



This Thesis Has Been

MICROFILMED

Negative No. T-

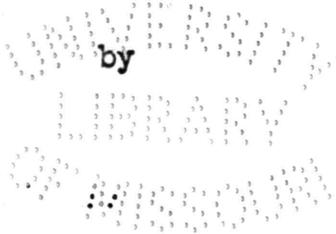
1368

1932

Form 26

THE EFFECT OF AGE AND CONDITION ON THE MINERAL
COMPOSITION OF THE ASH OF THE BOVINE

*Approved
May 10-1914
P. F. Howbridge*



Benjamin Elliott Sive, B. Ch. E.

SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE DEGREE OF
MASTER OF ARTS

in the

GRADUATE SCHOOL

of the

UNIVERSITY OF MISSOURI

1916

Table of Contents

	Page
Introduction	1
Historical	3
Experimental	11
Description of Animals	11
Slaughtering	11
Total Ash	13
Preparation of Ash	14
Methods of Ashing	14
Plan of Analyses	19
Analysis of Soluble Ash	20
Analysis of Insoluble Ash	27
Analysis of Total Ash	33
Calculations	34
Discussion	37
Effect of Age	37
Effect of Condition	43
General Relations	45
Summary	47
Appendix	51
Tables I to XXVI	51-75
Bibliography	76

THE EFFECT OF AGE AND CONDITION ON THE MINERAL COMPOSITION
OF THE ASH OF THE BOVINE

Introduction

It has long been known that the ash constituents or the mineral elements of the body bear an important relation to body structure and life processes of animals. However, the amount of data on the composition of the ash of animals is very small and in most cases incomplete. Furthermore, a great deal of the data as presented by various investigators cannot be compared because of lack of uniformity in reporting results as well as in the selection of samples for analysis.

Forbes¹ in his bulletin on "The mineral elements in animal nutrition" comments on this, saying "Our knowledge of the amounts and kinds of mineral matter required by animals is indefinite and fragmentary. Much progress is yet to be made in this field." This is especially true because the purpose of previous investigators seems to have been to get a representative analysis of different animals or parts of animals without any reference to factors which may or may not influence the mineral composition of the animal.

To the nutrition chemist data taken from experiments which have been systematized so that the results are comparable are of great value.

The data presented in this thesis are the first to be offered from a series of experiments now in progress. The ultimate end of this combined series is to study the distribution of mineral constituents in the ash of the different parts of steers as well as in the entire animal ash with a special view of determining, if possible, what effect the age or condition of a steer may have upon his ash composition. The entire work is a part of the so-called "Use of Food" experiment.

The work offered in this paper consists of data on six steers, three old and three young. The condition of each of the older steers was different but there was a steer in the group of young animals which was in a corresponding condition.

These data are limited to the effect of age and condition on the distribution of the ash constituents in the composite skeleton, composite lean and fat, and composite internal organs and internal fat. The work required the preparation and complete analysis of thirty samples, totaling about three hundred determinations in duplicate.

Historical

A careful review of the literature disclosed very little on the subject matter of this paper. Ash analyses of animals have been confined to a very few investigators each of whom seems to have been interested in some special phase of the work. Analyses have been reported on blood, wool, bones, some specific organ such as the liver and the gall, and in a few cases the entire animal composite was analyzed. However, the work as reported was confined chiefly to the comparison of a certain part of some animal, such as muscle or blood, with a similar part of animals of other species.

Lawes and Gilbert² who have probably done more research in ash work than any other investigators, analyzed ten different animals, giving, in addition to the composition of the entire animal, compositions of portions which they call the carcass and the collective offal parts. These divisions of the animal body are of such a character that no comparison of their work with the data offered here can be made.

There are several investigators, however, who have touched upon the problem in some manner and whose work is sufficiently related to be recorded here.

In 1872 E. Wildt³ reported results obtained at the Experiment Station at Proskau, Germany, on the effect of age on the composition of the ash of rabbit bones. He used the average composition of several animals of the same age, using rabbits ranging from new born to four years old. From these data he

concluded that the lime content increases slightly toward maturity, the carbon dioxide increases considerably, while both magnesia and phosphoric acid diminish.

Percentage Composition of Ash of Bones of Rabbits.
(E. Wildt)

No. of Animals used	Age	Ash	CaO	MgO	P ₂ O ₅	CO ₂
6	Newly born	53.39	52.17	1.38	42.02	3.65
5	Three days	50.82	52.16	1.36	42.13	3.84
4	Fourteen days	55.18	52.10	1.26	42.19	3.99
4	One month	58.94	51.91	1.22	42.20	4.00
2	Two months	65.63	52.10	1.09	41.64	4.52
3	Three months	67.68	52.49	1.01	41.03	4.69
3	Four months	68.72	52.60	1.02	40.80	4.92
2	Six months	70.26	52.64	1.05	40.80	4.94
2	Eight months	71.77	52.78	0.93	40.05	5.54
1	One year	74.24	52.61	0.91	40.04	5.71
1	Two years	72.90	52.76	0.93	39.78	5.81
1	Three-Four years	73.65	52.84	0.83	39.80	5.66

In addition, Wildt found only traces of soluble salts and no iron. In failing to find iron, he agrees with P. C. Plugge⁴ who says that iron is not a normal constituent of bone and, when found, is there as retained blood.

H. Weiske,⁵ assisted by E. Wildt, found in further experiments with rabbits that earthy phosphates when mixed with the food caused no noteworthy increase in magnesia, lime, or phosphoric acid. In addition, he reports that the age of an animal has an influence on the composition of the bones, those of old rabbits containing more salts than those of young rabbits. The percentage composition of the bone ash is very much the same in both but the proportion of lime is somewhat lower and of magnesia somewhat higher in young animals. The per cent of phosphoric acid is almost the same in all bones and averages about 42.17 per cent.

In 1891 Graffenberger,⁶ at the Institute of Animal Chemistry connected with the University of Breslau, under the direction of H. Weiske, worked on the composition of bones of aged rabbits. He reports results on two animals, one six and one-half and the other seven and one-half years old. He separated his samples into teeth, leg bones, and a composite of the remaining bones. Chlorine, sulphur, sodium, or potassium were not determined in teeth because of the lack of sufficient sample. He reports fluorine but does not vouch for the correctness of the results as the amount was obtained by difference. It might be mentioned in this connection that fluorine has been found to exist in bones. The amount is, however, very small and the methods for its quantitative determination very difficult so it was not deemed of sufficient value to be determined in the work presented in this paper. Graffenberger concludes

from his results that in the older animals there is more carbonic acid but a smaller amount of calcium phosphate than in rabbits three to four years of age.

Percentage Composition of Ash of Bones of Aged Rabbits
(Graffenberger)

	Male--			Female--		
	Six and a half years old			Seven and a half years old		
	Teeth	Leg Bones	Other Bones	Teeth	Leg Bones	Other Bones
Na ₂ O	---	0.49	0.81	---	0.65	0.59
K ₂ O	---	0.19	0.38	---	0.18	0.38
CaO	47.37	51.89	51.10	47.82	51.84	51.76
P ₂ O ₅	42.23	38.09	37.42	43.12	38.07	38.18
MgO	3.33	1.09	0.94	3.80	0.98	1.03
CO ₂	4.46	6.41	7.12	3.31	6.16	6.48
SO ₃	---	0.51	0.48	---	0.50	0.55
Cl	---	0.29	0.25	---	0.27	0.28
Fl	4.50	2.19	2.78	3.37	2.54	1.52

Summarizing the work of Weiske and his assistants, they conclude that there is a very slight gain in lime in the bones of rabbits up to an age of three to four years. At six and a half and seven and a half years this lime content was found to have decreased. The carbon dioxide increased with the age of the animal, the phosphoric acid decreased, and evidence of a diminution in the magnesia content was noted up to four years.

H. Brubacher⁷ analyzed the bones, muscles, organs, and entire bodies of three normal and five rachitic children. A complete analysis is not given and no very important conclusions drawn except that the foetus obtains most of its lime during the last month of its fetal development. Of the three normal children used (a) was a foetus of twenty weeks, (b) a newly born child, (c) a child four years old. The skeleton of (c) was not analyzed, the author using a rib which he considered a representative sample. Data obtained at this experiment station, however, show that no specific bone can be called representative of the entire skeleton.

Elements Reported in Ash of Bones of Children
(Brubacher)

	(a)	(b)	(c)
CaO	48.00	48.99	52.82
MgO	1.44	1.22	1.55
P ₂ O ₅	40.21	40.68	42.58

Elements Reported in Ash of Muscles of Children

	(a)	(b)	(c)
CaO	2.85	3.16	0.82
MgO	1.94	2.10	2.17
P ₂ O ₅	28.75	23.64	37.92
SiO ₂	1.36	0.35	1.80
Fe ₂ O ₃	1.16	1.39	0.75

S. Gabriel⁸ made a very extensive research on the methods

of analysis as well as on the composition of bone. One point of interest which he brings out is that there is more sodium than potassium in bone, the percentages, 1.1 per cent sodium and 0.2 per cent potassium, being rather constant. He also states that the sodium and potassium in bone are fixed so tightly that they cannot be washed out by water or by continued boiling with water.

A very interesting article by Hans Aron⁹ appeared in 1905. The paper is somewhat of a preliminary report on the influence of the alkalies upon the growth of bone. No further papers on the subject were found, however. In this report Aron says that when the sodium content of the food is diminished to a small amount and the potassium content is increased, there being just a sufficient quantity of calcium and phosphorus present, the calcium content and consequently the growth of the bone is below the normal. In this connection he analyzed the bones of several under-fed calves fed according to the above specifications. The results showed that only the absolute amount of the bone was diminished. The composition of the bone substance itself was the same as normal bone. He also found that the bone ash of diseased bone had the same composition as normal bone and verified Gabriel's conclusions that the sodium and potassium content was constant. No actual analyses were given.

An effort was made to obtain analyses of the composite skeleton of steers so that comparisons could be made with the work of other investigators. No data were available, how-

ever, in a form which could be used. The analysis of the shank bone of a steer might be worth recording.⁸

Analysis of the Shank Bone of a Steer
(Gabriel)

CaO	51.28
MgO	1.05
K ₂ O	0.18
Na ₂ O	1.09
Water of Crystallization	2.33
P ₂ O ₅	37.46
CO ₂	5.06
Cl	1.04

Several analyses of the ash of flesh were found in E. Wolff's¹⁰ "Ashen-Analysen".

