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# The Effect of DDT Residue on the Composition and Digestibility of Alfalfa Hay

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# THE EFFECT OF DDT RESIDUE ON THE COMPOSITION AND DIGESTIBILITY OF ALFALFA HAY

by

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# INTRODUCTION

DDT (2, 2 bis (p-chlorophenyl) 1, 1, 1-trichloroethane) was first synthesized in 1874 by Zeidler in Switzerland, however, its effectiveness as an insecticide was not known until about 1942. The active principle was first known as G.E.B. (Geigarol-Neocid Base): but in 1943 DDT was suggested as an abbreviation for Dichlore Diphenyl Trichloroethane (Kanegis, 1946). In fact DDT, as a powerful insecticide is one of the developments made during World War II. The importance of DDT in the control of the common insect enemies of mankind, such as mosquitoes, flies, moths, and many agricultural insect pests is realized more day by day. Many studies have been conducted to determine the uses, toxicity and residual effectiveness of DDT. However, the nutritive value of DDT dusted feedstuffs has not been investigated heretofore.

During recent years, in the west, alfalfa seed and hay production has declined as a result of injuries caused by certain species of insects such as lygus bugs and alfalfa weevil. DDT has been used successfully to control these insect pests. Because of this fact there is a need for more information on the chemical composition, digestibility and metabolizable energy content of DDT treated alfalfa hay.

## REVIEW OF LITERATURE

There are no references on the digestibility and metabolizable energy of DDT-treated feedstuffs. The following review, however, throws some light on work related to this problem.

Orr and Mott (1945) demonstrated that DDT is not acutely toxic to animals though some developed tremors and slight degeneration of the liver. In their investigation, 100 to 200 mgm. of DDT per kgm. of body weight was administered orally to cows, horses, and sheep. It was found that most of the DDT was eliminated with the feces and only a small amount was taken up by the blood stream.

In the investigation by the Bureau of Entomology and Plant Quarantine, three sheep were put in a pasture 48 hours after the pasture had been dusted with 40 pounds of 10-percent DDT per acre. The sheep showed extreme neurologic symptoms after 72 hours, but the animals eventually recovered. The field was dusted again a month later, and the sheep reacted in the same way, and again all recovered. One of the ewes was taken to the University of Southern California and fed 2 grams of DDT daily for 11 days. The amount was then increased to 4 grams of DDT per day for the next 40 days. During the entire period when DDT was fed orally, there were no noticeable symptoms of DDT poisoning. When the dosage was increased to 16 grams a day pronounced nervous symptoms were developed and death ensued 17 days later (Roark and McIndoo, 1947).

Nelson (1944) reported that among larger animals cows seem to be more sensitive to DDT. Characteristic symptoms such as tremors of hind legs and neck occured with doses of 0.05 to 0.1 grams per pound of body weight per day for three weeks. However, only one sheep out of three

tested showed liver damage while the other sheep and a horse remained unaffected.

Two goats having received single oral doses of 1.25 grams and 0.68 grams of DDT per pound of body weight eliminated sufficient amounts of DDT in the milk to produce toxic symptoms and death in white rats after 29 to 30 hours of milk feeding (Telford, 1945).

Kanegis (1946) stated in his review of DDT that absorption of DDT in powdered form, through the gut is variable and irregular notably depending on the fat content of the diet. In the case of DDT being administered in oil 50 to 95 percent was found to be absorbed. The body eliminated DDT in the feces with the bile, and some DDT was eliminated in the urine as a detoxified product. He further stated that in experiments with rabbits from 5 to 50 percent of a single dose administered in oil was excreted in the feces.

Leary, et al. (1946), reported that a half-grown kitten died with toxic symptoms from consuming goat's milk lactated by DDT fed goats: while neither the mother goats nor the kids died, apparently goats have a higher resistance to DDT than cats.

White and Sweeney (1945) in their investigation stated that eight rabbits weighing from 2 to 3 kgm. were given a dose of 100 mgm. of DDT per kilogram of body weight and the dose was administered in olive oil solution by means of a stomach tube. On analysis of urine it showed the presence of organic chlorine, namely di (p-chlorophenyl) acetic acid. It was further demonstrated by these investigators that di (p-chlorophenyl) acetic acid can be isolated from the urine.

From the above review it is known that DDT is a highly poisonous substance, and it has a cummulative effect. However, to large animals

small quantities of DDT do not seem to be toxic if it is used and handled properly. Most of the DDT consumed by the animal is excreted in milk, feces, and urine. Nevertheless, DDT excretion into milk and its concentration in the cream and butter causes great concern because of its injurious effects to nursing animals, milk fed babies and to human health generally.

## METHOD OF PROCEDURE

The general plan of the experiment was to determine whether residual quantities of DDT on alfalfa hay will affect its chemical composition, digestibility and metabolizable energy. The investigation was performed from March 8, 1948, to May 24, 1948.

Experimental design. A 4 x 4 Latin square design was employed (table 1) to determine the digestibility and metabolizable energy content of the DDT-dusted alfalfa hay. Four samples of alfalfa hay were fed at random to four sheep for four periods with the restriction of not repeating the hay already fed in a row or column.

Table 1. Experimental design.

| DDT per acre, lbs. | Alfalfa hay no. | Period number 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|
| | | Animal number | | | |
| 0 | I 1 | I 135 | I 133 | I 139 | I 137 |
| 1 | I 7 | I 133 | I 135 | I 137 | I 139 |
| 2 | I 9 | I 139 | I 137 | I 135 | I 133 |
| 4 | I 13 | I 137 | I 139 | I 133 | I 135 |

These levels of DDT were used because the recommended amount of actual DDT for insect control of alfalfa is from 1 to 2 pounds per acre. The level of 4 pounds per acre, approximately twice that of the recommended amount, was used in order to investigate the effects of the DDT when employed in excess.

Application of DDT. The hay was treated and harvested by the personnel mentioned in the acknowledgement. Four replicated plots of first crop alfalfa growing on the Kidman farm on the west slope of Cache Valley were dusted with DDT on May 29, 1947, at the following

levels per acre: (1) none, (2) 1 pound, (3) 2 pounds and (4) 4 pounds. The required amount of DDT was mixed with pyrophyllite and applied at the rate of 20 pounds per acre with a power duster. Because of inadequate facilities expense and time, hay from only four of the plots was used for digestion trials, however, the hay from all the 16 plots was chemically analysed.

Figure 1 shows the field a few days before cutting. The areas which were not dusted were greyish green in color, while the dusted areas were bright green in color. The leaves of the alfalfa on the undusted areas were skeletonized by weevil while the leaves on the dusted areas were not eated by weevil (figure 2). There was a marked line of color and height difference in the dusted and non-dusted alfalfa plots (figure 3).

On June 20, 1947, the hay was cut, raked into windrows, cured, baled, and labeled. The hay was then hauled to the Animal Husbandry Farm on July 1, 1947 and stacked. The hay was of good quality, free from foreign matter or mold and was stored in a clean dry place.

