., Philadelphia, Pa., 1955, 444-445.

87. Vandeputte, M., Denys, P. Jr., Leyten, R., and DeSomer, P.: The Oncogenic Activity of the Polyoma Virus in Thymectomized Rats. Life Sci., 7, (1963): 475-478.

88. VandeWater, J.M. and Katzman, H.: Studies of the Immune Mechanisms in Thymectomized Pups. J. Surg. Res., 4, (1964): 387-390.

89. Warner, N.L. and Szenberg, A.: Effect of Neonatal Thymectomy on the Immune Response in the Chicken. Nature, 196, (1962): 784-785.

90. Wilson, R., Sjodin, K., and Bealmear, M.: The Absence of Wasting in Thymectomized Germfree (Axenic) Mice. Proc. Soc. Exptl. Biol. and Med., 117, (1964): 237-239.

# APPENDIX

TABL

Ove

In
fo

W
e

TABLE 1. Erythrocyte counts for thymectomized and control calves (millions/cmm.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 8.61 | | | 8.55 |
| Individual means for 19 weeks | | T1 | 9.93 | | C1 | 10.41 |
| | | T2 | 11.10 | | C2 | 9.54 |
| | | T3 | 7.06 | | C3 | 7.51 |
| | | T4 | 7.81 | | C4 | 7.79 |
| | | T5 | 7.15 | | C5 | 7.50 |
| Weekly means for each group | week | 2 | 7.85 | week | 2 | 8.59 |
| | | 3 | 7.82 | | 3 | 8.64 |
| | | 4 | 8.44 | | 4 | 7.64 |
| | | 5 | 8.01 | | 5 | 8.42 |
| | | 6 | 8.61 | | 6 | 8.88 |
| | | 7 | 8.53 | | 7 | 8.60 |
| | | 8 | 8.57 | | 8 | 8.04 |
| | | 9 | 8.74 | | 9 | 8.90 |
| | | 10 | 8.43 | | 10 | 8.48 |
| | | 11 | 8.30 | | 11 | 8.55 |
| | | 12 | 8.61 | | 12 | 8.38 |
| | | 13 | 8.87 | | 13 | 8.77 |
| | | 14 | 8.47 | | 14 | 8.56 |
| | | 15 | 9.21 | | 15 | 8.49 |
| | | 16 | 9.28 | | 16 | 9.15 |
| | | 17 | 9.12 | | 17 | 8.82 |
| | | 18 | 9.04 | | 18 | 9.13 |
| | | 19 | 9.12 | | 19 | 8.23 |
| | | 20 | 8.59 | | 20 | 8.17 |

Analysis of variance table for erythrocyte values

| Source of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.179 | 1 | 0.179 | 0.004 | 0.91 |
| Error (a) | 386.015 | 8 | 48.252 | | |
| B** | 18.759 | 18 | 1.042 | 1.269 | 0.22 |
| AB*** | 10.246 | 18 | 0.569 | 0.693 | 0.81 |
| Remaining error | 118.226 | 144 | 0.821 | | |
| Total | 533.425 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 2. Hemoglobin values for thymectomized and control calves (gm./100 ml.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 10.49 | | | 10.40 |
| Individual means for 19 weeks | | T1 | 11.36 | | C1 | 11.64 |
| | | T2 | 12.70 | | C2 | 11.63 |
| | | T3 | 8.93 | | C3 | 9.30 |
| | | T4 | 10.01 | | C4 | 9.86 |
| | | T5 | 9.43 | | C5 | 9.55 |
| Weekly means for each group | week | 2 | 10.70 | week | 2 | 11.82 |
| | | 3 | 10.98 | | 3 | 10.96 |
| | | 4 | 10.24 | | 4 | 10.48 |
| | | 5 | 9.98 | | 5 | 10.18 |
| | | 6 | 10.34 | | 6 | 10.02 |
| | | 7 | 9.82 | | 7 | 9.96 |
| | | 8 | 10.18 | | 8 | 9.80 |
| | | 9 | 10.84 | | 9 | 10.50 |
| | | 10 | 10.66 | | 10 | 10.38 |
| | | 11 | 10.22 | | 11 | 10.36 |
| | | 12 | 10.28 | | 12 | 10.08 |
| | | 13 | 10.62 | | 13 | 10.56 |
| | | 14 | 10.50 | | 14 | 10.14 |
| | | 15 | 10.60 | | 15 | 10.36 |
| | | 16 | 10.92 | | 16 | 10.76 |
| | | 17 | 10.68 | | 17 | 10.40 |
| | | 18 | 10.64 | | 18 | 10.10 |
| | | 19 | 10.88 | | 19 | 10.58 |
| | | 20 | 10.16 | | 20 | 10.10 |

Analysis of variance table for hemoglobin values

| Source of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.380 | 1 | 0.380 | 0.011 | 0.88 |
| Error (a) | 279.541 | 8 | 34.943 | | |
| B** | 22.048 | 18 | 1.225 | 1.068 | 0.39 |
| AB*** | 6.001 | 18 | 0.333 | 0.291 | 1.00 |
| Remaining error | 165.131 | 144 | 1.147 | | |
| Total | 473.101 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TAB