In 100 parts of Pure Ash

	<u>Flesh of Man</u>	<u>Flesh of Ox</u>	<u>Flesh of Calf</u>	<u>Flesh of Hen</u>
Na ₂ O	22.9	14.5	25.6	20.5
K ₂ O	28.0	37.0	25.0	30.3
CaO	2.0	1.3	1.95	3.2
P ₂ O ₅	37.5	39.5	39.9	36.5
MgO	3.1	3.3	3.4	3.8
Cl	8.4	5.0	4.6	7.2

No iron, silica, sulphur, or carbon dioxide was reported. A small amount of CO₂ and a trace of SO₃ were found in the ash.

The flesh was ashed direct and not by chemical means.

Julius Katz¹¹ in his very complete paper on the mineral constituents of muscle flesh makes all his analyses on the fresh material because of the danger of losing constituents during ashing. He discusses methods and gives the analyses of the muscles of thirteen different animal species. These are calculated on the fresh and on the dry basis. A few of his analyses are recorded below.

Composition of Flesh on Fresh Basis

Parts per 1000

	<u>Man</u>	<u>Swine</u>	<u>Steer</u>	<u>Calf</u>
Na ₂ O	1.077	2.101	0.879	1.158
K ₂ O	3.857	3.057	4.411	4.578
CaO	0.105	0.113	0.030	0.200
P ₂ O ₅	4.656	4.870	3.894	5.029
MgO	0.351	0.468	0.403	0.504
Fe ₂ O ₃	0.210	0.084	0.352	0.125
S	2.076	2.043	1.868	2.259
Cl	0.701	0.484	0.567	0.672

No work could be found on the composition of the ash of internal organs.

Experimental Part

Description of Animals.

Portions of the following six steers were used in this experiment:

	Breed	Age	Weight	Group
Steer 501	Grade Hereford	47 months	1948 lbs.	I
Steer 512	Grade Hereford (or common)	48 months	1208 lbs.	II
Steer 500	Hereford-Shorthorn	48 months	1009 lbs.	III
Steer 547	Grade Hereford	8 months	455 lbs.	I
Steer 550	Grade Hereford	8 months	325 lbs.	II
Steer 558	Grade Shorthorn	8 months	239 lbs.	III

These animals were procured soon after birth and assigned to groups as soon as possible. Group I was full fed, Group II was fed for maximum growth without the laying on of appreciable fat, Group III was fed for retarded growth--about one-half pound gain daily while yearlings.

Slaughtering

Steers Nos. 501, 512, and 500 were slaughtered in March, 1911. The younger steers, Nos. 547, 550, and 558, were slaughtered in January, 1913, by the same expert from one of the large packing houses. At the time of slaughtering the internal organs were removed, cleaned, weighed, and put away in tight containers. Forty-eight hours later the carcass was cut up, the

weighed soft parts and internal organs ground in an Enterprise meat grinder and mixed thoroly. Samples were then weighed out for the various determinations and composites saved for complete ash analysis. The skeleton samples were ground thru a Mann bone grinder and samples taken.

The same expert cut up all six animals, so uniformity in preparing samples was assured. Only the skeleton composite, composite lean and fat, and internal organs are considered in this paper.

Total Ash

Triplicate samples of about ten grams each were taken from the thoroly mixed fresh samples of lean and fat composite, and composite internal organs and internal fat. These were thoroly dried and gently ignited in tared porcelain crucibles over a Fletcher burner until they were completely carbonized. They were then transferred to a muffle and heated to a dull redness until the combustion of the carbon was complete. They were then cooled and weighed.

Triplicate samples of the composite skeleton amounting to from 30 to 50 grams were ~~also~~ weighed out into large tubes and the moisture and fat determined. The sample was then ground, air dried, and ash determined. The per cent of ash was obtained on an air dry and practically fat-free basis and was then calculated back to the fresh basis. The foregoing operations were necessary in order to get an analysis on a uniform sample.

Table 1. Weight of Cuts (in grams) and Per Cent Ash in Cuts.

Steer	Lean and Fat Composite		Internal Organs and Internal Fat		Composite Skeleton	
	Weight	Ash	Weight	Ash	Weight	Ash
501	548,474	0.485	85,957	0.549	80,602	26.343
512	277,824	0.772	57,864	0.695	80,427	25.619
500	217,860	0.892	49,938	0.790	73,489	23.495
547	94,790	0.888	23,700	0.955	27,596	18.855
550	59,680	0.943	17,694	1.032	23,339	17.856
558	41,560	0.962	11,957	1.025	21,359	16.998

Preparation of the Ash

Preparation of Samples for Ashing

Representative samples amounting to 1000 to 2000 grams of composite skeleton, lean and fat composite, and the composite of the internal organs and internal fat were taken. These were preserved in 95 per cent alcohol in Whitehall-Tatum jars until ready for ashing.

Methods of Ashing

The entire sample was removed from the jars and placed in a large evaporating dish. The dish was placed on a steam bath and the alcohol evaporated off. More alcohol was added and the drying repeated until nearly all of the water was removed and the sample was dry. This operation is especially important in samples containing a large amount of fat because when the sample is heated later, water, when present, causes excessive spattering and loss of material.

Various methods have been used in ash work. Abderhalden¹² used barium hydroxide to aid in the ignition. Calcium acetate has been used to a great extent also. The use of chemicals is not advisable in the kind of work reported here; direct ashing is more practical.

Jarische¹³ in his work on blood used porcelain dishes and claimed that the amount of silica taken up by the ash was negligible.

Lawes and Gilbert² ashed their samples in large platinum

dishes in a muffle. Experience in this laboratory has taught us that this method is extremely expensive and therefore inadvisable because the dishes are ruined.

One of the methods¹⁴ used in this laboratory is as follows: baskets 7-8 centimeters in diameter and having about the same depth were made of platinum wire. Eight or ten ribs of the wire meeting at a common point in the bottom formed the framework. Platinum wire spirals were also wound around this framework, the wires being closer together at the bottom. Some of the material to be ignited was placed in the basket which was suspended from a tripod and was heated with the naked flame. Porcelain casseroles were placed under the baskets to catch the ash and any unignited portions which might fall. The latter were returned to the baskets and the heating continued until the material was completely carbonized and nearly all of the carbon oxidized. As it was not possible to get complete oxidation here the ash was transferred to platinum dishes. Hot water was added and the dishes placed on a water bath. The liquid was decanted off and more water added, this operation being repeated at intervals for several days. This liquid portion was filtered and washed thru ashless filters. The filter papers were added to the residue and the entire mass dried and ignited in platinum. This was then ground and mixed thoroly in an agate mortar. The ignited residue constituted the "insoluble ash"; the filtrate, the "soluble ash".

This method has its advantages and disadvantages. The strongest arguments against it are the danger of volatilization

of the alkali salts when the material is subjected to direct heat and the danger of the absorption of sulphur from the flame by the ash.

A method just as good and probably better is the one worked out recently at this laboratory. The material after drying is not taken from the porcelain dish but the dish and contents are placed on a sand bath and heated gently. The heat is raised gradually to the maximum capacity of the bath and when a greater part of the volatile material has been driven off a torch is applied and the entire mass is ignited. When much fat is present the ignition must not be attempted before enough of the volatile substances have distilled off to have a fairly small amount of liquid left or the material will creep up over the side of the dish. (Samples of lean and fat and internal organs become liquid before the ignition stage) When the flame has burned out and the mass is thoroly charred, a gas flame is directed against the outside of the dish. The carbon deposited on the sides of the dish is burned off in this way and a partly ignited, partly charred mass, is left in the bottom of the container. (Caution: the dish must have been heated at least three hours on the sand bath before the flame can be applied or it will crack.)

The charred mass can be removed easily with a spatula. It is then ground in a 'Wedgewood' mortar and put thru a millimeter sieve. A flat bottomed porcelain dish is then lined with platinum foil. A very good method is to place several pieces of porcelain or porous plate beneath the platinum so that an air space is created. The ground mass is

then placed in this container and put in the front part of a muffle, the back part of which is heated by a multiple Fletcher burner, and is kept below a bright red heat. The sample should be stirred occasionally with a platinum spatula and can be moved nearer the source of heat after the ignition is under way. Care must be taken not to get the dish to a red heat or the alkali chlorides will volatilize. A complete ignition cannot be obtained here or rather it is impractical in samples other than bone, teeth, horns, hoofs, etc. When the material has been reduced in volume to some degree and most of the carbon burned off it is transferred when cool to a casserole by means of hot water and a 'policeman'. The soluble ash and insoluble ash are then prepared as in the first method of ashing. No attempt was made to prepare a complete water soluble ash but the leaching was continued until enough was removed to insure a well ignited insoluble ash. This point is usually determined by the absence of chlorides in the wash water.

It is necessary to leach out the soluble salts when an abundance of them is present because during the ignition they fuse, occlude carbon, and so make it impossible to ignite the carbon unless such a heat is applied that it is certain to cause the volatilization of these salts.

In samples like bone the procedure is modified slightly. The mass in the larger porcelain dish usually does not catch fire but turns brown and gets brittle. It is then ground and passed thru a millimeter sieve and, as these samples are usually

very large, a representative portion is taken and finished. This is accomplished in the platinum lined dish. No soluble ash need be made as the amount of soluble salts present is very small. A total ash is obtained instead.

In the preparation of the soluble ash considerable trouble is given by a white colloidal precipitate which forms. Upon concentrating the solution this often separates out and apparently filters out. However, in most cases it goes thru the filter paper when washed. It is always well in such cases to take aliquots immediately and store until ready for use. Several samples of soluble ash used in this experiment stood for several years in volumetric flasks. The precipitate in these separated out and gave no trouble. Not enough substance could be obtained to prove the identity of this material, but it is believed to be silica.

Analysis

Qualitative Examination of Samples

It is necessary to make a qualitative examination of both the soluble and insoluble ashes before quantitative determinations are attempted. Chlorine, sulphur, carbon dioxide, sodium, potassium, phosphorus, and a trace of silica were found in the soluble ash. Silica, iron, calcium, magnesium, sodium, potassium, and phosphorus were found in the insoluble ash.

Quantitative Examination of the Soluble Ash

It is rather difficult to advise what aliquots to take in making the following determinations as this depends entirely upon the weight of total solids. The aliquots used here are based on total solids weighing from 5 to 8 grams. The plan of analyses given is so devised that when the amount of sample is limited a minimum amount is used.

Total Solids

This determination requires considerable care and is difficult because of the tendency of the residue to absorb moisture rapidly. Weighings must be taken to a constant weight.

Duplicate samples of a 1/25 aliquot were drawn into tared platinum dishes. The solutions were evaporated to dryness on a water bath, heated below redness, and weighed. The heatings and weighings were continued until a constant weight was obtained.

From these data the total weight of soluble ash was cal-

culated. These weights are recorded on tables assigned to each animal.