Animals. Four wether lambs (three Columbia x Rambouillet and one Rambouillet) approximately one year old, weighing from 85 to 90 pounds were used. The lambs were shorn before the experiment started. They were weighed at the beginning and at the end of each collection period. The animals were given the numbers I-133, I-135, I-137, and I-139.

Metabolism cages. Four metabolism cages were constructed for the purpose of collecting urine and feces separately from each of the individual lambs (figure 4). Each cage measures 2 feet 6 inches in width, 6 feet in length and 3 feet 6 inches in height; the legs measure 2 feet 6 inches. The inside has an adjustable partition on each side in order

Figure 1. Aerial photograph of alfalfa field taken the day before cutting the first crop. The light areas are the untreated plots and buffer strips. The light color was caused by the alfalfa weevil larvae feeding upon and damaging the alfalfa plants. The dark areas are the plots treated with DDT in which there is very little weevil damage.


Figure 2. Individual alfalfa plants from representative plots. No. 1 is from a plot receiving 2 lbs. of DDT per acre; No. 2 is from a plot receiving no DDT. Note the damage to the plant by the alfalfa weevil larvae in the latter.

Figure 3. The alfalfa in the foreground was treated with 2 lbs. of DDT per acre; the alfalfa immediately beyond in the light area is a buffer strip and received no DDT. The dark area immediately beyond the buffer strip is a plot that was treated with DDT. Note the sharp line of demarcation between the treated and untreated areas, and the difference in height of the alfalfa.

Figure 4. Metabolism cage showing the feed rack, urine and feces bottles in place.

that the animal cannot turn around. The feed trough is moveable and can be fastened after the ration has been placed in it. Just beneath the false bottom iron floor of the cage is a copper screen which prevents hay and feces from falling into the aluminum tray. The aluminum tray is constructed in such a way that urine and feces go into receiving bottles separately.

Feeding and care of sheep. Before the beginning of the experiment, the sheep were fed alfalfa hay for about one month to accustom them to this type of feed, and to acquaint the investigator with how to feed and care for them. The hays were chopped in about three inch lengths.

The sheep were fed between 8:00 and 9:00 a.m. and between 4:30 and 5:30 p.m. The hays were weighed on a balance to an accuracy of .01 lbs. The amount of hays were limited in each period, as near as possible, to that of the animals consuming the least. Each morning just before feeding the sheep, the orts (weigh-back), if there were any, were collected carefully [illegible] saved in a labeled sack for weighing and analysis.

Collection procedure. A preliminary period of 5 to 7 days was allowed before the beginning of the collection period. The hay under study was fed in constant daily amounts for a preliminary period in order to free the digestive tract of the material coming from the feed consumed prior to the start of feeding the hay under investigation.

Before the collection began, the floors, the screens, and the trays of the metabolism cages, and the collection bottles were thoroughly brushed, cleaned and dried. The collection started and ended at a definite time; the collection period lasted 7 days.

Fifty ml. of toluene was added to each urine bottle at the beginning of each collection period. Ten drops of toluene were added to the feces bottles once daily.

The day previous to the end of the collection period all of the scattered hay was carefully gathered from the floors and screens of the metabolism cages to insure the correct amount of feed intake. On the day of collection before the orts were weighed, the scattered hay was again carefully collected and added to the orts for weighing.

At the time of the collection, feces and urine bottles were removed first of all. With the aid of a steel brush all the feces were carefully brushed off the floor, screen and tray into the collection bottles.

Sampling of feeds, feces, and urine. A composite sample of hay for each plot was used for each of the four periods for chemical analysis. The total amount of orts was saved for each period for analysis.

At the end of the collection period the feces were weighed. They were then emptied from the bottle into a large aluminum bowl where they were thoroughly mixed by hand. A sample was put into a quart bottle, labeled and sealed air tight. Immediately the wet feces were weighed and dried in an air-drying oven at 60° C for 24 hours, with brisk air passing through. The feces were then reweighed and ground for analysis.

The urine, in the collection bottle, was thoroughly shaken and 20 percent of the measured urine was poured into a quart bottle for analysis.

The hays, orts, and feces were ground through a Wiley mill using a 1-mm screen.

Analysis procedures. Hays, orts, and feces were analyzed for ash, dry matter, ether extract, crude fiber, nitrogen-free extract, nitrogen,

crude protein, and gross energy. Calcium and phosphorus content of the hays were also determined. Urine was analyzed for nitrogen, and gross energy. All analyses were run in duplicate, and in cases where the results varied considerably, repeats were made.

Ash was determined by burning the samples in an electric furnace at 600° C for 3 hours with an automatic control pyrometer. Dry matter was determined by drying the samples in an oven at 100° C for 10 hours. Crude fat or ether extract was determined by extracting the samples with anhydrous ether for 16 hours. Crude fiber was determined by the method of the Association of Official Agricultural Chemists (1945, p 409). Nitrogen-free-extract was found by difference.

N.F.E. = 100% - (moisture % + ether extract % + crude fiber % + ash % + protein %).

Total nitrogen was determined by the Kjeldahl method of the Association of the Official Agricultural Chemists (1945, page 26 to 27) except the titration which was after Scales and Harrison (1920); boric acid is employed to hold the ammonia distilled in the process. Crude protein was obtained by multiplying the amount of nitrogen determined by the factor 6.25.

Calcium was determined by the method of the Official Agricultural Chemists (1945, p. 119-120) except the digestion which was done by the method of digesting biological materials for calcium and phosphorus analysis (Gerrits, 1935). Phosphorus was determined by the colorimetric method after Koenig and Johnson (1942). DDT residues on the hays were found by determining the total chlorine (i.e all 3 atoms of chlorine in DDT molecule) based on a method described by Umhoefer (1943), except the titration was by Volhard's method according to Gunther (1945).

Gross energy determinations: The ground air dry samples of alfalfa hay, oats, and feces were made into pellets by the use of a pellet press.

The urine was heated over a hot water bath at 45° C to dryness. The residue was transferred to a capsule containing a cellulose pellet using a minimum amount of distilled water. The contents of the capsule were again dried over a hot water bath.

The gross energy of the samples was determined by the use of a Parr Oxygen Bomb Adiabatic Calorimeter in accordance with the instructions contained in the Parr Manual No. 117.

Metabolizable energy was obtained by difference by the use of the following formula:

Metabolizable energy = gross energy in feed - gross energy in oats - (Gross energy in feces + urine + methane gas).

The amount of methane produced was estimated by using the following formula (Black 1948):

$$\text{methane in gms.} = 2.41x + 9.80$$

x represents hundreds of grams of carbohydrates digested. To convert grams of methane to Calories, the value of 13.344 was used.

Unless the animal is neither gaining nor losing body protein, the difference between the energy consumed and the energy lost in the solid, liquid, and gaseous excreta does not indicate the true metabolizable energy value of the feed. A correction for the gain or loss of body protein was made according to the factor suggested by Rubner and quoted by Hamilton (1928). For each gram of urinary nitrogen derived from the catabolism of body proteins (equal to negative nitrogen balance) 7.45 Calories were subtracted from the urinary energy, and for each gram of nitrogen stored in the body (equal to the positive nitrogen balance) 7.45 Calories were added to the urinary energy.

