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94 THE BUILDING OF TURKEYS
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They may not look it, but turkeys are relatively frail birds. Researchers at Snow Field Station are therefore trying to help producers cater effectively to their nutritional, housing, and general-environmental needs.

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114 ELISA—A New Method to Diagnose Disease
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ABOUT THE COVER
When livestock and poultry shift from healthy to sick, the culprit is likely to be bacterial or viral pathogens. Another likely candidate is the absence of necessary elements in the diet.

Commercial turkey production in Utah was once located in 12 counties from Box Elder and Cache on the north to Washington on the south. Today, it is limited to one part of central Utah. Producers face keen competition from several areas of the U.S. Utah does not produce all the feed required and transporting grain and soybean meal from the midwest is a serious obstacle. In an effort to remain competitive, Utah turkey producers have sponsored research at all three universities in the state and have given support to the Snow Field Station at Ephraim, Utah, where various management practices are tested. If the industry in Utah is to remain viable, producers must learn as much as possible about the nature of turkeys and find ways to solve several problems.

Advances by Biologists
Understanding the turkey requires that we understand some of the significant advances made by biologists during the last 30 years dealing with the nature of living organisms. Francois Jacob's book The Logic of Life: A History of Heredity summarizes these findings. A very important and basic realization is that living organisms are complex and highly
ordered systems that are programmed to reproduce. The turkey poult is programmed to take food from its surroundings, grow to maturity and then cooperate with an individual of the opposite sex to produce more turkeys. A turkey producer must discover the way to do this most efficiently and to avoid disruptions, such as disease, inadequate nutrition, and inefficient management practices that retard the whole process.

As an organism grows, it increases its mass of ordered matter and the degree of order of that mass increases. This at first seems to contravene the second law of thermodynamics, which states that in any isolated system undergoing spontaneous change, entropy (disorder) must increase. Some thought that living objects possessed a vital force not found in non-living. Upon further examination, it was realized that the increase in order in a living system is only a local phenomenon. If the whole system is considered (that is, the organism and matter flowing through it) entropy does increase. An organism cannot be a closed system; it must continually take in food (with low entropy) and excrete wastes (with high entropy). Today we regard living organisms as a site of a triple flow of matter, energy and information. Food must provide all three.

The term "information" has different meanings. Generally, information and entropy are regarded as two sides of the same coin. Entropy is a measure of the disorder and man’s ignorance of an organism’s internal structure, while information is a measure of the order of an organism and man’s knowledge of its structure. One is the negative of the other.

About 23 of the elements of the periodic table have roles in living organisms and must be a part of this flow of matter. Organic compounds composed of carbon, hydrogen, oxygen, sulfur and nitrogen form the bulk of this flow. Entering compounds must possess energy in the form of chemical bonds that the organism can transduce to the "high energy phosphate bond" form. The order found in a living system cannot be produced without using this energy.

The cell is the basic unit of all living organisms. Several types of compounds must be present in a living cell. But, the properties of living organisms are ultimately based on two entities: what the biochemists call proteins and what the geneticists call genes.
The Role of DNA and Protein of Cells

Proteins form the basic structural materials of cells. More important, they are the catalysts (enzymes) of living organisms that carry out chemical reactions. Some have the ability to sense certain conditions in the cell, change their shape and activity according to the conditions, and thus direct the flow of matter through the cell. Thousands of enzymes are required in a living cell and each is continually undergoing a rapid turnover. However, a new molecule of an enzyme is not produced by copying an old one. Proteins are organized from instructions found in the DNA of the genes. The DNA is the substance copied each time a cell divides giving each new cell a copy of this information.

DNA and protein are both linear polymers. Proteins are composed of 100 to 10,000 amino acid residues of 20 species. The structure and properties of a protein are determined by the sequence or linear order of the amino acid residues. DNA has alternating phosphate and deoxyribose (sugar) residues as its backbone. One of four organic bases is attached to each sugar residue; these give DNA its information carrying ability. A DNA chain can be considered coded instructions for the ordering of amino acid residues along a protein chain. A sequence of three bases is needed to specify which amino acid will be found in a given position. The genetic code is the rule that specifies which of 20 amino acids corresponds to each of the 64 possible triplets of bases.

Growth of an organism requires that it synthesize DNA and protein. Phosphate for DNA must come from the diet; the sugar and bases are usually present in the diet or are made in the normal flow of matter through the system. Eight of the 20 amino acids also are produced in the normal flow if the matter contains adequate amino nitrogen. Ten amino acids are not produced; these must come from the diet and are termed indispensable amino acids (IAAs). The amino acid cysteine can be produced in this flow only by using the IAA methionine; tyrosine can be produced from phenylalanine. Each such pair is often viewed as one entity in considering dietary requirements if the strictly IAAs provide at least half of the total. The food entering a living organism must also provide vitamins, minerals and at least one essential fatty acid. Not one of these factors can be ignored.