Chlorine

The residues from the determination of total solids were dissolved in hot water and washed into small beakers, using about one hundred cubic centimeters of water. The solution were made acid with dilute nitric acid and a slight excess of ten per cent silver nitrate solution was added slowly and with constant stirring. The contents of the beakers were boiled, covered with watch glasses and set aside in a dark place for a few hours or over night.

The precipitate of silver chloride was washed into tared Gooch crucibles with hot water containing a small amount of nitric acid. The crucibles were dried in an oven at 110° - 130° C., cooled and weighed.

This determination should give no trouble.

Carbon Dioxide

Duplicate samples of one-fifth aliquot were drawn for this determination. The carbon dioxide was absorbed in a tared U-tube containing soda lime. The absorption train consisted of a condenser, two sulphuric acid drying towers, the U-tube, a calcium chloride drying tower, safety bottle, and suction pump. The sample was placed in a three hundred cubic centimeter Erlenmeyer flask and the flask was attached to the train. Twenty cubic centimeters of 1:1 hydrochloric acid were admitted slowly by means of a dropping funnel,

equipped with a soda lime tube to prevent the entrance of any carbon dioxide from the air. The stop cock of the dropping funnel was then closed and the liquid warmed gently and heated to the boiling point. After boiling several minutes, the flame was taken away and the stop cock of the dropping funnel opened so that the pressure could be equalized. The suction was then started slowly and air was sucked thru the apparatus for forty-five minutes. The gain in weight of the U-tube gave the amount of carbon dioxide present in the aliquot.

A small amount of calcium chloride was placed in the side of the U-tube nearest the pump so that if the dry air coming thru absorbed any moisture from the soda lime the calcium chloride would reabsorb it, thus guarding against possible loss. To get satisfactory results, the "head" of sulphuric acid in the drying towers must not be too large and the manipulation, time of operation, etc., should be the same for all samples.

Silica

The solution from the carbon dioxide determination was evaporated to a smaller bulk and then transferred to platinum dishes, evaporated over a water bath, heated for an hour in an oven at 110° - 120° C., cooled, moistened with strong hydrochloric acid and about 20 cc. of hot water added. This solution was again dehydrated as above but this time the solution was boiled and filtered thru an ashless filter and thoroly washed. The filtrate was then dehydrated again and filtered, and the filtrate saved. The precipitates were combined, dried, ignited in a tared platinum crucible and weighed. The amount of sil-

ica found in the soluble ash was so very small that it was only reported as a trace.

The filtrate from this determination was made up to volume in a graduated flask.

Sulphuric Anhydride

One-eighth aliquots of the above filtrate, amounting to one-fortieth aliquot of the entire solution, were heated to boiling and an excess of ten per cent barium chloride solution was added drop by drop. After boiling for about five minutes the solution was allowed to stand over night. The precipitate was washed with hot water on to an ashless filter of such a texture that it was capable of holding barium sulphate. After washing until free from chlorides the filter paper and contents were transferred to a tared platinum crucible, ignited to whiteness in a muffle, and weighed as barium sulphate.

Good checks can be obtained without any trouble.

Sodium

The filtrate from the sulphuric anhydride determination was heated to boiling and ammonium hydroxide added. A heavy white precipitate of barium phosphate formed. This was allowed to settle and barium hydroxide was added to be sure all of the phosphate was precipitated. If the precipitate is exceedingly large, it is best to filter it off and wash it well. The filtrate is then used for the succeeding operations. If the precipitate is not too large, ammonium carbonate can be added in excess immediately and the solution allowed to stand on the water bath for about one hour.

The solution was then filtered into casseroles and the precipitate washed well with hot water, the filtrate and washings amounting to about 400 c.c. The casserole was placed on a hot plate and allowed to evaporate to a very small volume. It should not go to dryness unless the casserole is a large one as there is danger of spattering. The concentrated solution was transferred to a platinum dish and evaporated to dryness on a water bath. After drying in an oven at 110°C . it was ignited below redness to drive off all of the ammonium salts. The residue was then dissolved in hot water, filtered, and washed into small beakers.

A few drops of ammonium hydroxide were added and ammonium oxalate added in excess. Only a few drops of oxalate need be added, as there is no calcium in the soluble ash and this operation is only done as a precaution. In the insoluble ash this operation is very necessary. The solution was boiled and allowed to stand for several hours. It was then filtered into a weighed platinum dish, a few drops of hydrochloric acid added, evaporated to dryness, dried at 110°C . and heated below a dull red to expel ammonium salts. It was found best to heat the chlorides so that they just fuse. The residue was heated to constant weight and weighed as total sodium and potassium chlorides. After the potash is determined the sodium can be calculated by difference.

If, in this determination, a solution of soluble ash is used from which the silica has not been removed in some cases a light flocculent precipitate of silica appears when the above chlorides are dissolved in hot water. Altho this does

not weigh much it is best to filter, wash, evaporate to dryness with a few drops of hydrochloric acid and redetermine the total chlorides.

Quite a lot of trouble was experienced in this determination and it was often hard to get concordant results. As a result, the determinations had to be checked over. The trouble is due to the enormous quantity of phosphate present. Therefore, care must be taken to remove all of the phosphate and all of the barium used to precipitate the phosphate.

Potash

The residue of total chlorides was dissolved in 50 c.c. of hot water and placed on a water bath. A few drops of hydrochloric acid and platonic chloride in excess were added and the solution was evaporated to a thick paste. About 50 c.c. of eighty per cent alcohol was then added and the mixture allowed to stand for at least an hour. If the potassium chloroplatinate comes down in large plates it is advisable to break this up with an agate pestle so that no foreign material will be occluded.

The precipitate was washed both by decantation and on a tared Gooch crucible with eighty per cent alcohol until the filtrate was colorless. The precipitate was next washed six times with ammonium chloride solution saturated with potassium chloroplatinate and then thoroly with eighty per cent alcohol. It was then dried in an oven at 103° C. and weighed. The potash was calculated from the weight of the chloroplatinate precipitate.

This determination offered very little trouble, especially when enough platonic chloride was added and the precaution as to the condition of the chloroplatinate precipitate observed.

Phosphorus

From the filtrates from the silica determination one-eighth aliquots were drawn and placed in 250 c.c. beakers. The samples were neutralized with ammonium hydroxide and acidified with nitric acid in slight excess. About fifteen grams of solid ammonium nitrate were then added and the sample heated in a water bath up to 65° C. Seventy-five c.c. of ammonium molybdate solution¹⁵ were then added and the solution kept at 65° C. for at least one hour. After cooling, the yellow precipitate was filtered and washed with ten per cent ammonium nitrate solution. The original beaker containing the precipitate was then placed under the funnel and the precipitate dissolved on the filter and washed thru with a hot two per cent solution of ammonia. The solution was next neutralized with hydrochloric acid and made slightly ammoniacal. After cooling, 10 c.c. of magnesia mixture¹⁵ were added drop by drop with vigorous stirring. After fifteen minutes, 10 c.c. of ammonium hydroxide (sp.gr. 0.90) were added and the sample allowed to stand over night.

The solution was filtered thru an ashless filter and the precipitate washed thoroly with a 2.5 per cent solution of ammonia. The filter and contents were dried, transferred to a tared porcelain crucible, ignited in a muffle to whiteness,

and weighed as magnesium pyrophosphate.

Good results were obtained with this method.

Alkalinity of the Soluble Ash

This determination is not of very great value altho it is well to have such data on hand, ^{for future reference.} Aliquots of one-twenty-fifth were taken. The solution was titrated against standard hydrochloric acid (tenth-normal preferred), using phenolphthalein as an indicator. Methyl orange was then added and the titration continued.

Assuming the alkalinity to be due to carbonates, the difference between the two titrations would represent the hydrochloric acid required to neutralize one-half of the carbonate. However, the methyl orange titration so exceeds the phenolphthalein titration, which is supposed to indicate one-half of the carbonates, that it can readily be seen that some other combinations of the alkalies, such as the tertiary phosphates, are present and cause the variations noted.

Quantitative Analysis of the Insoluble Ash

After ignition, the insoluble ash was placed in a tared weighing bottle, heated to 110°C . in an oven, cooled, and the weight determined by difference, (For weight see tables for each animal). It is always best to heat up the ash before taking samples because of its tendency to absorb moisture. For this reason, samples were weighed out by difference.

The amount taken depends entirely upon the supply of ash on hand.

Preliminary investigations showed that in every case the insoluble ash could not be completely dissolved in acids. It was therefore necessary to fuse samples used in various determinations.

Silica

A sample of the insoluble ash was thoroly mixed in an agate mortar with four times its weight of a molecular mixture of sodium and potassium carbonates, then placed in a platinum crucible and fused. After cooling, it was dissolved in hot water, acidified with hydrochloric acid and silica dehydrated as described under the analysis of silica in the soluble ash.

The filtrates from this determination were then combined, made up to volume, and designated as the solution of insoluble ash. Phosphorus, calcium, magnesium, and iron were determined in this solution.

Iron

The method used was the Zimmerman-Reinhardt method as described by Mixer and Dubois¹⁶. One-tenth aliquots of the solution of insoluble ash were transferred to beakers and heated to boiling. Five drops of a stannous chloride solution (one to fifty) was added to the contents of the beakers.

Usually a yellow color which is present disappears. A beaker containing 400 c.c. of hot water, 5 c.c. of saturated solution of mercuric chloride, and 5 c.c. of a preventive* solution was prepared. The sample reduced by the stannous chloride was then washed into this large beaker and titrated immediately with a standard potassium permanganate solution; the end point was reached when the pink color disappeared in about fifteen seconds.

A blank must always be run. This method is a fairly good one, tho much depends on the technique of the operator, as all determinations must be carried out in exactly the same way. The permanganate should be restandardized before using.

Possibly a method⁺ which is worth while considering, especially when small quantities of iron exist, is a colorimetric one. To a definite aliquot, about 10 c.c. of a 20 per cent solution of ammonium sulphocyanate are added and the amount of iron is determined by comparison with a standard solution in a Duboscq colorimeter.

*Preventive solution: Dissolve 160 grams of $MnSO_4$ in 1750 c.c. of water, then add 330 c.c. of phosphoric acid (sp. gr. 1.7 syrupy) and 320 c.c. of sulphuric acid, (sp. gr. 1.82).

Phosphorus

One-tenth aliquots of the solution of insoluble ash were drawn and the analysis carried out as described under the determination of phosphorus in the soluble ash.

 +This method was not used in this work, but is recommended.