All the data were analyzed statistically by the analysis of variance as described by Snedecor (1946). Differences were considered to be significant at the 5% level and highly significant at the 1% level with a probability of 19: 1 and 99: 1 respectively.

## RESULTS AND DISCUSSION

The yield and chemical composition of alfalfa hay. There was a highly statistically significant difference in the yield of alfalfa hay in tons per acre between the undusted and DDT dusted hay (table 2). The average yield of undusted alfalfa hay was 1.12 tons per acre and the yield of DDT dusted alfalfa hay was 1.50, 1.40, and 1.47 tons per acre for the 1, 2, and 4 pound levels of DDT, respectively. However, there was no significant difference in yield among the 1, 2, and 4 pound levels of DDT. The increase in the yield of alfalfa hay was probably due to the protection furnished by the DDT from insect pests.

There was a highly significant difference in percentage ether extract between the undusted and DDT dusted alfalfa hay. The untreated hay contained 2.16 percent ether extract and the treated hay 2.46 percent, 2.46 percent, and 2.43 percent for the 1, 2, and 4 pound DDT treatments, respectively.

The DDT treated hay had a bright green color while the untreated hay had a grayish green color. The carotene content of the DDT treated hay was significantly greater than the untreated hay (table 3). There was no significant difference among the DDT treated hays.

The statistical analyses of the yield and chemical composition of the yield and chemical composition of the hays are given in appendix tables 1 and 2.

Food consumption and live-weight gain. The food consumption was held as constant as possible in the various periods in order to take out the effect of having varying food intakes. At the end of period

Table 2. Yield and chemical composition of first crop alfalfa hay

| Pounds of DDT per acre | Hay no. | Dry matter tons/ acre | DDT residue | Percent composition on dry basis | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Protein | Ether extract | Crude fiber | Ash | Nitrogen free extract | P | Ca |
| | | | ppm | | | | | | | |
| 0 | I-1 | 1.23 | 0 | 15.2 | 2.02 | 32.25 | 9.14 | 41.5 | 0.23 | 2.13 |
| 0 | I-2 | 1.23 | | 14.7 | 2.21 | 31.08 | 8.79 | 43.2 | 0.19 | 1.91 |
| 0 | I-3 | 1.05 | | 16.0 | 2.21 | 32.37 | 9.30 | 40.1 | 0.26 | 1.91 |
| 0 | I-4 | 0.98 | | 15.5 | 2.18 | 30.25 | 8.91 | 43.0 | 0.18 | 2.33 |
| Avg. | | 1.12 | | 15.4 | 2.16 | 31.48 | 9.03 | 41.9 | 0.22 | 2.07 |
| 1 | I-5 | 1.60 | | 15.8 | 2.59 | 27.18 | 9.55 | 44.9 | 0.20 | 2.16 |
| 1 | I-6 | 1.66 | | 16.5 | 2.35 | 32.64 | 9.07 | 39.5 | 0.25 | 1.81 |
| 1 | I-7 | 1.37 | 17 | 15.0 | 2.51 | 31.27 | 8.74 | 43.6 | 0.26 | 1.78 |
| 1 | I-8 | 1.37 | | 16.8 | 2.38 | 33.38 | 9.01 | 38.5 | 0.30 | 1.96 |
| Avg. | | 1.50 | | 16.0 | 2.46 | 31.10 | 9.09 | 41.4 | 0.23 | 1.93 |
| 2 | I-9 | 1.34 | 23 | 16.0 | 2.28 | 31.12 | 8.74 | 41.9 | 0.21 | 1.89 |
| 2 | I-10 | 1.60 | | 16.4 | 2.55 | 30.62 | 8.94 | 41.6 | 0.24 | 1.98 |
| 2 | I-11 | 1.34 | | 15.2 | 2.67 | 31.95 | 8.30 | 41.9 | 0.20 | 1.91 |
| 2 | I-12 | 1.32 | | 15.5 | 2.32 | 31.08 | 8.83 | 42.2 | 0.23 | 1.59 |
| Avg. | | 1.40 | | 15.8 | 2.46 | 31.19 | 8.70 | 41.9 | 0.22 | 1.84 |
| 4 | I-13 | 1.56 | 50 | 16.1 | 2.25 | 32.63 | 9.39 | 39.6 | 0.25 | 1.83 |
| 4 | I-14 | 1.73 | | 16.8 | 2.33 | 30.30 | 9.52 | 41.0 | 0.21 | 2.02 |
| 4 | I-15 | 1.28 | | 16.2 | 2.67 | 28.93 | 8.95 | 43.3 | 0.24 | 2.05 |
| 4 | I-16 | 1.30 | | 16.5 | 2.45 | 29.45 | 8.94 | 42.6 | 0.23 | 1.77 |
| Avg. | | 1.47 | | 16.4 | 2.43 | 30.32 | 9.20 | 41.6 | 0.23 | 1.92 |
| Minimum level of significance between treatment averages | | | | | | | | | | |
| P 0.05 | | 0.12 | | 1/ | 0.21 | 1/ | 1/ | 1/ | 1/ | 1/ |
| P 0.01 | | 0.17 | | | 0.30 | | | | | |

1/ Difference between treatment means are not considered significant.

Table 3. Carotene content of green alfalfa just before cutting for hay

| Pounds of DDT per acre | Plot no. | Hay no. | Carotene content |
|---|---|---|---|
| | | | PPM |
| 0 | 2 | I-1 | 123 |
| | | | 223 |
| 0 | 8 | I-2 | 332 |
| | | | 304 |
| 0 | 10 | I-3 | 297 |
| | | | 326 |
| 0 | 16 | I-4 | 369 |
| | | | 334 |
| Avg. | | | 288 |
| 1 | 3 | I-5 | 352 |
| | | | 345 |
| 1 | 6 | I-6 | 446 |
| | | | 289 |
| 1 | 9 | I-7 | 376 |
| | | | 332 |
| 1 | 13 | I-8 | 359 |
| | | | 328 |
| Avg. | | | 354 |
| 2 | 4 | I-9 | 153 |
| | | | 329 |
| 2 | 5 | I-10 | 410 |
| | | | 337 |
| 2 | 12 | I-11 | 529 |
| | | | 540 |
| 2 | 14 | I-12 | 380 |
| | | | 424 |
| Avg. | | | 388 |
| 4 | 1 | I-13 | 200 |
| | | | 203 |
| 4 | 7 | I-14 | 398 |
| | | | 421 |
| 4 | 11 | I-15 | 455 |
| | | | 635 |
| 4 | 15 | I-16 | 459 |
| | | | 321 |
| Avg. | | | 395 |

1 it was noted that the sheep would eat more food, therefore, the intake of alfalfa hay was increased.

There were no significant differences among the live-weight gains which could be attributed to the hays fed. However, there was a significant difference among the periods. The statistical analyses of the live-weight gain is shown in appendix table 11.