Ove

In
fo

We
e

TABLE 3. Packed cell volume for thymectomized and control calves (%)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 31.72 | | | 30.93 |
| Individual means for 19 weeks | | T1 | 35.09 | | C1 | 34.07 |
| | | T2 | 38.63 | | C2 | 33.88 |
| | | T3 | 27.05 | | C3 | 28.09 |
| | | T4 | 29.49 | | C4 | 29.83 |
| | | T5 | 28.32 | | C5 | 28.79 |
| Weekly means for each group | week | 2 | 34.04 | week | 2 | 36.20 |
| | | 3 | 35.56 | | 3 | 33.80 |
| | | 4 | 31.70 | | 4 | 32.60 |
| | | 5 | 30.84 | | 5 | 30.40 |
| | | 6 | 30.20 | | 6 | 30.50 |
| | | 7 | 29.60 | | 7 | 29.50 |
| | | 8 | 31.60 | | 8 | 28.56 |
| | | 9 | 32.00 | | 9 | 31.60 |
| | | 10 | 31.60 | | 10 | 30.60 |
| | | 11 | 30.60 | | 11 | 29.96 |
| | | 12 | 31.20 | | 12 | 30.90 |
| | | 13 | 32.20 | | 13 | 30.20 |
| | | 14 | 31.36 | | 14 | 29.52 |
| | | 15 | 31.36 | | 15 | 30.80 |
| | | 16 | 32.00 | | 16 | 31.30 |
| | | 17 | 31.76 | | 17 | 31.10 |
| | | 18 | 31.90 | | 18 | 30.30 |
| | | 19 | 32.50 | | 19 | 30.80 |
| | | 20 | 30.60 | | 20 | 29.10 |

Analysis of variance table for packed cell volume

| Source of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 29.133 | 1 | 29.133 | 0.094 | 0.76 |
| Error (a) | 2468.309 | 8 | 308.539 | | |
| B** | 372.383 | 18 | 20.688 | 1.308 | 0.19 |
| AB*** | 61.098 | 18 | 3.394 | 0.214 | 1.00 |
| Remaining error | 2278.074 | 144 | 15.820 | | |
| Total | 5208.999 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 4. Leukocytes counts for thymectomized and control calves (leukocytes/cmm.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 9317 | | | 9020 |
| Individual means for 19 weeks | | T1 | 13439 | | C1 | 13004 |
| | | T2 | 10979 | | C2 | 10798 |
| | | T3 | 9858 | | C3 | 8258 |
| | | T4 | 6574 | | C4 | 7163 |
| | | T5 | 5737 | | C5 | 5879 |
| Weekly means for each group | week | 2 | 7817 | week | 2 | 6928 |
| | | 3 | 8886 | | 3 | 11026 |
| | | 4 | 10639 | | 4 | 7884 |
| | | 5 | 8051 | | 5 | 8135 |
| | | 6 | 11745 | | 6 | 7155 |
| | | 7 | 12659 | | 7 | 9649 |
| | | 8 | 6880 | | 8 | 12189 |
| | | 9 | 7957 | | 9 | 6304 |
| | | 10 | 9659 | | 10 | 8324 |
| | | 11 | 12156 | | 11 | 10956 |
| | | 12 | 8513 | | 12 | 8366 |
| | | 13 | 9973 | | 13 | 8455 |
| | | 14 | 8400 | | 14 | 9141 |
| | | 15 | 8079 | | 15 | 8663 |
| | | 16 | 7819 | | 16 | 9506 |
| | | 17 | 9202 | | 17 | 9865 |
| | | 18 | 8584 | | 18 | 9700 |
| | | 19 | 9724 | | 19 | 9790 |
| | | 20 | 10284 | | 20 | 9352 |

Analysis of variance table for erythrocyte values

| Source of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 4184286 | 1 | 4184286 | 0.024 | 0.85 |
| Error (a) | 1393058899 | 8 | 174132362 | | |
| B** | 215410094 | 18 | 11967227 | 0.920 | 0.56 |
| AB*** | 210444677 | 18 | 11691371 | 0.898 | 0.58 |
| Remaining error | 1873604347 | 144 | 13011141 | | |
| Total | 3696702302 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 5a. Relative lymphocyte values for thymectomized and control calves (%)