Calcium

One-tenth aliquots were drawn here also. The solutions were neutralized with ammonium hydroxide, two grams of ammonium acetate were added and the solution was made strongly acid with acetic acid. The solution was stirred vigorously and allowed to stand one hour, after which it was filtered and washed by decantation on the filter with dilute acetic acid. The precipitate was in most cases very small. If large, the filtrate was removed and replaced by the beaker containing the original precipitate and the precipitate was washed into the beaker with hot dilute hydrochloric acid. A little dilute ammonia was then poured on the filter. The dissolved precipitate was treated with ammonium hydroxide, then acetic acid, and again filtered and washed. The resulting precipitate was rejected. The foregoing operations guard against absorption by the precipitate of any calcium or magnesium.

The filtrates were combined and ammonium oxalate added in excess to precipitate the calcium. The solution was then boiled, set aside for a few hours and then filtered and washed. The precipitate was dissolved thru the filter with hot, dilute hydrochloric acid and the filter paper made alkaline with ammonium hydroxide. The calcium was then reprecipitated, filtered, and washed as before. The combined filtrates were saved for the determination of magnesium.

A hole was punched thru the filter and the precipitate was washed into a volumetric flask with hot water and dilute

sulphuric acid. When cool the solution was made up to volume and aliquots drawn. These were diluted to 200 c.c. with hot water and 5 c.c. of dilute sulphuric acid added. The solution was titrated with standard potassium permanganate solution (preferably N/100) while hot. The temperature must not go below 60° C.

When the calcium content was small, the filter and precipitate were placed in a beaker, heated up with dilute sulphuric acid until solution took place and then titrated immediately. Care must be taken not to let the sulphuric acid char the filter paper. The permanganate must be restandardized before use. No difficulty was experienced with this determination.

Magnesium

The filtrates from the calcium determination were combined and evaporated to about 150 c.c. Ten c.c. of sodium or ammonium phosphate were added and the solution was neutralized with ammonium hydroxide. After stirring vigorously with a "policeman" the precipitate formed. After standing for fifteen minutes, fifteen c. c. of ammonia (sp. gr. 0.90) were added, and the beaker set aside over night. The solution was filtered thru an ashless filter and washed with two and one-half per cent ammonia solution. The filter and precipitate were dried, transferred to a tared porcelain crucible, ignited in the muffle to whiteness, and weighed as magnesium pyrophosphate.

Sodium and Potassium

Tests on the insoluble ash showed the presence of sodium and potassium. It was therefore necessary to run a J. Lawrence Smith fusion.

Sodium

A weighed sample of the ash was mixed in an agate mortar with an equal weight of ammonium chloride. An amount of calcium carbonate equal to about eight times the weight of the sample was weighed out and a small portion placed in the bottom of a J. Lawrence Smith platinum crucible. The greater part of the carbonate was intimately mixed with the sample in the mortar, the remainder was used to rinse the mortar. The mixture was transferred to the crucible, the mortar rinsed with the carbonate and this was used to cover the charge. The covered crucible was heated gently near the middle and gradually toward the bottom until the fumes of ammonia were driven off. It was next heated at a high temperature for one hour, and then cooled. Care was taken to heat only the lower half of the crucible.

The contents of the crucible were removed easily with a few drops of water and a little hydrochloric acid and were placed in a porcelain casserole. The mixture was boiled for about a half hour with several hundred cubic centimeters of water and then filtered into a large Erlenmeyer flask. The solid residue was ground up with a pestle and washed on to the filter paper with hot water. The filtrate was made alka-

line with ammonium hydroxide and then an excess of ammonium carbonate was added and the solution brought to boiling on a hot plate. The contents of the flask were filtered and washed into a large casserole with hot water. The casserole was placed on a hot plate and evaporated to a small bulk and the analysis continued as described under the determination of sodium in the soluble ash. Blanks were run and corrections made for impurities in the reagents.

Potassium

Potassium was determined in the total chlorides from the sodium determination by the method given in the analysis of the soluble ash.

Analysis of Total Ash

No division of the skeleton ash was necessary and the sample was analyzed as total ash. The methods used were those used in the analysis of the soluble and insoluble ashes.

Calculations

Note: Totals which have been marked by the symbol * indicate that the total should be 100 per cent but differ in the third decimal place because of figures carried forward or dropped.

No mention need be made of the methods of calculation of the ordinary quantitative analyses used in this work, but it is of value to trace out the methods of calculation used in preparing certain tables.

In order that the methods may be followed more closely, a concrete example will be used. Noting table I, Steer No. 501, a certain analysis is recorded for both the soluble and insoluble ash. Since all the constituents except chlorine are reported as oxide, the oxygen equivalent of chlorine must be deducted to get the correct total. The constituents do not total up to 100 per cent so in order that comparison might be made the error is distributed equally among the individual constituents in each sample of ash. This is done by dividing each per cent of constituent by 102.367 in the case of the soluble ash and 101.917 in the case of the insoluble ash and then multiplying by 100. This gives the soluble ash and insoluble ash calculated to 100 per cent. In order to get an analysis of the total ash, these two results must be combined. From the actual weights of the two ashes, it was found that 75.7 per cent of the constituents in the soluble ash were contained

in the total ash, therefore this percentage for the insoluble ash would be 100 per cent minus 75.7 per cent, or 24.3 per cent. Multiplying the per cent of each constituent in each ash by the corresponding factor, the total ash calculated to 100 per cent is obtained, the error being distributed in the ash to which it belongs.

Thinking that probably results calculated on a silica free basis would be of value, the per cent of silica was subtracted from 100 per cent and this new value divided into the total ash percentages and multiplied by 100. The new figures were percentages figured on a silica free basis.

In calculating to a fresh basis, the total ash figures are multiplied by the per cent of ash in the sample and summarized by multiplying by ten and reporting as parts per thousand.

Because of the fact that the silica in the internal organs may be extraneous, this sample was calculated to a fresh basis silica free. This was done by obtaining the exact weight of each mineral constituent in the sample by multiplying the total weight of the part by the per cent of constituents in the part. The actual weight of silica was then subtracted from the total weight of the internal organs and a silica free weight was obtained which when divided into the actual weight of the mineral constituents gave the per cent in the fresh material silica free. This was reported in parts per thousand.

The same plan of calculation was used for the work on internal organs. The skeleton was analyzed as total ash and

since no silica was present, it was only necessary to calculate to 100 per cent and then to the fresh basis.

In calculating Tables XIX and XXIV, in order to get the constituents on a fat free basis, the amount in grams of each mineral constituent in the fresh composite was figured. The weight of the soluble fat was then subtracted from each composite and this new weight formed the divisor. The quotients represented the amounts in the fat free part. This can all be calculated in one operation by obtaining the ratio of fat free substance to total substance. When the amount of constituents in Tables XVIII and XXII are multiplied by this ratio the figures in Table XIX and XXIV are obtained.

DiscussionEffect of AgeAsh

Considering first the ash of the composite skeleton, Table XIV, (all summary tables, etc., are included in the Appendix) the sodium is consistently lower in the older animals than it is in the young animals of the same group. Moreover, the average amount of sodium in the ash of the skeleton of the older steers as compared with that in the younger steers would indicate that the older an animal, the less its sodium content.

The same conclusions can be drawn when referring to potassium, as the difference is fully as marked.

Calcium, the largest constituent of bone, also shows a very significant change. It seems that in steers the calcium content increases with age. The same observation with regard to rabbits was made by Weiske⁵ and Wildt³.

The per cent of phosphorus is surprisingly constant. There is no change due to age, altho it would seem from comparing the data on Steers 500 and 558 that in thin animals the phosphorus decreased. Weiske and Wildt differ slightly on this point, the former reporting the phosphorus content as constant in rabbit bones, the latter reporting a decrease.

Iron, altho present in all six animals in seemingly large quantities, is shown to be present in a greater amount

in the bones of the younger steers.

Moreover, the four year old animals seem to have a smaller per cent of magnesium than the young animals of the same group. The general average would indicate this also.

Too much stress must not be laid on the determination of carbon dioxide as there is no definite knowledge as to its value. It may, however, indicate the amount of acid radical lacking. It is an interesting fact that the carbon dioxide content is higher in the skeleton of the more mature animals, a fact which may be due to the increased alkali content. This same point was noted by Weiske and his assistants.

The same view used in interpreting the data on carbon dioxide might be applied in the case of chlorine and sulphur since Hogan and Vanatta¹⁴ of this Station have shown that there is a varying loss due to ashing. Therefore, the results reported here are per cents remaining in the ash. Notwithstanding, one cannot ignore the fact that age seems to have an effect on this amount in the cuts analyzed. On inspection, it will be noted that the skeleton ash of the older steers has less chlorine and sulphur fixed as salts.

Table XVI shows the composition of the ash of the composite lean and fat. It is more logical to use this table rather than the one calculated to a silica free basis since there is nothing to indicate conclusively that the silica in this cut is extraneous.

In this ash, generally speaking, the sodium increases

with age while the potassium decreases even tho the Group II animals do not show this. In this connection, it might be said that there are many anomalies in the data presented here that cannot be explained until more complete data are offered on the subject. For this reason it seems desirable to consider the general relations only.

The per cent calcium when referred to corresponding animals shows a decrease with increasing age. The figures on phosphorus indicate a peculiar relation in which the Group I animals show a decrease with age, Group II no change, and Group III an increase, while the numerical averages of the three old animals and of the three young animals are very nearly alike. To come to a definite conclusion on this constituent would be impossible.

Disregarding the large iron content of Steer 501, which probably came from some outside source, it can be said that in the ash of lean and fat iron increases with age. This is contrary to the data on skeleton ash.

The magnesium content agrees with that in the skeleton, decreasing toward maturity.

As to the presence of silica, no importance is attached since there is no evidence to indicate that it does or does not belong in the ash.

Here again, as in the skeleton, the percentage of carbon dioxide increases with age. The sulphur increases toward maturity, but the chlorine decreases.

In the analysis of the ash of the composite internal organs and internal fat, an exceedingly large per cent of silica was found. It is hard to believe that the organs themselves contained such an amount of silica so a probable source of error was investigated. An animal often eats a large quantity of dirt and sand and this is often found in its intestines and stomach. In cleaning these before analysis it is nearly impossible to clean them thoroly even with water, so some of the silica must certainly have been left in.

Since this seems to be the most logical explanation, it is not fair to consider the silica as a constituent of the organs and therefore it should be eliminated. Table XXI gives the mineral composition of the ash of the internal organs and internal fat calculated to a silica free basis and it is from this table that the following observations are made.

As in the ash of the lean and fat, animals of greater maturity show an increase in sodium content but a decrease in potassium, altho the difference in the latter is not striking.

Here too, calcium shows a marked decrease with age. Magnesium, however, remains practically constant.

An apparent decrease in phosphorus is shown in the older animals. The per cent of iron is less also.