Nitrogen balance. The nitrogen balance was significantly in favor of the untreated alfalfa hay when compared to the hay which had been dusted with 2 and 4 pounds of DDT per acre. However, there was no significant difference between the undusted hay and the hay which was dusted with one pound of DDT. Also there was no significant difference among the DDT dusted hays, although as the amount of DDT on the hay increased the nitrogen balance decreased. From these results it appears that the concentration of DDT on the hay influences the amount of nitrogen stored.

The statistical analyses of the nitrogen balance is in appendix table 10.

Digestibility of alfalfa hay. There were no significant differences among the digestion coefficients for protein (Nitrogen X 6.25), ether extract, crude fiber, ash and nitrogen free extract (table 5). However, the digestion coefficients for ether extract approached significance ( F value was 4.49, the significant value at P = 0.05 being 4.76). There was also no significant differences among the hays as regards the total digestible nutrients. From these results it appears that the DDT residues on the hay did not effect the digestibility of the above constituents when the hay was fed to sheep.

The statistical analyses for the digestibility of the hays are given in appendix tables 4, 5, 6, 7 and 8.

Table 4. The dry matter intake, average body weight, gain and nitrogen balances of sheep fed alfalfa hay dusted with various amounts of DDT

| Sheep no. | Period no. | DDT per acre | Hay no. | Avg. wt. | Period gain | Dry matter intake | Nitrogen in Feed offered | Orts | Feces | Urine | Balance* |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | lb. | | lb. | lb. | gm. | gm. | gm. | gm. | gm. | gm. |
| I-133 | 2 | 0 | I-1 | 94.3 | - 0.6 | 1114 | 28.7 | 0.9 | 8.2 | 15.6 | 4.0 |
| I-135 | 1 | 0 | I-1 | 93.9 | 0.7 | 1004 | 24.6 | 0.1 | 7.4 | 15.0 | 2.1 |
| I-137 | 4 | 0 | I-1 | 97.5 | - 5.0 | 1169 | 28.4 | 0.0 | 8.5 | 16.8 | 3.1 |
| I-139 | 3 | 0 | I-1 | 88.8 | 6.5 | 1165 | 28.7 | 0.2 | 9.9 | 15.2 | 3.4 |
| Avg. | | | | 93.6 | 1.5 | 1113 | 27.6 | 0.3 | 8.5 | 15.6 | 3.1 |
| I-133 | 1 | 1 | I-7 | 94.1 | 1.1 | 997 | 24.1 | 0.2 | 4.4 | 17.3 | 2.2 |
| I-135 | 2 | 1 | I-7 | 94.4 | 0.3 | 1124 | 27.9 | 0.7 | 8.6 | 16.4 | 2.2 |
| I-137 | 3 | 1 | I-7 | 96.8 | 6.5 | 1167 | 27.9 | 0.0 | 9.1 | 17.1 | 1.7 |
| I-139 | 4 | 1 | I-7 | 91.0 | - 2.0 | 1146 | 27.9 | 0.3 | 8.6 | 18.0 | 1.0 |
| Avg. | | | | 94.1 | 1.5 | 1109 | 26.9 | 0.3 | 7.7 | 17.2 | 1.8 |
| I-133 | 4 | 2 | I-9 | 98.5 | - 4.0 | 1052 | 29.2 | 2.8 | 7.6 | 17.8 | 1.0 |
| I-135 | 3 | 2 | I-9 | 98.0 | 7.0 | 1169 | 29.0 | 0.0 | 7.9 | 21.8 | -0.7 |
| I-137 | 2 | 2 | I-9 | 94.1 | - 1.1 | 1151 | 29.2 | 0.6 | 7.3 | 17.9 | 3.4 |
| I-139 | 1 | 2 | I-9 | 84.8 | - 0.5 | 996 | 24.9 | 0.1 | 7.6 | 19.1 | -1.9 |
| Avg. | | | | 93.8 | 0.3 | 1092 | 28.1 | 0.9 | 7.6 | 19.1 | 0.5 |
| I-133 | 3 | 4 | I-13 | 97.3 | 6.5 | 1131 | 29.8 | 0.4 | 8.5 | 19.2 | 1.7 |
| I-135 | 4 | 4 | I-13 | 99.0 | - 5.0 | 1165 | 30.1 | 0.0 | 9.6 | 20.9 | -0.4 |
| I-137 | 1 | 4 | I-13 | 94.5 | 0.6 | 936 | 24.7 | 0.3 | 7.5 | 19.2 | - 2.3 |
| I-139 | 2 | 4 | I-13 | 85.0 | 1.0 | 1135 | 30.4 | 0.6 | 9.2 | 19.1 | 1.5 |
| Avg. | | | | 93.9 | 0.8 | 1092 | 28.8 | 0.3 | 8.7 | 19.6 | 0.1 |

* Grams of nitrogen per animal

Table 5. Digestion coefficients and total digestible nutrients of alfalfa hay dusted with various amounts of DDT when fed to sheep.

| Sheep No. | Period No. | DDT per acre | Protein | Ether extract | Crude fiber | Nitrogen free extract | Total digestible nutrients |
|---|---|---|---|---|---|---|---|
| | | lb. | % | % | % | % | % |
| I-133 | 2 | 0 | 71 | 57 | 44 | 70 | 53 |
| I-135 | 1 | 0 | 70 | 53 | 48 | 73 | 55 |
| I-137 | 4 | 0 | 70 | 33 | 44 | 75 | 53 |
| I-139 | 3 | 0 | 65 | 42 | 46 | 62 | 49 |
| Avg. | | | 69 | 46 | 46 | 70 | 53 |
| I-133 | 1 | 1 | 64 | 55 | 46 | 74 | 54 |
| I-135 | 2 | 1 | 69 | 49 | 41 | 72 | 53 |
| I-137 | 3 | 1 | 68 | 46 | 50 | 71 | 54 |
| I-139 | 4 | 1 | 69 | 43 | 47 | 75 | 55 |
| Avg. | | | 68 | 48 | 46 | 73 | 54 |
| I-133 | 4 | 2 | 72 | 28 | 48 | 78 | 56 |
| I-135 | 3 | 2 | 74 | 32 | 50 | 72 | 55 |
| I-137 | 2 | 2 | 75 | 35 | 50 | 77 | 58 |
| I-139 | 1 | 2 | 69 | 55 | 48 | 72 | 55 |
| Avg. | | | 73 | 38 | 49 | 75 | 56 |
| I-133 | 3 | 4 | 71 | 29 | 54 | 73 | 55 |
| I-135 | 4 | 4 | 68 | 29 | 47 | 73 | 53 |
| I-137 | 1 | 4 | 69 | 50 | 50 | 71 | 54 |
| I-139 | 2 | 4 | 69 | 43 | 46 | 73 | 53 |
| Avg. | | | 69 | 38 | 49 | 73 | 54 |
| Coefficient of variation | | | 3.3 | 12.3 | 5.0 | 3.9 | 3.0 |

Metabolizable energy. There was a highly significant difference in the metabolizable energy between the undusted and DDT dusted hay (table 6). The untreated hay contained 2108 Calories per kilogram of dry matter, while the dusted hay contained 2582, 2601 and 2573 Calories per kilogram of dry matter for the 1, 2 and 4 pound DDT treatments, respectively. There was no significant difference among the hays which were treated with DDT.