### Specific nutrients in a turkey poults diet are critical to its health

#### Feeding Pouls

Most of the dollar cost of growing farm animals goes to provide energy and protein. Our work with chicks indicated that about 550 KJ of energy were required per kilogram of metabolic body weight per day just to maintain this living system. Producing rapid growth required additional inputs. The efficiency and rate of this gain depends on the balance among the nutrients entering the system. Because the turkey poults' diet is so critical to its health, the poult has been a valuable tool in our research into factors affecting rate and efficiency of gain.

We first studied the effect of dietary levels of the IAAs on rate of gain and feed efficiency in pouls. We knew that pouls required at least 28 percent good protein in their diet to achieve a near-maximum gain and feed efficiency. A basic diet was formulated with 28 percent protein and a total of 14 percent IAAs in what was considered well balanced proportions. Ingredients in this diet were selected to allow us to alter the level of each IAA from a clearly inadequate to a more than adequate level. Graded levels of each IAA were then fed to determine the minimum level which would allow near maximum performance. Only the level of the single IAA being studied was reduced in an experiment. These are the minimums found (given as percentage of the diet):

<table>
<thead>
<tr>
<th>IAA</th>
<th>Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arginine</td>
<td>1.60</td>
</tr>
<tr>
<td>Histidine</td>
<td>0.58</td>
</tr>
<tr>
<td>Lysine</td>
<td>1.68</td>
</tr>
<tr>
<td>Leucine</td>
<td>1.86</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>1.10</td>
</tr>
<tr>
<td>Valine</td>
<td>1.20</td>
</tr>
<tr>
<td>Phenylalanine and tyrosine</td>
<td>1.80</td>
</tr>
<tr>
<td>Methionine and cystine</td>
<td>1.04</td>
</tr>
<tr>
<td>Threonine</td>
<td>1.00</td>
</tr>
</tbody>
</table>

**TOTAL** 12.12

These totaled only 12.12 percent compared with the 14 percent in our starting basal diet. In the next experiment, graded total levels from 12.12 to 14 percent were fed in the proportions given above. Results are plotted in Figure 1. When a 28 percent protein diet contained the minimum levels of all IAAs given above, gain was about 84 percent and feed efficiency 90 percent of that noted when all IAAs were increased 15.15 percent (14 percent total). Yet any one could be reduced from the higher to the minimum given, without a significant reduction in performance. Thus, the poult's overall IAA requirement is greater than the sum of the individual minimums.

The Snow Field Station has conducted a test each year since the early sixties to compare commercial turkey strains. It soon became apparent that strains from breeder A used less feed to produce a pound of gain than those from breeder B although both turkeys were widely used in the industry. This raised a question as to whether strains that differed in feed efficiency also differed in programming or the degree or kind of order developed. As an indication that such differences exist, we expected to find differences in other measures of performance and examined reports of the tests. R. E. Warnick, director of Snow Field Station, identified the strains for us.

In these tests, pouls of all strains are identified but raised together for the first eight weeks. Feed efficiency by strain is reported only for the period from eight weeks of age to market. Mean feed/gain ratios noted with four strains of interest in the 1980, 1981 and 1982 tests are given in Figure 2. Turkeys of strain B required about 8 percent more feed per pound of gain during the growing period than those of the two A strains. However, B strain pouls hatched better, showed faster early growth and much better survivability (Figure 2). Some of this latter difference could be due to a higher disease load carried through the egg to A strain pouls, but the intermediate performance of the pouls produced by crossing the two strains by breeder A suggests a strain difference. Growth rate and mortality after eight weeks of age were similar for the four strains. Strain B turkeys yield less meat parts and more skin if the carcasses are further processed.

All evidence available to us indicates that strain B pouls are more vigorous than those of strain A. But, if a grower wants to grow pouls programmed for this vigor, he must pay for it by using more feed. Must pouls use more feed if they are programmed for greater vigor? This important question is not answered. B. K. McNab suggested that a similar relationship is found among mammalian species. He stated that reproductive output increases with an increase in energy expenditure for homeostasis.
The young of species with the higher energy expenditure can be programmed for more rapid postnatal growth.

**IAAs and Strain Differences**

Does the overall level of IAAs in the diet make as much difference to strain B poults as to strain A? A recent test was designed to answer this by feeding the following diets.

1. A diet meeting the minimum requirements given previously, plus a little more (12.75 percent total), and 25.35 percent protein.
2. As 1, plus three dispensable amino acids to increase protein to 28 percent.
3. A diet with added levels of all IAAs and three dispensable amino acids to make a total of 14 percent IAA and 28 percent protein.

These diets were fed to poults of strains A₁, A₂ and B from one to three weeks of age with the results given in Figure 3. Feed/gain ratio was greater with strain B even at this age, although the strain B gained more. Adding extra dispensable amino acids alone seemed to increase gain slightly but did not influence feed efficiency. The greatest and most efficient gain was attained with the higher levels of both IAAs and protein. Increasing both tended to improve performance more with strain A than B.