Another similarity to the ash of the skeleton and of the

lean and fat is shown by the fact that the carbon dioxide and chlorine decrease towards maturity. The sulphur, however, increases.

Effect of Age.

Composition of the Fresh Material

In referring the mineral constituents of the ash back to the fresh material, there is a factor which is worthy of careful consideration, namely, the fat content. A brief inspection of Tables XVIII and XXII will show that there is a gradual increase in the amount of constituents from Group I to Group III, especially in the older animals where the per cent of fat is larger. In other words, the fatter the animal the smaller the amount of its mineral matter figured on a fresh basis. This fact is very evident here because the samples are composites containing fat. The fat in bone would not cause such a big variation because the difference in the amount present in each sample is very small. The effect of the fat is so marked in the other composites that there is a tendency to attribute all differences to it. Therefore, the composites of lean and fat and of internal organs were calculated to a fat free basis, the fat being ether soluble fat which would contain no ash. These new tables were then used in the subsequent comparisons.

The most important constituents of the skeleton, calcium and phosphorus, show a very decided increase with age when referred to the fresh bone. The amount of sodium and potas-

sium decreases. The Group III animals show an exception to this. Magnesium is a little higher in the older steers, Group III having practically a constant content of this constituent. The amount of iron, disregarding Group I, shows no effect of age.

The determinations of carbon dioxide, chlorine, and sulphur will not be discussed because these values do not represent the true amount in the fresh material.

Referring to Table XIX, the results of the analysis of the fresh lean and fat, with the exception of calcium and magnesium, fail to show any definite relations. These two constituents decrease with maturity. There seems to be a tendency toward an increase in sodium with age, no change in potassium, and an increase in iron. Nothing can be concluded from the phosphorus data.

Because of the silica present in the internal organs and coming from an outside source, the fresh material was calculated to a silica free basis (Table XXIII). By comparing this table with Table XXII it will be noted that on the fresh basis the silica hardly changed the figures.

Calculated free from fat, the data on the internal organs offered better opportunity for comparison than did the lean and fat (Table XXIV). Calcium, phosphorus, and magnesium show clearly a diminution with age. Potassium and iron also show this, while sodium indicates an increase.

Effect of Condition

In considering the effect of the condition of the animal on its mineral composition, the determinations of carbon dioxide, chlorine, and sulphur are of little value because they only represent that found in the ash and are influenced by the varying losses of these constituents during ignition. They will not be discussed in this section. The results on iron vary so much that nothing definite is shown.

Ash

Referring back to Table XIV showing the analysis of the ash of the composite skeleton, it can readily be seen that condition has no effect on the two most important constituents of bone ash, calcium and phosphorus. Nothing definite is shown in regard to potassium and magnesium, altho there seems to be no effect on the latter in the young animals. A peculiar relation is shown in the case of sodium. From the data recorded it seems that in the thinner four year old animals the skeleton ash contained more sodium, while in the eight month old steers it is just the opposite. This difference cannot be explained and may be due to some experimental error.

A study of Table XVI, the composition of the ash of the lean and fat, did not bring out any striking results. The per cents of calcium and magnesium are fairly constant as shown by small differences in the three groups. Sodium fails to show any relation, while potassium and phosphorus

have a tendency toward constant values.

The analysis of ash of the internal organs and internal fat shows more uniformity than that of the lean and fat. Noting the composition of the ash of the internal organs and internal fat, Table XXI, both the data on the young and the old animals point out that the fatter the steer the more calcium and magnesium in his internal organs. Sodium, however, is less in the fatter animal. The potassium appears to be constant, but the results on phosphorus bear no definite relation to one another.

Composition of the Fresh Material

The tables used in the following discussion are those used in the section on 'Effect of Age' and are calculated to a fat free basis.

The relation of the calcium and phosphorus content of the fresh skeleton composite (Table XV) to the condition of the animal is shown very nicely. These constituents are found in smaller quantities in the retarded growth animals, Group III, than in the full fed animals, Group I. This is apparent in both the young and old steers. The four year old animals give rather obscure results on sodium, potassium, and magnesium. The amount of sodium in the animals eight months of age seems to be less in the thin steer. The values for magnesium are the same. The relation of the condition to the amount of potassium is not shown very well.

The irregularity of the data on the lean and fat again shows up in the composition of the fresh sample. From a general survey of the figures (Table XIX) on animals of both

ages, one might say that there is a tendency for the sodium and potassium content to be constant. This would also apply to calcium, phosphorus, and magnesium.

The effect of condition on the amount of mineral constituents in the composite internal organs and internal fat is fairly well defined in Table XXIV. The full fed animals contain less sodium than the retarded growth animals. In the younger animals the results on potassium seem to indicate the reverse, while in the older animals there is hardly any change. The amounts of both calcium and magnesium are less in the thin animals while the phosphorus seems to lean toward a constant value.

General Relations

There are a few general conclusions which arise from these data on the mineral composition of the ash of the three samples studied which are very interesting.

There is more sodium than potassium in bone ash. Furthermore, at least half of the skeleton ash is lime.

The principal constituents of the ash of lean and fat are sodium, potassium, and phosphorus. The potash content is much greater than the content of sodium oxide, being three times as large in most cases. A brief study of the data on the six samples of ash of lean and fat brings out a very interesting fact. There seems to be a definite relation between the per cent of lime and magnesia, the ratio being one part of lime to one and a half parts of magnesia.

In the ash of the internal organs the potash exceeds the sodium oxide by a comparatively small amount. The calcium in this sample also bears a relation to the magnesium, but the ratio is different for different ages. In the animals four years old the ratio is one part of magnesia to 1.19 parts of lime; in the eight months old steers it is 1:1.44.

SummaryEffect of Age

The sodium content of the ash of the skeleton decreases with increasing age, while in the ash of the lean and fat and internal organs and internal fat it increases. Referred to the fresh basis*, this constituent in these three samples shows the same relation as it did in the ash.

That potassium is less the older the animal, is shown in the ash of all three parts examined, the fact being more clearly defined in the skeleton. The same is true when referred to the fresh sample except in the lean and fat where a constant ratio is indicated whether the animal be young or old.

In both the skeleton ash and the fresh skeleton, the calcium content increases toward maturity. The other parts and ash of these parts show a decrease.

Age evidently has no effect upon the per cent of phosphorus in the bone ash, but a decided increase in the older animals is noted in the fresh composite. The mature animals have less phosphorus in their internal organs and in the ash of their internal organs. No conclusion can be drawn from the data on this constituent in the lean and fat on either basis.

*The comparisons of the fresh parts of lean and fat and internal organs and internal fat are all made on a fat-free basis.

Magnesium shows a decrease in the ash of the skeleton and lean and fat of the older animals, while in the ash of the internal organs it is constant. In the fresh skeleton it shows a slight increase and a decrease in the other parts.

The older animals have a smaller per cent of iron in their bone ash, but when referred to the fresh bone no difference is shown. In their internal organs they also have less iron, which is apparent in the fresh material as well. The per cent of this constituent is larger, however, in the ash of the fresh sample of lean and fat of the mature animals.

There is a marked increase with age in the carbon dioxide content of all ash samples.

There is less chlorine in each of the three ash samples of the older animals, less sulphur in the skeleton of the older animals, but more in the other two composites analyzed.

With the possible exception of the phosphorus and magnesium of the skeleton, the conclusions drawn from the ash with reference to age also apply to the fresh material.

Effect of Condition

The data on sodium in the skeleton does not satisfactorily show any relation to condition. This is also true of the sodium content of the lean and fat. The fat animals have less sodium in their internal organs and in the ash of this part than the thinner animals.

Nothing definite can be derived from the potassium content of bones. Condition has no effect on this constituent in the lean and fat sample, or in the ash of the internal organs. It is only in the fresh sample of the internal organs of the eight months old steers that an increase in potassium in the full fed animal is shown.

The condition of the animal has no effect on the calcium content of the skeleton and lean and fat ash. The fatter the steer the more calcium in his internal organs. In the fresh skeleton, the full fed steers show a decided increase in calcium over the retarded growth animals. This is also true in the internal organs. There is no change in the lean and fat.

The skeleton ash shows no difference in the per cent phosphorus due to a change in condition, but the fresh skeleton indicates an increase in the full fed steers. Neither sample of lean and fat shows any effect. The data on the phosphorus content in the ash of the internal organs are rather unsatisfactory, but the fresh composite shows a constant amount.

Nothing definite can be said of the effect on magnesium in the skeleton except in the fresh composite of the younger animals where a constant value is noted. Condition has no effect on this constituent in the lean and fat. The fat animals, however, have more magnesium in their internal organs and internal fat.

From these data one of the observations which stands out is the fact that condition seems to have no effect on the mineral composition of the lean and fat.

Additional Observations

There is more magnesium than calcium in the lean and fat of steers. This seems to be in a rather definite ratio of 1:1.5.

There is more sodium than potassium in the skeleton of steers.

There is more potassium than sodium in both the lean and fat and the internal organs, altho in the latter the difference is not very large.

APPENDIX

Tables I to XXVI

Bibliography

Table I. Composition of Ash of Lean and Fat CompositeSteer No. 501

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00485 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	19.550	1.530	19.098	1.501	14.822	0.0719	15.163
K ₂ O	36.818	21.453	35.967	21.049	32.342	0.1569	33.086
CaO	none	7.411	none	7.272	1.767	0.0086	1.808
P ₂ O ₅	35.286	42.139	34.470	41.346	36.141	0.1753	36.973
Fe ₂ O ₃	none	8.463	none	8.304	2.018	0.0098	2.064
MgO	none	11.485	none	11.269	2.738	0.0133	2.801
SiO ₂	trace	9.436	trace	9.259	2.250	0.0109	-----
CO ₂	4.720	none	4.611	none	3.491	0.0169	3.571
SO ₃	1.114	none	1.088	none	0.824	0.0040	0.843
Cl	6.301	none	6.155	none	4.659	0.0226	4.766
Total	103.789	101.917	101.389	100.000	101.052	0.4902	101.075
O=Cl	1.422	-----	1.389	-----	1.051	0.0051	1.075
Total	102.367	101.917	100.000	100.000	100.000*	0.4851	100.000

Weight of Soluble Ash -- 4.0788 grams
 Weight of Insoluble Ash -- 1.3090 grams

Factor - Soluble Ash to Total Ash -- 0.7570
 Factor - Insoluble Ash to Total Ash -- 0.2430

*See Section on Calculations.