From the appearance of the hays in figure 2 it would appear that there should be some difference in the utilization of the hays. If these facts are considered it seems that metabolizable energy is a better measure of utilization than total digestible nutrients.

The statistical analyses for the metabolizable energy content of the hays is shown in appendix table 9.

Table 6. The gross energy, its losses, and the metabolizable energy of the alfalfa hay dusted with various amounts of DDT when fed to sheep

| Sheep no. | Period no. | DDT per acre | Dry matter intake | Energy in hay eaten | Energy | | | Correction for N balance | Metabolizable energy | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | In feces | In urine | In methane | | In dry matter | per kgm dry matter |
| | | lb. | gm. | Cal. | Cal. | Cal. | Cal. | Cal. | Cal. | Cal. |
| I-133 | 2 | 0 | 1114 | 4872 | 2051 | 231 | 285 | ∮29.8 | 2235 | 2096 |
| I-135 | 1 | 0 | 1004 | 4393 | 1713 | 223 | 279 | ∮15.6 | 2194 | 2185 |
| I-137 | 4 | 0 | 1169 | 5114 | 2039 | 225 | 277 | ∮23.1 | 2596 | 2221 |
| I-139 | 3 | 0 | 1165 | 5103 | 2358 | 238 | 283 | ∮25.3 | 2249 | 1931 |
| Avg. | | | | | | | | | | 2108 |
| I-133 | 1 | 1 | 997 | 4796 | 1836 | 242 | 321 | -15.6 | 2381 | 2389 |
| I-135 | 2 | 1 | 1124 | 5424 | 2081 | 238 | 233 | ∮16.4 | 2888 | 2570 |
| I-137 | 3 | 1 | 1167 | 5605 | 2115 | 230 | 303 | ∮12.7 | 2970 | 2545 |
| I-139 | 4 | 1 | 1146 | 5512 | 1949 | 238 | 279 | ∮ 7.5 | 3054 | 2664 |
| Avg. | | | | | | | | | | 2542 |
| I-133 | 4 | 2 | 1052 | 4996 | 1615 | 241 | 268 | ∮ 7.45 | 2880 | 2737 |
| I-135 | 3 | 2 | 1169 | 5489 | 1948 | 284 | 304 | - 5.20 | 2948 | 2522 |
| I-137 | 2 | 2 | 1151 | 5419 | 1825 | 253 | 309 | ∮25.3 | 3057 | 2656 |
| I-139 | 1 | 2 | 996 | 4681 | 1685 | 229 | 276 | -14.2 | 2477 | 2487 |
| Avg. | | | | | | | | | | 2601 |
| I-133 | 3 | 4 | 1131 | 5439 | 1868 | 292 | 299 | ∮12.7 | 2992 | 2639 |
| I-135 | 4 | 4 | 1165 | 5587 | 2080 | 253 | 271 | - 3.0 | 2980 | 2558 |
| I-137 | 1 | 4 | 936 | 4505 | 1660 | 226 | 264 | -17.1 | 2338 | 2498 |
| I-139 | 2 | 4 | 1135 | 5464 | 1999 | 240 | 291 | ∮11.2 | 2945 | 2595 |
| Avg. | | | | | | | | | | 2573 |

## SUMMARY AND CONCLUSIONS

1. DDT has been successfully used to control lygus bugs and alfalfa weevil which cause injuries to the production of alfalfa seed and hay. However, the nutritive value of DDT dusted feedstuff has not been investigated heretofore. Because of this fact this study was conducted during the spring of 1948 at the Utah State Agricultural College, Logan, Utah.

2. Four replicated plots of first crop alfalfa growing on the Kidman Farm on the west slope of Cache Valley were dusted with DDT on May 29, 1947, at the following levels per acre: (1) none (2) 1 pound (3) 2 pounds and (4) 4 pounds in connection with the Utah Agricultural Experiment Station, project number 289.

3. The four hays were fed at random to four sheep in metabolism cages for four periods with the restriction of not repeating the hay already fed in a row or column (Latin square design).

4. A preliminary period of 5 to 7 days was allowed before the beginning of a collection period. The collection of feces and urine started and ended at a definite time; each collection period lasted 7 days.

5. Samples of feeds, feces, and orts (weigh-backs) were prepared and analyzed for ash, dry matter, ether extract, crude fiber, nitrogen, and gross energy. In case of urine, nitrogen and gross energy were determined. Calcium, phosphorus, carotene and DDT residues were also determined on the hays.

6. There was a highly significant difference in the yield of alfalfa hay in tons per acre between the undusted and DDT dusted hay. The average yield of DDT dusted alfalfa hay increased from 1.12 tons

per acre in the undusted hay to 1.50, 1.40, and 1.47 tons per acre in the 1, 2, and 4 pound levels of DDT respectively.

7. There was a highly significant difference in percentage ether extract between the undusted and DDT dusted alfalfa hay, the untreated hay containing 2.16 percent ether extract while the treated hay contained 2.46, 2.46 and 2.43 percent for the 1, 2, and 4 pound DDT treatments respectively.

8. The carotene content of DDT treated hay was significantly higher than the untreated hay. The carotene content increased from 288 parts per million in the untreated hay to 354 parts per million, 388 parts per million, and 395 parts per million in the 1, 2, and 4 pound DDT treatments respectively.

9. There was no significant difference among the live weight gains which could be attributed to the hays fed. However, there was significant differences among the periods.

10. The nitrogen balance was significantly in favor of the untreated alfalfa hay when compared to 2 and 4 pounds DDT treated hay. However, there was no significant difference between the undusted and 1 pound DDT dusted hay.

11. There were no significant differences among the digestion coefficients for protein, ether extract, crude fiber, ash and nitrogen free extract. However, the digestion coefficient for ether extract approached significance (F value was 4.49, the significant value being 4.76).

12. There was also no significant difference among the hays in regard to the total digestible nutrients.

13. There was a highly significant difference in the metaboliz̃able

energy between the undusted and DDT dusted hay. The undusted hay contained 2108 Calories per kilogram of dry matter, while the dusted hay contained 2542, 2601, and 2573 Calories per kilogram of dry matter for 1, 2, and 4 pounds DDT treatments, respectively. However, there were no significant differences among the DDT dusted hays.