| | | Thymectomized | | Control | |
|---|---|---|---|---|---|
| Overall mean | | | 47.87 | | 55.18 |
| Individual means for 19 weeks | | T1 | 48.16 | C1 | 55.00 |
| | | T2 | 46.32 | C2 | 44.16 |
| | | T3 | 38.32 | C3 | 54.10 |
| | | T4 | 49.74 | C4 | 57.63 |
| | | T5 | 56.84 | C5 | 65.00 |
| Weekly means for each group | week | 2 | 35.60 | week 2 | 63.60 |
| | | 3 | 61.80 | 3 | 55.60 |
| | | 4 | 55.00 | 4 | 63.20 |
| | | 5 | 55.20 | 5 | 65.00 |
| | | 6 | 40.80 | 6 | 67.40 |
| | | 7 | 38.00 | 7 | 51.80 |
| | | 8 | 58.00 | 8 | 48.80 |
| | | 9 | 54.40 | 9 | 61.20 |
| | | 10 | 40.00 | 10 | 54.80 |
| | | 11 | 40.60 | 11 | 43.20 |
| | | 12 | 45.00 | 12 | 45.20 |
| | | 13 | 49.60 | 13 | 49.40 |
| | | 14 | 53.60 | 14 | 48.40 |
| | | 15 | 50.60 | 15 | 48.20 |
| | | 16 | 48.20 | 16 | 54.00 |
| | | 17 | 43.00 | 17 | 50.60 |
| | | 18 | 47.60 | 18 | 53.00 |
| | | 19 | 44.00 | 19 | 58.40 |
| | | 20 | 48.80 | 20 | 66.60 |

Analysis of variance table for relative lymphocyte values

| Source of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 2535 | 1 | 2535 | 2.649 | 0.14 |
| Error (a) | 7655 | 8 | 957 | | |
| B** | 5004 | 18 | 278 | 1.407 | 0.14 |
| AB*** | 4778 | 18 | 265 | 1.344 | 0.17 |
| Remaining error | 28448 | 144 | 198 | | |
| Total | 48419 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 5b. Absolute lymphocyte values for thymectomized and control calves (lymphocytes/cmm.)

| | | Thymectomized | | Control | |
|---|---|---|---|---|---|
| Overall mean | | | 4066 | | 4781 |
| Individual means for 19 weeks | | T1 | 5977 | C1 | 7259 |
| | | T2 | 4812 | C2 | 4455 |
| | | T3 | 3369 | C3 | 4338 |
| | | T4 | 3054 | C4 | 4038 |
| | | T5 | 3119 | C5 | 3816 |
| Weekly means for each group | week | 2 | 2503 | week 2 | 4251 |
| | | 3 | 5134 | 3 | 6308 |
| | | 4 | 5737 | 4 | 4690 |
| | | 5 | 4394 | 5 | 4926 |
| | | 6 | 3079 | 6 | 4585 |
| | | 7 | 3271 | 7 | 4640 |
| | | 8 | 3645 | 8 | 5382 |
| | | 9 | 4465 | 9 | 3855 |
| | | 10 | 3639 | 10 | 4833 |
| | | 11 | 4377 | 11 | 4231 |
| | | 12 | 3690 | 12 | 3574 |
| | | 13 | 3974 | 13 | 4219 |
| | | 14 | 4215 | 14 | 4282 |
| | | 15 | 3936 | 15 | 3903 |
| | | 16 | 3860 | 16 | 5151 |
| | | 17 | 4057 | 17 | 5080 |
| | | 18 | 4200 | 18 | 5008 |
| | | 19 | 4436 | 19 | 5872 |
| | | 20 | 4645 | 20 | 6049 |

Analysis of variance table for absolute lymphocyte values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 24276753 | 1 | 24276753 | 0.703 | 0.43 |
| Error (a) | 276299199 | 8 | 34537400 | | |
| B** | 66205228 | 18 | 3678068 | 1.634 | 0.06 |
| AB*** | 31399065 | 18 | 1744392 | 0.775 | 0.73 |
| Remaining error | 324147049 | 144 | 2251021 | | |
| Total | 722327294 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 6a. Relative neutrophil values for thymectomized and control calves (%)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 43.96 | | | 41.20 |
| Individual means for 19 weeks | | T1 | 46.00 | | C1 | 38.53 |
| | | T2 | 45.74 | | C2 | 49.10 |
| | | T3 | 38.32 | | C3 | 54.10 |
| | | T4 | 54.84 | | C4 | 36.21 |
| | | T5 | 34.89 | | C5 | 28.05 |
| Weekly means for each group | week | 2 | 47.00 | week | 2 | 51.20 |
| | | 3 | 46.20 | | 3 | 35.80 |
| | | 4 | 40.40 | | 4 | 40.00 |
| | | 5 | 38.80 | | 5 | 33.00 |
| | | 6 | 42.80 | | 6 | 30.20 |
| | | 7 | 48.00 | | 7 | 39.20 |
| | | 8 | 39.20 | | 8 | 46.60 |
| | | 9 | 50.60 | | 9 | 39.20 |
| | | 10 | 40.40 | | 10 | 34.60 |
| | | 11 | 48.20 | | 11 | 49.80 |
| | | 12 | 43.60 | | 12 | 48.00 |
| | | 13 | 48.20 | | 13 | 39.40 |
| | | 14 | 34.00 | | 14 | 42.60 |
| | | 15 | 44.00 | | 15 | 46.60 |
| | | 16 | 38.60 | | 16 | 39.40 |
| | | 17 | 43.00 | | 17 | 41.00 |
| | | 18 | 44.40 | | 18 | 44.00 |
| | | 19 | 48.40 | | 19 | 44.80 |
| | | 20 | 49.40 | | 20 | 37.40 |