Table II. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 501

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00549 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	25.909	4.250	25.496	4.133	19.699	0.1081	20.600
K ₂ O	26.865	16.882	26.437	16.416	23.719	0.1302	24.804
CaO	none	12.502	none	12.157	3.298	0.0181	3.449
P ₂ O ₅	40.656	40.703	40.009	39.581	39.893	0.2190	41.719
Fe ₂ O ₃	none	0.953	none	0.927	0.251	0.0014	0.262
MgO	none	10.959	none	10.657	2.891	0.0159	3.023
SiO ₂	trace	16.587	trace	16.130	4.376	0.0240	-----
CO ₂	4.480	none	4.409	none	3.213	0.0176	3.360
SO ₃	2.161	none	2.127	none	1.550	0.0085	1.621
Cl	1.998	none	1.966	none	1.433	0.0079	1.499
Total	102.069	102.836	100.444	100.001	100.324	0.5507	100.337
O=Cl	0.451	-----	0.444	-----	0.324	0.0018	0.339
Total	101.618	102.836	100.000	100.000*	100.000	0.5489	100.000*

Weight of Soluble Ash -- 3.4930 grams

Weight of Insoluble Ash -- 1.3004 grams

Factor - Soluble Ash to Total Ash -- 0.7287

Factor - Insoluble Ash to Total Ash -- 0.2713

*See Section on Calculations.

Table III. Composition of Ash of Lean and Fat CompositeSteer No. 512

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00772 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	14.841	1.679	14.601	1.652	11.784	0.0910	11.910
K ₂ O	40.077	22.014	39.428	21.656	35.562	0.2745	35.941
CaO	none	8.164	none	8.031	1.747	0.0135	1.766
P ₂ O ₅	36.530	47.906	35.938	47.127	38.371	0.2962	38.780
Fe ₂ O ₃	none	4.099	none	4.032	0.877	0.0068	0.886
MgO	none	12.866	none	12.657	2.753	0.0213	2.782
SiO ₂	trace	4.926	trace	4.846	1.054	0.0081	-----
CO ₂	3.107	none	3.057	none	2.392	0.0185	2.417
SO ₃	1.241	none	1.221	none	0.955	0.0074	0.965
Cl	7.556	none	7.434	none	5.817	0.0449	5.879
Total	103.352	101.654	101.679	100.001	101.312	0.7822	101.326
O=Cl	1.705	-----	1.677	-----	1.312	0.0101	1.326
Total	101.647	101.654	100.000*	100.000*	100.000	0.7721	100.000

Weight of Soluble Ash -- 8.0713 grams

Weight of Insoluble Ash -- 2.2430 grams

Factor - Soluble Ash to Total Ash -- 0.7825

Factor - Insoluble Ash to Total Ash -- 0.2175

*See Section on Calculations.

Table IV. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 512

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00695 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	26.108	5.015	25.367	4.892	20.764	0.1443	21.404
K ₂ O	25.495	14.295	24.772	13.946	22.338	0.1553	23.026
CaO	none	13.224	none	12.901	2.900	0.0202	2.989
P ₂ O ₅	42.359	41.245	41.157	40.237	40.950	0.2846	42.212
Fe ₂ O ₃	none	3.504	none	3.418	0.768	0.0053	0.792
MgO	none	11.593	none	11.310	2.542	0.0177	2.620
SiO ₂	trace	13.629	trace	13.296	2.989	0.0208	-----
CO ₂	4.570	none	4.440	none	3.442	0.0239	3.548
SO ₃	2.621	none	2.547	none	1.974	0.0137	2.035
Cl	2.282	none	2.217	none	1.719	0.0119	1.772
Total	103.435	102.505	100.500	100.000	100.386	0.6977	100.398
O=Cl	0.515	-----	0.500	-----	0.388	0.0027	0.400
Total	102.920	102.505	100.000	100.000	100.000*	0.6950	100.000*

Weight of Soluble Ash -- 3.0360 grams

Weight of Insoluble Ash -- 0.8806 grams

Factor - Soluble Ash to Total Ash -- 0.7752

Factor - Insoluble Ash to Total Ash -- 0.2248

*See Section on Calculations.

Table V. Composition of Ash of Lean and Fat CompositeSteer No. 500

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00892 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	17.740	2.102	17.474	2.036	13.054	0.1164	13.270
K ₂ O	40.118	24.982	39.517	24.195	35.130	0.3134	35.711
CaO	none	7.366	none	7.134	2.042	0.0182	2.076
P ₂ O ₅	36.061	48.569	35.520	47.039	38.818	0.3463	39.460
Fe ₂ O ₃	none	3.325	none	3.220	0.922	0.0082	0.937
MgO	none	11.035	none	10.687	3.060	0.0273	3.111
SiO ₂	trace	5.873	trace	5.688	1.628	0.0145	-----
CO ₂	4.225	none	4.162	none	2.970	0.0265	3.019
SO ₂	1.403	none	1.382	none	0.986	0.0088	1.002
Cl	2.551	none	2.513	none	1.794	0.0160	1.824
Total	102.098	103.252	100.568	99.999	100.404	0.8956	100.410
O=Cl	0.576	-----	0.567	-----	0.405	0.0036	0.412
Total	101.522	103.252	100.000*	100.000*	100.000*	0.8920	100.000*

Weight of Soluble Ash -- 2.9825 grams
 Weight of Insoluble Ash -- 1.1963 grams

Factor - Soluble Ash to Total Ash -- 0.7137
 Factor - Insoluble Ash to Total Ash -- 0.2863

*See Section on Calculations.

Table VI. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 500

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00790 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	26.744	7.325	26.154	7.350	22.011	0.1739	23.013
K ₂ O	26.729	12.609	26.139	12.653	23.168	0.1830	24.223
CaO	none	12.415	none	12.458	2.744	0.0217	2.869
P ₂ O ₅	40.073	36.380	39.189	36.506	38.598	0.3049	40.355
Fe ₂ O ₃	none	1.557	none	1.562	0.344	0.0027	0.360
MgO	none	9.666	none	9.700	2.137	0.0169	2.234
SiO ₂	trace	19.702	trace	19.770	4.355	0.0344	-----
CO ₂	3.158	none	3.088	none	2.408	0.0190	2.518
SO ₃	2.710	none	2.650	none	2.066	0.0163	2.160
Cl	3.670	none	3.589	none	2.798	0.0221	2.925
Total	103.084	99.654	100.809	99.999	100.629	0.7949	100.657
O=Cl	0.828	-----	0.810	-----	0.632	0.0050	0.661
Total	102.256	99.654	100.000*	100.000*	100.000*	0.7899	100.000*

Weight of Soluble Ash -- 3.7210 grams
 Weight of Insoluble Ash -- 1.0512 grams

Factor - Soluble Ash to Total Ash -- 0.7797
 Factor - Insoluble Ash to Total Ash -- 0.2203

*See Section on Calculations.

Table VII. Composition of Ash of Lean and Fat CompositeSteer No. 547

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00888 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	15.448	1.159	14.925	1.162	11.563	0.1027	11.617
K ₂ O	41.958	22.533	40.537	22.589	36.152	0.3210	36.322
CaO	none	8.742	none	8.764	2.141	0.0190	2.151
P ₂ O ₅	35.086	49.686	33.898	49.809	37.785	0.3355	37.963
Fe ₂ O ₃	none	2.354	none	2.360	0.577	0.0051	0.580
MgO	none	13.369	none	13.402	3.274	0.0291	3.289
SiO ₂	trace	1.911	trace	1.916	0.468	0.0042	-----
CO ₂	3.547	none	3.427	none	2.590	0.0230	2.602
SO ₃	1.046	none	1.011	none	0.764	0.0068	0.768
Cl	8.290	none	8.009	none	6.052	0.0537	6.080
Total	105.375	99.754	101.807	100.002	101.366	0.9001	101.372
O=Cl	1.870	-----	1.807	-----	1.366	0.0121	1.372
Total	103.505	99.754	100.000	100.000*	100.000	0.8880	100.000

Weight of Soluble Ash -- 6.6615 grams

Weight of Insoluble Ash -- 2.1530 grams

Factor - Soluble Ash to Total Ash -- 0.7557

Factor - Insoluble Ash to Total Ash -- 0.2443

*See Section on Calculations.

Table VIII. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 547

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00955 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	21.215	1.222	21.424	1.255	14.821	0.1415	15.440
K ₂ O	28.106	14.940	28.383	15.347	24.115	0.2303	25.123
CaO	none	12.336	none	12.672	4.149	0.0396	4.322
P ₂ O ₅	42.449	45.055	42.867	46.281	43.984	0.4200	45.822
Fe ₂ O ₃	none	3.421	none	3.514	1.150	0.0110	1.198
MgO	none	8.449	none	8.679	2.842	0.0271	2.961
SiO ₂	trace	11.928	trace	12.253	4.012	0.0383	-----
CO ₂	0.684	none	0.691	none	0.465	0.0044	0.484
SO ₃	0.880	none	0.889	none	0.598	0.0057	0.623
Cl	7.348	none	7.420	none	4.991	0.0477	5.200
Total	100.682	97.351	101.674	100.001	101.127	0.9656	101.173
O=Cl	1.658	-----	1.674	-----	1.126	0.0108	1.173
Total	99.024	97.351	100.000	100.000*	100.000*	0.9548	100.000

Weight of Soluble Ash -- 7.6430 grams
 Weight of Insoluble Ash -- 3.7204 grams

Factor - Soluble Ash to Total Ash -- 0.6726
 Factor - Insoluble Ash to Total Ash -- 0.3274

*See Section on Calculations.

Table IX. Composition of Ash of Lean and Fat CompositeSteer No. 550

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00943 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	16.371	2.199	15.888	2.156	12.547	0.1183	12.547
K ₂ O	40.238	23.650	39.042	23.190	35.185	0.3318	35.185
CaO	none	9.928	none	9.735	2.369	0.0223	2.369
P ₂ O ₅	36.514	48.638	35.437	47.692	38.418	0.3623	38.418
Fe ₂ O ₃	none	1.911	none	1.874	0.456	0.0043	0.456
MgO	none	15.657	none	15.353	3.735	0.0352	3.735
SiO ₂	trace	trace	trace	trace	trace	trace	-----
CO ₂	3.104	none	3.012	none	2.279	0.0215	2.279
SO ₃	0.933	none	0.905	none	0.685	0.0065	0.685
Cl	7.606	none	7.382	none	5.586	0.0527	5.586
Total	104.756	101.983	101.666	100.000	101.260	0.9549	101.260
O=Cl	1.716	-----	1.665	-----	1.260	0.0119	1.260
Total	103.040	101.983	100.000*	100.000	100.000	0.9430	100.000

Weight of Soluble Ash -- 7.0650 grams
 Weight of Insoluble Ash -- 2.2712 grams

Factor - Soluble Ash to Total Ash -- 0.7567
 Factor - Insoluble Ash to Total Ash -- 0.2433

*See Section on Calculations.