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Appendix table 1. Analysis of variance for yield and percentage chemical composition of alfalfa hay

| Source of variation | Degrees of freedom | Mean squares | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Dry matter tons/acre | Protein | Ether extract | Crude fiber | Ash | Nitrogen free extract | Phosphorus | Calcium |
| Treatments | 3 | 0.1146** | 0.780 | 0.08543* | 0.9788 | 0.1841 | 0.287 | 0.000267 | 0.03610 |
| Replicates | 3 | 0.0880** | 0.237 | 0.03960 | 0.1059 | 0.1143 | 0.410 | 0.000633 | 0.000737 |
| Error | 9 | 0.0052 | 0.361 | 0.01649 | 3.8139 | 0.0733 | 4.354 | 0.000788 | 0.03754 |
| Coefficient of variability | | 5.27 % | 3.78 % | 5.43 % | 6.29% | 3.0 % | 5.0 % | 12.76 % | 9.99 % |

* Significant P 0.05
** Highly significant P 0.01

Appendix table 2. Analysis of variance for carotene content of alfalfa

| Source of variation | Degrees of freedom | | Mean squares |
|---|---|---|---|
| Treatments | 3 | 9482 | |
| Untreated vs DDT | 1 | | 24,457* |
| Between levels of DDT | 2 | | 7.977 |
| Replicates | 3 | 27,186* | |
| Experimental error | 9 | 4,667* | |
| Sampling error | 16 | 1,818 | |

* Significant P 0.05

Coefficient of variability = 19.18% Based on experimental error
Coefficient of variability = 16.94% Based on sampling error

Appendix table 3. Composition and gross energy content of alfalfa hay, orts, feces and urine

| Sheep No. | Period No. | DDT per acre | Item | Daily wt. | Dry matter | Protein | Ether extract | Crude fiber | Ash | Nitrogen free Extract | Nitrogen per ml. | Gross energy per gm. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | lb. | | gm. | % | % | % | % | % | % | mgm. | Cal. |
| I-133 | 2 | 0 | Hay II | 1270 | 92.9 | 14.1 | 1.88 | 29.9 | 8.49 | 38.5 | - | 4065 |
| | | | Orts | 71 | 93.4 | 8.19 | 0.750 | 40.2 | 7.18 | 37.1 | - | 4109 |
| | | | Feces | 1114 | 41.3 | 4.58 | 0.911 | 17.5 | 6.04 | 12.3 | - | 1842 |
| | | | Urine | 794 1/ | - | - | - | - | - | - | 19.6 | 291 1/ |
| I-135 | 1 | 0 | Hay II | 1089 | 92.9 | 14.1 | 1.88 | 29.9 | 8.49 | 38.5 | - | 4065 |
| | | | Orts | 8 | 93.5 | 8.06 | 1.08 | 49.4 | 6.60 | 28.4 | - | 4261 |
| | | | Feces | 973 | 40.5 | 4.76 | 0.993 | 17.1 | 5.89 | 11.8 | - | 1761 |
| | | | Urine | 699 1/ | - | - | - | - | - | - | 21.5 | 319 1/ |
| I-137 | 4 | 0 | Hay II | 1258 | 92.9 | 14.1 | 1.88 | 29.9 | 8.49 | 38.5 | - | 4065 |
| | | | Orts | - | There were no orts | | | | | | - | - |
| | | | Feces | 1296 | 40.5 | 4.11 | 1.22 | 16.2 | 5.67 | 9.4 | - | 1296 |
| | | | Urine | 803 | - | - | - | - | - | - | 20.9 | 803 |
| I-139 | 3 | 0 | Hay II | 1270 | 92.9 | 14.1 | 1.88 | 29.9 | 8.49 | 38.5 | - | 4065 |
| | | | Orts | 16 | 93.9 | 9.56 | 0.870 | 45.0 | 8.11 | 30.4 | - | 3774 |
| | | | Feces | 1276 | 42.4 | 4.85 | 1.07 | 15.8 | 6.41 | 14.3 | - | 1848 |
| | | | Urine | 1266 1/ | - | - | - | - | - | - | 12.0 | 188 1/ |
| I-133 | 1 | 1 | Hay 17 | 1095 | 92.1 | 13.8 | 2.31 | 28.8 | 8.05 | 39.2 | - | 4424 |
| | | | Orts | 12 | 93.6 | 11.9 | 1.20 | 39.6 | 9.73 | 31.2 | - | 4002 |
| | | | Feces | 950 | 43.2 | 5.73 | 1.20 | 17.6 | 6.72 | 11.95 | - | 1933 |
| | | | Urine | 824 1/ | - | - | - | - | - | - | 21.0 | 294 1/ |
| I-135 | 2 | 1 | Hay 17 | 1270 | 92.1 | 13.8 | 2.31 | 28.8 | 8.05 | 39.2 | - | 4424 |
| | | | Orts | 49 | 93.2 | 9.00 | 0.570 | 41.4 | 8.32 | 33.9 | - | 3959 |
| | | | Feces | 1114 | 41.6 | 4.82 | 1.32 | 17.5 | 5.65 | 12.3 | - | 1868 |
| | | | Urine | 806 | - | - | - | - | - | - | 20.4 | 295 1/ |

Appendix table 3 continued

| Sheep no. | Period no. | DDT per acre | Item | Daily wt. | Dry matter | Protein | Ether extract | Crude fiber | Ash | Nitrogen free extract | Nitrogen per ml. | Gross energy per gm. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | lb. | lb. | gm. | % | % | % | % | % | % | mgm. | Cal. |
| I-137 | 3 | 1 | Hay I7 | 1267 | 92.1 | 13.8 | 2.31 | 28.8 | 8.05 | 39.2 | - | 4424 |
| | | | Orts | 1393[1] | There were no orts | | | | | | | |
| | | | Feces | 1040 | 34.7 | 4.08 | 1.13 | 13.2 | 5.97 | 10.3 | - | 1519 |
| | | | Urine | 4 | - | - | - | - | - | - | 16.4 | 221[1] |
| I-139 | 4 | 1 | Hay I7 | 1270 | 92.1 | 13.8 | 2.31 | 28.8 | 8.05 | 39.2 | - | 4424 |
| | | | Orts | 26 | 92.0 | 8.06 | 1.50 | 47.11 | 6.28 | 29.2 | - | 4068 |
| | | | Feces | 1153 | 39.1 | 4.67 | 1.84 | 16.[illegible] | 6.[illegible] | 10.6 | - | 1698 |
| | | | Urine | 914[1] | - | - | - | - | - | - | 19.7 | 268[1] |
| I-133 | 4 | 2 | Hay I9 | 1270 | 92.7 | 14.8 | 2.11 | 28.8 | 8.10 | 38.9 | - | 4353 |
| | | | Orts | 134 | 91.9 | 13.1 | 2.09 | 35.8 | 9.[illegible] | 31.7 | - | 3912 |
| | | | Feces | 810 | 48.6 | 5.88 | 2.14 | 20.3 | 8.30 | 11.98 | - | 1194 |
| | | | Urine | 583[1] | - | - | - | - | - | - | - | 443[1] |
| I-135 | 3 | 2 | Hay I9 | 1261 | 92.7 | 14.8 | 2.11 | 28.8 | 8.10 | 38.9 | - | 4353 |
| | | | Orts | There were no orts | | | | | | | | |
| | | | Feces | 1056 | 44.3 | 4.66 | 1.70 | 17.1 | 8.13 | 12.8 | - | 1845 |
| | | | Urine | 971[1] | - | - | - | - | - | - | 22.5 | 292[1] |
| I-137 | 1 | 2 | Hay I9 | 1270 | 92.7 | 14.8 | 2.11 | 28.8 | 8.10 | 38.9 | - | 4353 |
| | | | Orts | 28 | 93.4 | 12.8 | 1.01 | 34.2 | 11.2 | 34.2 | - | 3887 |
| | | | Feces | 1341 | 30.7 | 3.40 | 1.28 | 13.2 | 4.65 | 8.13 | - | 1361 |
| | | | Urine | 831[1] | - | - | - | - | - | - | 21.5 | 305[1] |
| I-139 | 1 | 2 | Hay I9 | 1082 | 92.7 | 14.8 | 2.11 | 28.8 | 8.10 | 38.9 | - | 4353 |
| | | | Orts | 7 | 94.2 | 8.00 | 0.575 | 50.8 | 6.18 | 29.4 | - | 4137 |
| | | | Feces | 862 | 46.4 | 5.49 | 1.40 | 18.6 | 7.76 | 13.4 | - | 1955 |
| | | | Urine | 1354[1] | - | - | - | - | - | - | 13.9 | 168[1] |