Analysis of variance table for relative neutrophil values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 361 | 1 | 361 | 0.226 | 0.65 |
| Error (a) | 12800 | 8 | 1600 | | |
| B** | 2760 | 18 | 153 | 0.671 | 0.84 |
| AB*** | 2029 | 18 | 113 | 0.494 | 0.96 |
| Remaining error | 32886 | 144 | 228 | | |
| Total | 50836 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 6b. Absolute neutrophil values for thymectomized and control calves (neutrophils/cmm.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 4589 | | | 3678 |
| Individual means for 19 weeks | | T1 | 6723 | | C1 | 4953 |
| | | T2 | 5342 | | C2 | 5653 |
| | | T3 | 5810 | | C3 | 3453 |
| | | T4 | 2992 | | C4 | 2674 |
| | | T5 | 2076 | | C5 | 1657 |
| Weekly means for each group | week | 2 | 4773 | week | 2 | 2500 |
| | | 3 | 3202 | | 3 | 4161 |
| | | 4 | 4001 | | 4 | 2476 |
| | | 5 | 3005 | | 5 | 2863 |
| | | 6 | 8009 | | 6 | 2185 |
| | | 7 | 8915 | | 7 | 4536 |
| | | 8 | 2656 | | 8 | 6259 |
| | | 9 | 3070 | | 9 | 1971 |
| | | 10 | 5350 | | 10 | 3387 |
| | | 11 | 7084 | | 11 | 6103 |
| | | 12 | 4249 | | 12 | 4139 |
| | | 13 | 4972 | | 13 | 3730 |
| | | 14 | 3634 | | 14 | 4067 |
| | | 15 | 3526 | | 15 | 4188 |
| | | 16 | 3342 | | 16 | 3651 |
| | | 17 | 4378 | | 17 | 3984 |
| | | 18 | 3585 | | 18 | 3785 |
| | | 19 | 4527 | | 19 | 3221 |
| | | 20 | 4908 | | 20 | 2681 |

Analysis of variance table for absolute neutrophil values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 39375711 | 1 | 39375711 | 0.634 | 0.45 |
| Error (a) | 496712418 | 8 | 62089052 | | |
| B** | 201517588 | 18 | 11195422 | 1.169 | 0.29 |
| AB*** | 184808612 | 18 | 10267145 | 1.072 | 0.39 |
| Remaining error | 1378697497 | 144 | 9574288 | | |
| Total | 2301111826 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 7a. Relative monocyte values for thymectomized and control calves (%)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 5.10 | | | 5.14 |
| Individual means for 19 weeks | | T1 | 4.21 | | C1 | 5.21 |
| | | T2 | 7.05 | | C2 | 5.32 |
| | | T3 | 4.74 | | C3 | 4.74 |
| | | T4 | 4.26 | | C4 | 5.42 |
| | | T5 | 5.26 | | C5 | 5.00 |
| Weekly means for each group | week | 2 | 4.40 | week | 2 | 4.40 |
| | | 3 | 4.60 | | 3 | 5.00 |
| | | 4 | 6.60 | | 4 | 5.60 |
| | | 5 | 6.00 | | 5 | 3.60 |
| | | 6 | 4.80 | | 6 | 3.60 |
| | | 7 | 3.20 | | 7 | 4.20 |
| | | 8 | 5.80 | | 8 | 4.40 |
| | | 9 | 4.40 | | 9 | 5.60 |
| | | 10 | 5.20 | | 10 | 5.00 |
| | | 11 | 5.60 | | 11 | 4.80 |
| | | 12 | 4.40 | | 12 | 5.80 |
| | | 13 | 4.60 | | 13 | 4.80 |
| | | 14 | 5.00 | | 14 | 6.40 |
| | | 15 | 5.00 | | 15 | 6.20 |
| | | 16 | 5.20 | | 16 | 5.80 |
| | | 17 | 6.40 | | 17 | 7.00 |
| | | 18 | 5.60 | | 18 | 6.00 |
| | | 19 | 5.20 | | 19 | 5.00 |
| | | 20 | 5.00 | | 20 | 4.40 |

Analysis of variance table for relative monocyte values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.047 | 1 | 0.047 | 0.003 | 0.91 |
| Error (a) | 109.432 | 8 | 13.679 | | |
| B** | 85.916 | 18 | 4.773 | 0.995 | 0.47 |
| AB*** | 50.253 | 18 | 2.792 | 0.582 | 0.91 |
| Remaining error | 690.568 | 144 | 4.796 | | |
| Total | 936.216 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 7b. Absolute monocyte values for thymectomized and control calves (monocytes/cmm.)