Table X. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 550

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.01032 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	26.837	1.736	26.209	1.758	17.318	0.1787	19.869
K ₂ O	27.048	13.107	26.415	13.274	21.637	0.2233	24.834
CaO	none	8.938	none	9.052	3.291	0.0340	3.776
P ₂ O ₅	39.774	31.216	38.843	31.614	36.215	0.3737	41.550
Fe ₂ O ₃	none	2.511	none	2.543	0.925	0.0095	1.061
MgO	none	6.364	none	6.445	2.343	0.0242	2.688
SiO ₂	trace	34.870	trace	35.314	12.840	0.1325	-----
CO ₂	1.213	none	1.185	none	0.754	0.0078	0.865
SO ₃	1.208	none	1.180	none	0.751	0.0078	0.862
Cl	8.157	none	7.966	none	5.070	0.0523	5.817
Total	104.237	98.742	101.798	100.000	101.144	1.0438	101.312
O=Cl	1.840	-----	1.797	-----	1.144	0.0118	1.313
Total	102.397	98.742	100.000*	100.000	100.000	1.0320	100.000*

Weight of Soluble Ash -- 5.7930 grams

Weight of Insoluble Ash -- 3.3092 grams

Factor - Soluble Ash to Total Ash -- 0.6364

Factor - Insoluble Ash to Total Ash -- 0.3636

*See Section on Calculations.

Table XI. Composition of Ash of Lean and Fat CompositeSteer No. 558

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.00962 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	14.875	2.402	14.593	2.453	11.562	0.1112	11.644
K ₂ O	40.658	24.565	39.887	25.085	36.191	0.3482	36.448
CaO	none	8.785	none	8.971	2.240	0.0215	2.256
P ₂ O ₅	35.985	44.642	35.302	45.587	37.870	0.3643	38.138
Fe ₂ O ₃	none	1.822	none	1.861	0.465	0.0045	0.468
MgO	none	12.949	none	13.223	3.302	0.0318	3.325
SiO ₂	trace	2.761	trace	2.819	0.704	0.0068	-----
CO ₂	3.410	none	3.345	none	2.510	0.0241	2.528
SO ₃	1.017	none	0.998	none	0.749	0.0072	0.754
Cl	7.734	none	7.587	none	5.693	0.0548	5.733
Total	103.679	97.926	101.712	99.999	101.286	0.9744	101.294
O=Cl	1.745	----	1.712	-----	1.285	0.0124	1.294
Total	101.934	97.926	100.000	100.000*	100.000*	0.9620	100.000

Weight of Soluble Ash -- 7.7290 grams
 Weight of Insoluble Ash -- 2.5724 grams

Factor - Soluble Ash to Total Ash -- 0.7503
 Factor - Insoluble Ash to Total Ash -- 0.2497

*See Section on Calculations.

Table XII. Composition of Ash of Internal Organs and InternalFat CompositeSteer No. 558

	Soluble Ash Per Cent	Insoluble Ash Per Cent	Soluble Ash Calculated to 100 Per Cent	Insoluble Ash Calculated to 100 Per Cent	Total Ash Calculated to 100 Per Cent	Total Ash Calculated on Fresh Basis Factor 0.01025 Per Cent	Total Ash Calculated on Silica Free Basis Per Cent
Na ₂ O	26.701	2.298	26.357	2.264	19.626	0.2012	20.248
K ₂ O	26.262	18.645	25.924	18.368	23.813	0.2441	24.568
CaO	none	12.082	none	11.903	3.326	0.0341	3.431
P ₂ O ₅	39.884	45.289	39.370	44.617	40.836	0.4186	42.130
Fe ₂ O ₃	none	3.821	none	3.764	1.052	0.0108	1.085
MgO	none	8.211	none	8.089	2.260	0.0232	2.332
SiO ₂	trace	11.161	trace	10.995	3.072	0.0315	-----
CO ₂	0.627	none	0.619	none	0.446	0.0046	0.460
SO ₃	0.882	none	0.871	none	0.628	0.0064	0.648
Cl	8.974	none	8.858	none	6.383	0.0654	6.585
Total	103.330	101.507	101.999	100.000	101.442	1.0399	101.487
O=Cl	2.025	-----	1.999	-----	1.440	0.0148	1.486
Total	101.305	101.507	100.000	100.000	100.000*	1.0251	100.000*

Weight of Soluble Ash -- 9.4940 grams

Weight of Insoluble Ash -- 3.6820 grams

Factor - Soluble Ash to Total Ash -- 0.7206

Factor - Insoluble Ash to Total Ash -- 0.2794

*See Section on Calculations.

Table XIII. Composition of Total Ash of Skeleton.

	Steer No. 501 Per Cent	Steer No. 512 Per Cent	Steer No. 500 Per Cent	Steer No. 547 Per Cent	Steer No. 550 Per Cent	Steer No. 558 Per Cent
Na ₂ O	1.790	2.323	2.325	3.902	3.682	2.728
K ₂ O	0.322	0.351	0.604	0.815	0.943	0.744
CaO	53.615	53.769	52.900	50.535	50.565	50.356
P ₂ O ₅	41.587	41.350	40.575	40.437	41.415	40.669
Fe ₂ O ₃	0.617	0.867	0.457	1.105	1.180	0.625
MgO	1.452	1.731	1.195	1.661	1.719	1.699
SiO ₂	trace	trace	trace	trace	trace	trace
CO ₂	1.148	1.112	1.790	0.540	trace	0.491
SO ₃	0.658	0.764	0.940	0.969	1.137	0.890
Cl	0.434	0.410	0.396	0.574	0.584	0.700
Total	101.623	102.677	101.182	100.538	101.225	98.902
O=Cl	0.098	0.092	0.089	0.129	0.132	0.158
Total	101.525	102.585	101.093	100.409	101.093	98.744

Table XIV. Composition of Total Ash of Skeleton Calculated
to 100 Per Cent.

Na ₂ O	1.763	2.264	2.300	3.886	3.642	2.763
K ₂ O	0.317	0.342	0.597	0.812	0.933	0.753
CaO	52.810	52.414	52.328	50.329	50.018	50.997
P ₂ O ₅	40.962	40.308	40.136	40.272	40.967	41.186
Fe ₂ O ₃	0.608	0.845	0.452	1.101	1.167	0.633
MgO	1.430	1.687	1.182	1.654	1.700	1.721
SiO ₂	trace	trace	trace	trace	trace	trace
CO ₂	1.131	1.084	1.771	0.538	trace	0.497
SO ₃	0.648	0.745	0.930	0.965	1.125	0.901
Cl	0.427	0.400	0.392	0.572	0.578	0.709
Total	100.096	100.089	100.088	100.129	100.130	100.160
O=Cl	0.096	0.090	0.088	0.128	0.131	0.160
Total	100.000	100.000*	100.000	100.000*	100.000*	100.000

*See section on Calculations.

Table XV. SummaryComposition of Total Ash of Composite SkeletonCalculated to Fresh BasisParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
<u>Factor</u>	<u>2.6343</u>	<u>2.5619</u>	<u>2.3495</u>	<u>1.8855</u>	<u>1.7856</u>	<u>1.6998</u>
Na ₂ O	4.644	5.800	5.404	7.327	6.503	4.697
K ₂ O	0.835	0.876	1.403	1.531	1.666	1.280
CaO	139.117	134.279	122.945	94.895	89.312	86.685
P ₂ O ₅	107.906	103.265	94.300	75.933	73.151	70.008
Fe ₂ O ₃	1.602	2.165	1.062	2.076	2.084	1.076
MgO	3.767	4.322	2.777	3.119	3.036	2.925
SiO ₂	trace	trace	trace	trace	trace	trace
CO ₂	2.979	2.777	4.161	1.014	trace	0.845
SO ₃	1.707	1.909	2.185	1.820	2.009	1.532
Cl	1.125	1.025	0.921	1.079	1.032	1.205
<u>Total</u>	<u>263.682</u>	<u>256.418</u>	<u>235.158</u>	<u>188.794</u>	<u>178.793</u>	<u>170.253</u>
O=Cl	0.253	0.231	0.207	0.241	0.234	0.272
<u>Total</u>	<u>263.429</u>	<u>256.187</u>	<u>234.951</u>	<u>188.553</u>	<u>178.559</u>	<u>169.981</u>

Table XVI. SummaryComposition of Total Ash of Lean and Fat CompositeCalculated to 100 Per Cent

	Steer No. 501	Steer No. 512	Steer No. 500*	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	14.822	11.784	13.054	11.563	12.547	11.562
K ₂ O	32.342	35.562	35.130	36.152	35.185	36.191
CaO	1.767	1.747	2.042	2.141	2.369	2.240
P ₂ O ₅	36.141	38.371	38.818	37.785	38.418	37.870
Fe ₂ O ₃	2.018	0.877	0.922	0.577	0.456	0.465
MgO	2.738	2.753	3.060	3.274	3.735	3.302
SiO ₂	2.250	1.054	1.628	0.468	trace	0.704
CO ₂	3.491	2.392	2.970	2.590	2.279	2.510
SO ₃	0.824	0.955	0.986	0.764	0.685	0.749
Cl	4.659	5.817	1.794	6.052	5.586	5.693
Total	101.052	101.312	100.404	101.366	101.260	101.286
O=Cl	1.051	1.312	0.405	1.366	1.260	1.285
Total	100.000*	100.000	100.000*	100.000	100.000	100.000*

*See Section on Calculations.

Table XVII. SummaryComposition of Total Ash of Lean and Fat CompositeSilica Free and Calculated to 100 Per Cent

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	15.163	11.910	13.270	11.617	12.547	11.644
K ₂ O	33.086	35.941	35.711	36.322	35.185	36.448
CaO	1.808	1.766	2.076	2.151	2.369	2.256
P ₂ O ₅	36.973	38.780	39.460	37.963	38.418	38.138
Fe ₂ O ₃	2.064	0.886	0.937	0.580	0.456	0.468
MgO	2.801	2.782	3.111	3.289	3.735	3.325
CO ₂	3.571	2.417	3.019	2.602	2.279	2.528
SO ₃	0.843	0.965	1.002	0.768	0.685	0.754
Cl	4.766	5.879	1.824	6.080	5.586	5.733
Total	101.075	101.326	100.410	101.372	101.260	101.294
O=Cl	1.075	1.326	0.412	1.372	1.260	1.294
Total	100.000	100.000	100.000*	100.000	100.000	100.000

*See Section on Calculations.