Appendix table 3 continued

| Sheep No. | Period No. | EDT per acre | Item | Daily wt. | Dry matter | Protein | Ether extract | Crude fiber | Ash | Nitrogen free extract | Nitrogen per ml. | Gross energy per gm. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | lb. | lb. | gm. | % | % | % | % | % | % | mgm. | Cal. |
| I-133 | 3 | 4 | Hay I13 | 1246 | 92.5 | 14.9 | 2.08 | 30.2 | 8.68 | 36.6 | - | 4438 |
| | | | Orts | 24 | 92.1 | 10.5 | 0.265 | 38.9 | 9.69 | 32.8 | - | 3808 |
| | | | Feces | 985 | 45.3 | 5.43 | 1.82 | 17.3 | 8.28 | 12.5 | - | 1896 |
| | | | Urine | 943 1/ | - | - | - | - | - | - | 20.4 | 310 1/ |
| I-135 | 4 | 4 | Hay I13 | 1259 | 92.5 | 14.9 | 2.08 | 30.2 | 8.68 | 36.6 | - | 4438 |
| | | | Orts | There were no orts | | | | | | | | |
| | | | Feces | 1076 | 44.8 | 5.56 | 1.73 | 18.8 | 7.30 | 11.4 | - | 1934 |
| | | | Urine | 891 | - | - | - | - | - | - | 25.3 | 284 1/ |
| I-137 | 1 | 4 | Hay I15 | 1034 | 92.5 | 14.9 | 2.08 | 30.2 | 8.68 | 36.6 | - | 4438 |
| | | | Orts | 21 | 93.8 | 10.2 | 0.550 | 44.0 | 7.73 | 31.3 | - | 3984 |
| | | | Feces | 881 | 42.6 | 5.32 | 1.22 | 17.2 | 6.57 | 12.3 | - | 1884 |
| | | | Urine | 1117 1/ | - | - | - | - | - | - | 17.2 | 202 1/ |
| I-139 | 2 | 4 | Hay I13 | 1270 | 92.5 | 14.9 | 2.08 | 30.2 | 8.68 | 36.6 | - | 4438 |
| | | | Orts | 43 | 93.7 | 9.19 | 2.88 | 14.9 | 7.90 | 28.9 | - | 4000 |
| | | | Feces | 1114 | 42.0 | 5.15 | 1.29 | 17.7 | 6.83 | 11.0 | - | 1794 |
| | | | Urine | 1271 1/ | - | - | - | - | - | - | 15.0 | 189 1/ |

1/ This is in terms of ml. of urine

Appendix table 4. Statistical analysis of the digestibility by sheep of protein in alfalfa hay when treated with various amounts of DDT

| Sheep | Digestibility Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:70 | $I_7$:69 | $I_9$:74 | $I_{13}$:68 | 281 |
| I 133 | $I_7$:64 | $I_1$:71 | $I_{13}$:71 | $I_9$:72 | 278 |
| I 139 | $I_9$:69 | $I_{13}$:69 | $I_1$:65 | $I_7$:69 | 272 |
| I 137 | $I_{13}$:69 | $I_9$:75 | $I_7$:68 | $I_1$:70 | 282 |
| Sum | 272 | 284 | 278 | 279 | 1113 |

| | Summary by treatment (alfalfa hay 1/) | | | | |
|---|---|---|---|---|---|
| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
| Sum | 276 | 270 | 290 | 277 | 1113 |
| Mean | 69.0 | 67.5 | 72.5 | 69.3 | 69.6 |

| Analysis of variance | | | | |
|---|---|---|---|---|
| Source of variation | Degrees of freedom | Sum of square | Mean square | F value |
| Total | 15 | 118 | 7.87 | |
| Hays | 3 | 53 | 17.7 | 3.34 |
| Period | 3 | 18 | 6.0 | 1.13 |
| Sheep | 3 | 15 | 5.0 | .94 |
| Error | 6 | 32 | 5.3 | |

1/ Treatments of alfalfa hay: I 1 no DDT; I 7 one pound DDT per acre; I 9 2 pounds DDT per acre; I 13 4 pounds DDT per acre.

Appendix table 5. Statistical analysis of the digestibility by sheep of ether extract in alfalfa hay when treated with various amounts of EDT

Digestibility

| Sheep | Period | | | | Sum |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | |
| I 135 | $I_1$:53 | $I_7$:49 | $I_9$:32 | $I_{13}$:29 | 163 |
| I 133 | $I_7$:55 | $I_1$:57 | $I_{13}$:29 | $I_9$:28 | 169 |
| I 139 | $I_9$:55 | $I_{13}$:43 | $I_1$:42 | $I_7$:43 | 183 |
| I 137 | $I_{13}$:50 | $I_9$:35 | $I_7$:46 | $I_1$:33 | 164 |
| Sum | 213 | 184 | 149 | 133 | 679 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 185 | 193 | 150 | 151 | 679 |
| Mean | 46.3 | 48.3 | 37.5 | 37.8 | 42.4 |

Analysis of variance

| Source of variation | Degree of freedom | Sum of square | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 1576 | 105.1 | |
| Hays | 3 | 379 | 126.33 | 4.49 |
| Periods | 3 | 964 | 321.3** | 11.39** |
| Sheep | 3 | 64 | 21.3 | 0.76 |
| Error | 6 | 169 | 28.16 | |

** Highly significant P 0.05

Appendix table 6. Statistical analysis of the digestibility by sheep of crude fiber in alfalfa hay when treated with various amounts of DDT

Digestibility

| Sheep | Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:48 | $I_7$:41 | $I_9$:50 | $I_{13}$:47 | 186 |
| I 133 | $I_7$:46 | $I_1$:45 | $I_{13}$:54 | $I_9$:48 | 193 |
| I 139 | $I_9$:48 | $I_{13}$:46 | $I_1$:46 | $I_7$:47 | 187 |
| I 137 | $I_{13}$:50 | $I_9$:50 | $I_7$:50 | $I_1$:44 | 194 |
| Sum | 192 | 182 | 200 | 186 | 760 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 183 | 184 | 196 | 197 | 760 |
| Mean | 45.8 | 46.0 | 49.0 | 49.3 | 47.5 |