| | | Thymectomized | | Control |
|---|---|---|---|---|
| Overall mean | | 465.0 | | 460.9 |
| Individual means for 19 weeks | | T1 526.2 | | C1 702.5 |
| | | T2 724.5 | | C2 527.5 |
| | | T3 468.0 | | C3 393.6 |
| | | T4 277.9 | | C4 385.2 |
| | | T5 328.4 | | C5 295.6 |
| Weekly means for each group | week 2 | 371.2 | week 2 | 332.2 |
| | 3 | 408.4 | 3 | 525.0 |
| | 4 | 684.6 | 4 | 498.6 |
| | 5 | 536.0 | 5 | 302.6 |
| | 6 | 595.6 | 6 | 280.2 |
| | 7 | 368.0 | 7 | 403.8 |
| | 8 | 427.8 | 8 | 470.2 |
| | 9 | 339.6 | 9 | 336.2 |
| | 10 | 478.4 | 10 | 370.4 |
| | 11 | 580.0 | 11 | 416.6 |
| | 12 | 359.4 | 12 | 500.4 |
| | 13 | 384.0 | 13 | 380.2 |
| | 14 | 415.0 | 14 | 545.6 |
| | 15 | 439.6 | 15 | 535.8 |
| | 16 | 425.2 | 16 | 592.4 |
| | 17 | 569.8 | 17 | 668.8 |
| | 18 | 499.6 | 18 | 604.4 |
| | 19 | 522.6 | 19 | 543.0 |
| | 20 | 430.6 | 20 | 450.4 |

Analysis of variance table for absolute monocyte values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 813 | 1 | 813 | 0.002 | 0.92 |
| Error (a) | 4277701 | 8 | 534713 | | |
| B** | 1043819 | 18 | 57990 | 1.139 | 0.32 |
| AB*** | 851372 | 18 | 47298 | 0.929 | 0.55 |
| Remaining error | 7329869 | 144 | 50902 | | |
| Total | 13503574 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 8a. Relative eosinophil values for thymectomized and control calves (%)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 8.30 | | | 5.20 |
| Individual means for 19 weeks | | T1 | 1.32 | | C1 | 1.26 |
| | | T2 | 0.37 | | C2 | 0.58 |
| | | T3 | 0.47 | | C3 | 0.16 |
| | | T4 | 1.10 | | C4 | 0.32 |
| | | T5 | 1.10 | | C5 | 0.42 |
| Weekly means for each group | week | 2 | 0.60 | week | 2 | 0.00 |
| | | 3 | 0.80 | | 3 | 0.40 |
| | | 4 | 0.60 | | 4 | 0.80 |
| | | 5 | 0.40 | | 5 | 0.20 |
| | | 6 | 0.20 | | 6 | 0.60 |
| | | 7 | 0.40 | | 7 | 0.20 |
| | | 8 | 0.20 | | 8 | 0.80 |
| | | 9 | 0.60 | | 9 | 1.20 |
| | | 10 | 0.80 | | 10 | 0.40 |
| | | 11 | 1.00 | | 11 | 0.80 |
| | | 12 | 1.20 | | 12 | 0.60 |
| | | 13 | 0.60 | | 13 | 0.80 |
| | | 14 | 0.60 | | 14 | 0.60 |
| | | 15 | 0.40 | | 15 | 0.00 |
| | | 16 | 1.40 | | 16 | 0.20 |
| | | 17 | 1.00 | | 17 | 0.20 |
| | | 18 | 2.60 | | 18 | 1.60 |
| | | 19 | 1.80 | | 19 | 0.20 |
| | | 20 | 1.40 | | 20 | 0.80 |

Analysis of variance table for relative eosinophil values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 5.058 | 1 | 5.058 | 1.466 | 0.12 |
| Error (a) | 27.600 | 8 | 3.450 | | |
| B** | 32.379 | 18 | 1.799 | 1.556 | 0.08 |
| AB*** | 15.642 | 18 | 0.869 | 0.752 | 0.74 |
| Remaining error | 166.400 | 144 | 1.156 | | |
| Total | 247.079 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 8b. Absolute eosinophil values for thymectomized and control calves (eosinophils/cmm.)

| | | Thymectomized | | | Control |
|---|---|---|---|---|---|
| Overall mean | | | 87.31 | | 54.10 |
| Individual means for 19 weeks | | T1 | 167.84 | C1 | 164.79 |
| | | T2 | 90.89 | C2 | 60.00 |
| | | T3 | 44.16 | C3 | 12.95 |
| | | T4 | 81.79 | C4 | 22.00 |
| | | T5 | 74.84 | C5 | 25.00 |
| Weekly means for each group | week | 2 | 46.80 | week 2 | 0.00 |
| | | 3 | 118.80 | 3 | 32.20 |
| | | 4 | 64.40 | 4 | 71.60 |
| | | 5 | 49.20 | 5 | 21.40 |
| | | 6 | 24.20 | 6 | 42.00 |
| | | 7 | 88.00 | 7 | 34.20 |
| | | 8 | 19.00 | 8 | 100.20 |
| | | 9 | 235.20 | 9 | 94.00 |
| | | 10 | 87.40 | 10 | 45.40 |
| | | 11 | 101.60 | 11 | 128.20 |
| | | 12 | 113.60 | 12 | 77.60 |
| | | 13 | 79.80 | 13 | 81.40 |
| | | 14 | 38.20 | 14 | 83.20 |
| | | 15 | 26.60 | 15 | 0.00 |
| | | 16 | 98.00 | 16 | 30.40 |
| | | 17 | 86.40 | 17 | 22.40 |
| | | 18 | 214.60 | 18 | 125.40 |
| | | 19 | 148.40 | 19 | 13.40 |
| | | 20 | 106.00 | 20 | 79.00 |