Table XVIII. SummaryComposition of Total Ash of Lean and Fat CompositeCalculated to Fresh BasisParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	0.719	0.910	1.164	1.027	1.183	1.112
K ₂ O	1.569	2.745	3.134	3.210	3.318	3.482
CaO	0.086	0.135	0.182	0.190	0.223	0.215
P ₂ O ₅	1.753	2.962	3.463	3.355	3.623	3.643
Fe ₂ O ₃	0.098	0.068	0.082	0.051	0.043	0.045
MgO	0.133	0.213	0.273	0.291	0.352	0.318
SiO ₂	0.109	0.081	0.145	0.042	trace	0.068
CO ₂	0.169	0.185	0.265	0.230	0.215	0.241
SO ₃	0.040	0.074	0.088	0.068	0.065	0.072
Cl	0.226	0.449	0.160	0.537	0.527	0.548
Total	4.902	7.822	8.956	9.001	9.549	9.744
O=Cl	0.051	0.101	0.036	0.121	0.119	0.124
Total	4.851	7.721	8.920	8.880	9.430	9.620

Table XIX. SummaryComposition of Total Ash of Lean and Fat CompositeCalculated to Fresh Basis Fat* FreeParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	1.534	1.269	1.406	1.251	1.381	1.176
K ₂ O	3.347	3.829	3.786	3.910	3.872	3.684
CaO	0.183	0.188	0.220	0.231	0.260	0.227
P ₂ O ₅	3.739	4.132	4.183	4.086	4.228	3.854
Fe ₂ O ₃	0.209	0.095	0.099	0.062	0.050	0.048
MgO	0.284	0.297	0.330	0.354	0.411	0.336
SiO ₂	0.232	0.113	0.175	0.051	-----	0.072
CO ₂	0.360	0.258	0.320	0.280	0.251	0.255
SO ₃	0.085	0.103	0.106	0.083	0.076	0.076
Cl	0.482	0.526	0.193	0.654	0.615	0.580

*Ether Soluble Fat.

Table XX. SummaryComposition of Total Ash of Internal Organs and Internal FatCompositeCalculated to 100 Per Cent

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	19.699	20.764	22.011	14.821	17.318	19.626
K ₂ O	23.719	22.338	23.168	24.115	21.637	23.813
CaO	3.298	2.900	2.744	4.149	3.291	3.326
P ₂ O ₅	39.893	40.950	38.598	43.984	36.215	40.836
Fe ₂ O ₃	0.251	0.768	0.344	1.150	0.925	1.052
MgO	2.891	2.542	2.137	2.842	2.343	2.260
SiO ₂	4.376	2.989	4.355	4.012	12.840	3.072
CO ₂	3.213	3.442	2.408	0.465	0.754	0.446
SO ₃	1.550	1.974	2.066	0.598	0.751	0.628
Cl	1.433	1.719	2.798	4.991	5.070	6.383
Total	100.324	100.386	100.629	101.127	101.144	101.442
O=Cl	0.324	0.388	0.632	1.126	1.144	1.440
Total	100.000	100.000*	100.000*	100.000*	100.000	100.000*

*See Section on Calculations.

Table XXI. SummaryComposition of Total Ash of Internal Organs and Internal FatCompositeSilica Free and Calculated to 100 Per Cent

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	20.600	21.404	23.013	15.440	19.869	20.248
K ₂ O	24.804	23.026	24.223	25.123	24.824	24.568
CaO	3.449	2.989	2.869	4.322	3.776	3.431
P ₂ O ₅	41.719	42.212	40.355	45.822	41.550	42.130
Fe ₂ O ₃	0.262	0.792	0.360	1.198	1.061	1.085
MgO	3.023	2.620	2.234	2.961	2.688	2.332
CO ₂	3.360	3.548	2.518	0.484	0.865	0.460
SO ₃	1.621	2.035	2.160	0.623	0.862	0.648
Cl	1.499	1.772	2.925	5.200	5.817	6.585
Total	100.337	100.398	100.657	101.173	101.312	101.487
O=Cl	0.339	0.400	0.661	1.173	1.313	1.486
Total	100.000*	100.000*	100.000*	100.000	100.000*	100.000*

*See Section on Calculations.

Table XXII. SummaryComposite of Total Ash of Internal Organs and Internal FatCompositeCalculated to Fresh BasisParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	1.081	1.443	1.739	1.415	1.787	2.012
K ₂ O	1.302	1.553	1.830	2.303	2.233	2.441
CaO	0.181	0.202	0.217	0.396	0.340	0.341
P ₂ O ₅	2.190	2.846	3.049	4.200	3.737	4.186
Fe ₂ O ₃	0.014	0.053	0.027	0.110	0.095	0.108
MgO	0.159	0.177	0.169	0.271	0.242	0.232
SiO ₂	0.240	0.208	0.344	0.383	1.325	0.315
CO ₂	0.176	0.239	0.190	0.044	0.078	0.046
SO ₃	0.085	0.137	0.163	0.057	0.078	0.064
Cl	0.079	0.119	0.221	0.477	0.523	0.654
Total	5.507	6.977	7.949	9.656	10.438	10.399
O=Cl	0.018	0.027	0.050	0.108	0.118	0.148
Total	5.489	6.950	7.899	9.548	10.320	10.251

Table XXIII. SummaryComposition of Total Ash of Internal Organs and Internal FatCompositeCalculated to Fresh Basis Silica FreeParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	1.081	1.443	1.740	1.416	1.789	2.013
K ₂ O	1.302	1.553	1.831	2.304	2.236	2.442
CaO	0.181	0.202	0.217	0.396	0.340	0.341
P ₂ O ₅	2.191	2.847	3.050	4.202	3.742	4.187
Fe ₂ O ₃	0.014	0.053	0.027	0.110	0.095	0.108
MgO	0.159	0.177	0.169	0.271	0.242	0.232
CO ₂	0.176	0.239	0.190	0.044	0.078	0.046
SO ₃	0.085	0.137	0.163	0.057	0.078	0.064
Cl	0.079	0.119	0.221	0.477	0.524	0.654
Total	5.268	6.770	7.608	9.277	9.124	10.087
O=Cl	0.018	0.027	0.050	0.108	0.118	0.148
Total	5.250	6.743	7.558	9.169	9.006	9.939

Table XXIV. SummaryComposition of Total Ash of Internal Organs and Internal FatCompositeCalculated to Fresh Basis Fat* FreeParts per Thousand

	Steer No. 501	Steer No. 512	Steer No. 500	Steer No. 547	Steer No. 550	Steer No. 558
Na ₂ O	2.144	2.240	2.476	1.798	2.262	2.211
K ₂ O	2.582	2.410	2.606	2.927	2.827	2.683
CaO	0.359	0.314	0.309	0.503	0.430	0.375
P ₂ O ₅	4.343	4.417	4.342	5.338	4.731	4.600
Fe ₂ O ₃	0.028	0.082	0.038	0.140	0.120	0.119
MgO	0.315	0.275	0.241	0.344	0.306	0.255
SiO ₂	0.476	0.323	0.490	0.487	1.677	0.346
CO ₂	0.349	0.371	0.271	0.056	0.099	0.051
SO ₃	0.169	0.213	0.232	0.072	0.099	0.070
Cl	0.157	0.185	0.315	0.606	0.662	0.719

*Ether Soluble Fat.

Table XXV. Summary. Weights of Parts and Weights of Ether Soluble Fat in Parts (grams).

<u>Lean and Fat Composite</u>				
	Weight of Part	Weight of Fat	Weight of Part Fat Free	Factor - Total Wt. to Fat Free Weight
Steer 501	548,474	291,323	257,151	2.133
512	277,824	78,606	199,218	1.395
500	217,860	37,538	180,322	1.208
547	94,790	16,981	77,809	1.218
550	59,680	8,532	51,148	1.167
558	41,560	2,292	39,268	1.058
<u>Composite Internal Organs and Internal Fat</u>				
Steer 501	85,957	42,608	43,349	1.983
512	57,864	20,574	37,290	1.552
500	49,938	14,873	35,065	1.424
547	23,700	5,055	18,645	1.271
550	17,694	3,717	13,977	1.266
558	11,957	1,078	10,879	1.099

Table XXVI. Alkalinity Titrations

Steer No.	c.c. N/10 HCl per gram of Soluble Ash Indicator- Phenolphthalein	c.c. N/10 HCl per gram of Soluble Ash Indicator- Methyl Orange
Lean and Fat Composite		
501	16.11	48.75
512	13.91	44.92
500	12.51	63.02
547	11.87	48.18
550	13.92	46.14
558	13.91	47.55
Internal Organs and Internal Fat Composite		
501	11.50	66.08
512	10.76	61.91
500	14.87	56.60
547	9.34	46.71
550	5.18	51.40
558	8.08	46.91

Bibliography

1. E. B. Forbes--''The Mineral Elements in Animal Nutrition.'' Ohio Exp. Sta. Bull. 201 (1909).
2. Lawes and Gilbert--Phil. Trans. (1883), p. 865.
3. E. Wildt--Land. Versuchs Stationen, 15, 404-454.
Abstracted J. Chem. Soc. 26, 291 (1873).
4. P. C. Plugge--Pfluger's Archiv, IV, p. 101-103.
Abstracted J. Chem. Soc. 25, 257 (1872).
5. H. Weiske--Zeitschr. f. Biologie, VIII, 239-245.
Abstracted J. Chem. Soc. 25, 897 (1872).
6. L. Graffenberger--Land. Versuchs Stationen, 39, 115-126 (1891).
7. H. Brubacher--Zeit. Biol., 27, 517-549.
8. S. Gabriel--Zeitschr. f. Physiolog. Chem. 18, p. 257-302(1894).
9. Hans Aron--Pfluger's Archiv. 106, 91 (1905).
10. E. Wolff--''Aschen-Analysen'', Part II (1870-1880).
11. Julius Katz--Pflugers Archiv. 63, 83 (1896).
12. Emil Abderhalden--Quantitative analysis of blood. Zeit. Physiol. Chem. 23, 221-231 (1897)
13. A. Jarische--Analysis of Blood Ash. Chem. Centr. 1877, 7.
Abstracted J. Chem. Soc., 2, 940, (1877).
14. E. E. Vanatta--Thesis, Master of Arts, University of Missouri, 1911.
A. G. Hogan--Thesis, Master of Arts, University of Missouri, 1912.
W. B. Connell--Thesis, Master of Arts, University of Missouri, 1914.
15. Bulletin 107, U. S. Dept. of Agriculture, Bureau of Chemistry.
16. Mixer and Dubois--Modification of the Zimmerman-Reinhardt Method. J. Am. Chem. Soc., 17, 405.

Acknowledgement.

The author wishes to express his indebtedness to the various members of the Agricultural Chemistry staff of the University of Missouri Agricultural Experiment Station, whose assistance and advice have been invaluable, and especially to Dr. P. F. Trowbridge under whose direction this work was done.