Analysis of variance

| Source of variation | Degree of freedom | Sum of squares | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 136 | | |
| Hays | 3 | 43 | 14.3 | 2.57 |
| Periods | 3 | 46 | 15.3 | 2.70 |
| Sheep | 3 | 13 | 4.33 | 0.76 |
| Error | 6 | 34 | 5.67 | |

Appendix table 7. Statistical analysis of the digestibility by sheep of nitrogen free extract in alfalfa hay when treated with various amounts of DDT

Digestibility

| Sheep | Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:73 | $I_7$:72 | $I_9$:72 | $I_{13}$:73 | 290 |
| I 133 | $I_7$:74 | $I_1$:70 | $I_{13}$:73 | $I_9$:78 | 295 |
| I 139 | $I_9$:72 | $I_{13}$:73 | $I_1$:62 | $I_7$:75 | 282 |
| I 137 | $I_{13}$:71 | $I_9$:77 | $I_7$:71 | $I_1$:75 | 294 |
| Sum | 290 | 292 | 278 | 301 | 1161 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 280 | 292 | 299 | 290 | 1161 |
| Mean | 70 | 73 | 74.8 | 72.5 | 72.6 |

Analysis of variance

| Source of variation | Degree of freedom | Sum of square | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 188 | | |
| Hays | 3 | 46 | 15.33 | 1.88 |
| Period | 3 | 67 | 22.33 | 3.62 |
| Sheep | 3 | 26 | 8.67 | 1.06 |
| Error | 6 | 49 | 8.17 | |

Appendix table 8. Statistical analysis for total digestible nutrients by sheep in alfalfa hay when treated with various amounts of DDT

Total digestible nutrients

| Sheep | Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:55 | $I_7$:53 | $I_9$:55 | $I_{13}$:53 | 216 |
| I 133 | $I_7$:54 | $I_1$:53 | $I_{13}$:55 | $I_9$:57 | 219 |
| I 139 | $I_9$:55 | $I_{13}$:53 | $I_1$:49 | $I_7$:55 | 212 |
| I 137 | $I_{13}$:54 | $I_9$:58 | $I_7$:54 | $I_1$:53 | 219 |
| Sum | 218 | 217 | 213 | 218 | 866 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 210 | 216 | 225 | 215 | 866 |
| Mean | 52.5 | 54 | 56.3 | 53.8 | 54.1 |

Analysis of variance

| Source of variation | Degrees of freedom | Sum of squares | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 60 | | |
| Days | 3 | 30 | 10.00 | 3.75 |
| Periods | 3 | 5 | 1.67 | |
| Sheep | 3 | 9 | 3.00 | -1.12 |
| Error | 6 | 16 | 2.67 | |

Appendix table 9. Statistical analysis of the metabolizable energy by sheep in calories when treated with various amounts of DDT

| Sheep | Metabolizable energy<br>Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:2185 | $I_7$:2570 | $I_9$:2522 | $I_{13}$:2558 | 9835 |
| I 133 | $I_7$:2389 | $I_1$:2096 | $I_{13}$:2639 | $I_9$:2737 | 9861 |
| I 139 | $I_9$:2487 | $I_{13}$:2595 | $I_1$:1931 | $I_7$:2664 | 9677 |
| I 137 | $I_{13}$:2498 | $I_9$:2656 | $I_7$:2545 | $I_1$:2221 | 9920 |
| Sum | 9559 | 9917 | 9637 | 10180 | 39293 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 9433 | 10168 | 10402 | 10290 | 39293 |
| Mean | 2108 | 2542 | 2601 | 2573 | 2456 |

Analysis of variance

| Source of variation | Degree of freedom | Sum of squares | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 791916 | | |
| Hays | 3 | 651113 | 217043 | 17.97 |
| Period | 3 | 60144 | 20048 | 1.66 |
| Sheep | 3 | 8078 | 2693 | 0.22 |
| Error | 6 | 72481 | 12080 | |

Appendix table 10. Statistical analysis of nitrogen balance by sheep in alfalfa hay when treated with various amounts of DDT

Nitrogen balance

| Sheep | Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:2.1 | $I_7$:2.2 | $I_9$:-0.7 | $I_{13}$:-0.4 | 3.2 |
| I 133 | $I_7$:2.2 | $I_1$:4.0 | $I_{13}$:1.7 | $I_9$:1.0 | 8.9 |
| I 139 | $I_9$:-1.9 | $I_{13}$:1.5 | $I_1$:3.4 | $I_7$:1.0 | 4.0 |
| I 137 | $I_{13}$:-2.3 | $I_9$:3.4 | $I_7$:1.7 | $I_1$:3.1 | 5.9 |
| Sum | 0.1 | 11.1 | 6.1 | 4.7 | 22.0 |

Summary by treatment (alfalfa Hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 12.6 | 7.1 | 1.8 | 0.5 | 22 |
| Mean | 3.2 | 1.8 | 0.5 | 0.1 | 5.5 |

Analysis of variance

| Source of variation | Degrees of freedom | Sum of squares | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 52.15 | | |
| Hays | 3 | 22.92 | 7.64 | 5.09 * |
| Period | 3 | 15.38 | 5.13 | 3.42 |
| Sheep | 3 | 4.81 | 1.60 | 1.07 |
| Error | 6 | 9.04 | 1.50 | |

Appendix table 11. Statistical analysis of live weight gain by sheep on the alfalfa dusted with various amounts of DDT

Gain in body weight

| Sheep | Period 1 | 2 | 3 | 4 | Sum |
|---|---|---|---|---|---|
| I 135 | $I_1$:.7 | $I_7$:0.3 | $I_9$:20 | $I_{13}$:-5.0 | 3.0 |
| I 133 | $I_7$:1.1 | $I_1$:-0.6 | $I_{13}$:6.5 | $I_9$:-4.0 | 3.0 |
| I 139 | $I_9$:-0.5 | $I_{13}$:1.0 | $I_1$:6.5 | $I_7$:-2.0 | 5.0 |
| I 137 | $I_{13}$:0.6 | $I_9$:-1.1 | $I_7$:6.5 | $I_1$:-5.0 | 1.0 |
| Sum | 1.9 | -0.4 | 26.5 | -16.0 | 12.0 |

Summary by treatment (alfalfa hay)

| | $I_1$ | $I_7$ | $I_9$ | $I_{13}$ | |
|---|---|---|---|---|---|
| Sum | 1.6 | 5.9 | 1.4 | 3.1 | 12 |
| Mean | 0.4 | 1.5 | 0.4 | 0.8 | 3 |

Analysis of variance

| Source of variation | Degrees of freedom | Sum of squares | Mean square | F value |
|---|---|---|---|---|
| Total | 15 | 241.7 | | |
| Hays | 3 | 3.2 | 1.06 | .09 |
| Period | 3 | 168.1 | 56.03 | 4.91 |
| Sheep | 3 | 2.0 | .7 | 0.006 |
| Error | 6 | 68.4 | 11.4 | |