Analysis of variance table for absolute eosinophil values

| Sources of variance | Sum of squares | Degs. of free-dom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 58048 | 1 | 58048 | 1.008 | 0.45 |
| Error (a) | 460900 | 8 | 57613 | | |
| B** | 305359 | 18 | 16964 | 1.030 | 0.43 |
| AB*** | 147792 | 18 | 8211 | 0.499 | 0.96 |
| Remaining error | 2370644 | 144 | 16463 | | |
| Total | 3342752 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE

Over

Indi
for

Wei
ea

TABLE 9a. Relative basophil values for thymectomized and control calves (%)

| | | Thymectomized | | Control | |
|---|---|---|---|---|---|
| Overall mean | | 1.23 | | 0.93 | |
| Individual means for 19 weeks | | T1 | 0.37 | C1 | 0.58 |
| | | T2 | 0.84 | C2 | 0.79 |
| | | T3 | 1.95 | C3 | 0.84 |
| | | T4 | 1.10 | C4 | 0.89 |
| | | T5 | 1.89 | C5 | 1.53 |
| Weekly means for each group | week | 2 | 0.60 | week 2 | 0.20 |
| | | 3 | 0.40 | 3 | 0.00 |
| | | 4 | 1.60 | 4 | 0.80 |
| | | 5 | 0.80 | 5 | 0.00 |
| | | 6 | 0.60 | 6 | 1.00 |
| | | 7 | 0.60 | 7 | 1.20 |
| | | 8 | 1.80 | 8 | 0.80 |
| | | 9 | 0.40 | 9 | 1.20 |
| | | 10 | 1.20 | 10 | 1.20 |
| | | 11 | 0.80 | 11 | 0.80 |
| | | 12 | 1.60 | 12 | 1.00 |
| | | 13 | 2.00 | 13 | 0.80 |
| | | 14 | 1.40 | 14 | 0.80 |
| | | 15 | 2.20 | 15 | 0.60 |
| | | 16 | 1.80 | 16 | 1.00 |
| | | 17 | 1.20 | 17 | 1.40 |
| | | 18 | 1.00 | 18 | 2.00 |
| | | 19 | 1.20 | 19 | 1.80 |
| | | 20 | 2.20 | 20 | 1.00 |

Analysis of variance table for relative basophil values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 4.426 | 1 | 4.426 | 0.786 | 0.40 |
| Error (a) | 45.074 | 8 | 5.634 | | |
| B** | 30.716 | 18 | 1.706 | 1.980 | 0.01 |
| AB*** | 25.474 | 18 | 1.415 | 1.642 | 0.06 |
| Remaining error | 124.126 | 144 | 0.862 | | |
| Total | 229.816 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 9b. Absolute basophil values for thymectomized and control calves (basophils/cmm.)

| | | Thymectomized | | Control | |
|---|---|---|---|---|---|
| Overall mean | | 100.62 | | 73.84 | |
| Individual means for 19 weeks | | T1 | 46.79 | C1 | 87.84 |
| | | T2 | 94.89 | C2 | 68.58 |
| | | T3 | 166.74 | C3 | 64.10 |
| | | T4 | 77.26 | C4 | 63.47 |
| | | T5 | 117.42 | C5 | 85.21 |
| Weekly means for each group | week | 2 | 43.40 | week 2 | 17.60 |
| | | 3 | 22.00 | 3 | 0.00 |
| | | 4 | 175.80 | 4 | 60.80 |
| | | 5 | 67.20 | 5 | 0.00 |
| | | 6 | 38.20 | 6 | 50.80 |
| | | 7 | 35.40 | 7 | 88.60 |
| | | 8 | 137.20 | 8 | 62.80 |
| | | 9 | 25.20 | 9 | 63.40 |
| | | 10 | 103.80 | 10 | 86.60 |
| | | 11 | 52.00 | 11 | 46.00 |
| | | 12 | 121.00 | 12 | 76.00 |
| | | 13 | 178.60 | 13 | 56.60 |
| | | 14 | 129.60 | 14 | 63.80 |
| | | 15 | 153.00 | 15 | 37.00 |
| | | 16 | 129.20 | 16 | 80.80 |
| | | 17 | 110.20 | 17 | 132.20 |
| | | 18 | 85.20 | 18 | 206.60 |
| | | 19 | 111.20 | 19 | 179.80 |
| | | 20 | 193.60 | 20 | 93.60 |

Analysis of variance table for absolute basophil values

| Sources of variance | Sum of squares | Degs. of free-dom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 34063 | 1 | 34063 | 1.652 | 0.23 |
| Error (a) | 165015 | 8 | 20627 | | |
| B** | 315486 | 18 | 17527 | 2.362 | 0.00 |
| AB*** | 206365 | 18 | 11465 | 1.545 | 0.08 |
| Remaining error | 1068763 | 144 | 7422 | | |
| Total | 1789692 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 10. Total protein values for thymectomized and control calves (gm./100 ml.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 5.25 | | | 5.23 |
| Individual means for 19 weeks | | T1 | 5.73 | | C1 | 5.80 |
| | | T2 | 5.63 | | C2 | 5.31 |
| | | T3 | 5.24 | | C3 | 4.96 |
| | | T4 | 5.12 | | C4 | 5.26 |
| | | T5 | 4.54 | | C5 | 4.82 |
| Weekly means for each group | week | 2 | 4.24 | week | 2 | 4.56 |
| | | 3 | 3.74 | | 3 | 4.47 |
| | | 4 | 4.66 | | 4 | 4.49 |
| | | 5 | 4.69 | | 5 | 4.64 |
| | | 6 | 5.01 | | 6 | 4.84 |
| | | 7 | 5.06 | | 7 | 5.10 |
| | | 8 | 5002 | | 8 | 4.77 |
| | | 9 | 5.60 | | 9 | 5.39 |
| | | 10 | 5.40 | | 10 | 5.25 |
| | | 11 | 5.36 | | 11 | 5.25 |
| | | 12 | 5.41 | | 12 | 5.27 |
| | | 13 | 5.50 | | 13 | 5.39 |
| | | 14 | 5.53 | | 14 | 5.32 |
| | | 15 | 5.48 | | 15 | 5.75 |
| | | 16 | 5.68 | | 16 | 5.57 |
| | | 17 | 5.73 | | 17 | 5.71 |
| | | 18 | 5.78 | | 18 | 5.73 |
| | | 19 | 5.99 | | 19 | 5.83 |
| | | 20 | 5.88 | | 20 | 6.04 |

Analysis of variance for total protein values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.020 | 1 | 0.020 | 0.006 | 0.90 |
| Error (a) | 27.991 | 8 | 3.499 | | |
| B** | 49.071 | 18 | 2.726 | 10.618 | 0.00 |
| AB*** | 2.647 | 18 | 0.147 | 0.573 | 0.91 |
| Remaining error | 36.972 | 144 | 0.257 | | |
| Total | 116.701 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 11. Albumin values for thymectomized and control calves (gm./100 ml.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 3.18 | | | 3.25 |
| Individual means for 19 weeks | | T1 | 3.15 | | C1 | 3.15 |
| | | T2 | 3.12 | | C2 | 3.26 |
| | | T3 | 3.20 | | C3 | 3.26 |
| | | T4 | 3.52 | | C4 | 3.52 |
| | | T5 | 2.89 | | C5 | 3.07 |
| Weekly means for each group | week | 2 | 2.89 | week | 2 | 3.16 |
| | | 3 | 2.39 | | 3 | 2.95 |
| | | 4 | 2.96 | | 4 | 2.99 |
| | | 5 | 3.03 | | 5 | 2.85 |
| | | 6 | 3.12 | | 6 | 3.15 |
| | | 7 | 3.06 | | 7 | 3.22 |
| | | 8 | 3.05 | | 8 | 2.95 |
| | | 9 | 3.42 | | 9 | 3.35 |
| | | 10 | 3.15 | | 10 | 3.21 |
| | | 11 | 3.14 | | 11 | 3.25 |
| | | 12 | 3.18 | | 12 | 3.27 |
| | | 13 | 3.26 | | 13 | 3.29 |
| | | 14 | 3.24 | | 14 | 3.37 |
| | | 15 | 3.29 | | 15 | 3.64 |
| | | 16 | 3.35 | | 16 | 3.41 |
| | | 17 | 3.41 | | 17 | 3.45 |
| | | 18 | 3.43 | | 18 | 3.33 |
| | | 19 | 3.51 | | 19 | 3.48 |
| | | 20 | 3.48 | | 20 | 3.48 |

Analysis of variance table for albumin values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.268 | 1 | 0.268 | 0.354 | 0.57 |
| Error (a) | 6.068 | 8 | 0.758 | | |
| B** | 8.656 | 18 | 0.481 | 4.014 | 0.00 |
| AB*** | 1.332 | 18 | 0.074 | 0.618 | 0.88 |
| Remaining error | 17.251 | 144 | 0.120 | | |
| Total | 33.576 | 189 | | | |

* Treated
** Trials
***Treated vs. Trials

TABLE 12. Globulin values for thymectomized and control calves (gm./100 ml.)

| | | Thymectomized | | | Control | |
|---|---|---|---|---|---|---|
| Overall mean | | | 2.07 | | | 1.98 |
| Individual means for 19 weeks | | T1 | 2.57 | | C1 | 2.65 |
| | | T2 | 2.50 | | C2 | 2.05 |
| | | T3 | 2.03 | | C3 | 1.70 |
| | | T4 | 1.60 | | C4 | 1.74 |
| | | T5 | 1.64 | | C5 | 1.74 |
| Weekly means for each group | week | 2 | 1.35 | week | 2 | 1.40 |
| | | 3 | 1.35 | | 3 | 1.52 |
| | | 4 | 1.70 | | 4 | 1.50 |
| | | 5 | 1.66 | | 5 | 1.79 |
| | | 6 | 1.87 | | 6 | 1.70 |
| | | 7 | 2.00 | | 7 | 1.89 |
| | | 8 | 1.97 | | 8 | 1.82 |
| | | 9 | 2.18 | | 9 | 2.04 |
| | | 10 | 2.24 | | 10 | 2.03 |
| | | 11 | 2.22 | | 11 | 1.99 |
| | | 12 | 2.23 | | 12 | 2.00 |
| | | 13 | 2.24 | | 13 | 2.10 |
| | | 14 | 2.29 | | 14 | 1.95 |
| | | 15 | 2.19 | | 15 | 2.12 |
| | | 16 | 2.33 | | 16 | 2.16 |
| | | 17 | 2.32 | | 17 | 2.26 |
| | | 18 | 2.35 | | 18 | 2.40 |
| | | 19 | 2.48 | | 19 | 2.35 |
| | | 20 | 2.40 | | 20 | 2.56 |

Analysis of variance table for globulin values

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.422 | 1 | 0.422 | 0.119 | 0.73 |
| Error (a) | 28.369 | 8 | 3.546 | | |
| B** | 18.101 | 18 | 1.006 | 10.031 | 0.00 |
| AB*** | 0.921 | 18 | 0.051 | 0.510 | 0.95 |
| Remaining error | 14.436 | 144 | 0.100 | | |
| Total | 62.250 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 13. Albumin:globulin ratio for thymectomized and control calves

| | | Thymectomized | | Control | |
|---|---|---|---|---|---|
| Overall mean | | | 1.61 | | 1.75 |
| Individual means for 19 weeks | | T1 | 1.21 | C1 | 1.24 |
| | | T2 | 1.31 | C2 | 1.65 |
| | | T3 | 1.63 | C3 | 1.96 |
| | | T4 | 2.21 | C4 | 2.08 |
| | | T5 | 1.68 | C5 | 1.79 |
| Weekly means for each group | week | 2 | 2.15 | week 2 | 2.27 |
| | | 3 | 1.52 | 3 | 1.94 |
| | | 4 | 1.82 | 4 | 2.03 |
| | | 5 | 1.86 | 5 | 1.71 |
| | | 6 | 1.71 | 6 | 1.71 |
| | | 7 | 1.64 | 7 | 1.86 |
| | | 8 | 1.66 | 8 | 1.76 |
| | | 9 | 1.53 | 9 | 1.80 |
| | | 10 | 1.49 | 10 | 1.73 |
| | | 11 | 1.52 | 11 | 1.83 |
| | | 12 | 1.52 | 12 | 1.75 |
| | | 13 | 1.57 | 13 | 1.66 |
| | | 14 | 1.52 | 14 | 1.75 |
| | | 15 | 1.63 | 15 | 1.79 |
| | | 16 | 1.49 | 16 | 1.66 |
| | | 17 | 1.48 | 17 | 1.62 |
| | | 18 | 1.48 | 18 | 1.41 |
| | | 19 | 1.45 | 19 | 1.53 |
| | | 20 | 1.51 | 20 | 1.37 |

Analysis of variance table for albumin:globulin ratio

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 0.902 | 1 | 0.902 | 0.364 | 0.57 |
| Error (a) | 19.794 | 8 | 2.474 | | |
| B** | 5.507 | 18 | 0.306 | 3.238 | 0.00 |
| AB*** | 1.009 | 18 | 0.056 | 0.593 | 0.90 |
| Remaining error | 13.603 | 144 | 0.094 | | |
| Total | 40.814 | 189 | | | |

* Treated
** Trials
***Treated vs. trials

TABLE 14. Cumulative weight gains for thymectomized and control calves (kg.)

| | Thymectomized | | Control | |
|---|---|---|---|---|
| Mean weights | | 25.8 | | 27.6 |
| Biweekly cumulative weights | week 0 | 0.0 | week 0 | 0.0 |
| | 2 | -1.0 | 2 | -1.4 |
| | 4 | 0.9 | 4 | 3.3 |
| | 6 | 6.8 | 6 | 6.9 |
| | 8 | 10.6 | 8 | 14.4 |
| | 10 | 17.5 | 10 | 22.8 |
| | 12 | 27.9 | 12 | 27.8 |
| | 14 | 33.6 | 14 | 36.9 |
| | 16 | 44.4 | 16 | 46.7 |
| | 18 | 53.9 | 18 | 55.0 |
| | 20 | 63.0 | 20 | 63.8 |
| Actual final cumulative weights | | 64.4 | | 69.0 |

Analysis of variance table for cumulative weight gains****

| Sources of variance | Sum of squares | Degs. of freedom | Mean square | F stat. | Approx. signif. prob. of F stat. |
|---|---|---|---|---|---|
| A* | 412 | 1 | 412 | 1.375 | 0.25 |
| Error (a) | 11992 | 40 | 300 | | |
| B** | 221269 | 9 | 24585 | 178.917 | 0.00 |
| AB*** | 391 | 9 | 43 | 0.732 | 0.68 |
| Remaining error | 2372 | 40 | 59 | | |
| Total | 236437 | 99 | | | |

* Treated
** Trials
*** Treated vs. trials
****These figures are presented as they were originally analyzed in pounds.