The difference noted in performance can be better understood if we consider the nature of a living organism as now presented by biologists. Protein, DNA and other substances are all essential for life, but not one of these has life by itself. The properties of living organisms are not the sum of the properties of the parts.

A turkey poult’s overall IAA requirement is greater than the sum of the individual minimum requirements although these are important. Even the levels of the dispensable amino acids as a whole affects performance. Poult performances were best with the proportions and levels near those in diet 3 above, but one IAA can be reduced as much as 15 percent without inducing a significant decrease. Decreasing the level of one IAA increases the efficiency of its utilization, but decreases the efficiency of utilization of other nutrients.

The turkey poult’s diet is more critical to its well-being than is that of chicks or other farm animals. This made the poult a valuable means of demonstrating these dietary/growth relationships. Turkey growers recognize that several other phases of management are also especially critical with poults.

**References**


**ABOUT THE AUTHOR**

Jay O. Anderson, professor, Animal, Dairy and Veterinary Sciences Dept., received his PhD at the University of Maryland. His research deals primarily with amino acids in nutrition of chicks and turkeys.
Turkeys were first raised in Sanpete County in the 1920s, with the first commercial flock of 500 birds being raised in 1927. The number of birds raised increased to over 35,000 in 1935. During that year, the first modern turkey processing plant in Utah was constructed near Moroni and operated by the Utah Poultry Producers Cooperative headquartered in Salt Lake City.

As the industry began to expand, so did their problems such as disease, feeds, equipment, and husbandry practices. Under the leadership of a vocational agricultural teacher and other adult education leaders, workshops were held to teach better nutrition and management practices. Leading experts from Utah State University and throughout the nation were invited to speak at these problem-solving meetings.¹

The Moroni Feed Company was officially incorporated under the cooperative statute of Utah in 1938. Since then close ties have been maintained between the Company and the personnel of the USU Agricultural Experiment Station and the USU Extension Service. Utah's turkey industry has many unique problems, many of which are associated with the high altitude and normally dry conditions of the area. To help solve them, research projects were started at the Snow Field Station in Ephraim in the late 1950s and expanded in the early 1960s. With the cooperation of Utah State University Agricultural Experiment Station, Animal, Dairy and Veterinary Sciences Department, Utah's turkey industry, and Snow College, a very productive turkey research facility has been developed with a feed mill, a large confinement barn, range facilities, and a new solar building for brooding.

Feeding Schedules

The seasonal feeding schedules (Table 1) used by the Utah turkey growers were originally developed by Donald Dobson at the Snow Field Station and have played an important role in helping Utah's turkey industry remain competitive. These feeding schedules require rations higher in protein and lower in added fat than those used in other turkey growing areas.

In 1981, rations and feeding schedules used in Utah were compared with those used in Iowa and Minnesota. The turkeys on the Utah schedule were 1.5 lbs. heavier than birds fed according to the midwest schedules.²

Animal fat is added to turkey diets as an energy source and has been shown to improve growth rate and feed efficiency. The many tests with added fat conducted in Utah have shown little or no value when levels of fat exceed 2 percent of the diet. An example of test results from a study reported in 1981 are listed in Table 2.

Strain Test

Research into strain evaluation has continued at the Snow Field Station for many years. Eggs from different strains of turkeys are brought to the Moroni Hatchery and hatched. The poult's are then taken to the research facility and grown out. All strains are fed the same type of diets as commercial birds in this area following the same feeding schedules. The birds are processed at two different ages for both sexes. A large sample of whole-bodied, A grade birds are further processed to provide meat yield data for all strains.

Table 3 lists partial results obtained from the hens of four strains tested in 1981. These data contain most of the extremes found in this test. The birds from strain 3 were smaller at 17 weeks but had the best feed conversion and meat yield. In contrast, strain 10 birds were the heaviest and had the highest feed conversion but the lowest meat yield.

At 23 weeks, strain 6 hens had the best feed conversion and white meat yield, but the lowest thigh meat yield. Feed conversions were quite high with strains 8 and 10, with lower white meat yields. Strains 3 and 6 are slower maturing strains that tend to produce more white meat with more efficient feed utilization at older ages.

The data obtained from the toms (Table 4) at 20 weeks followed a similar pattern to that of the hens; however, there was a wider range in live weight differences at both ages. Strains 3 and 6 both gained significantly more be-

 FIGURE 1. Single slope shelter used for range turkeys.

 FIGURE 2. Portable shelter with gabled roof.

 FIGURE 3. Flat-roofed bowery-type shelter.

 FIGURE 4. Pole barn used for semi-confinement rearing of turkeys.

 FIGURE 5. Large feeders inside the research pole barn at Snow Field Station.

 FIGURE 6. View of solar panels on the new solar building.

 FIGURE 7. Casablanca-type fans are used for air circulation in the solar building.

 FIGURE 8. Brooding pen showing brooder, water containers, and feeders.

 FIGURE 9. Heat ducts provide an added source of heat.

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The data obtained from the toms (Table 4) at 20 weeks followed a similar pattern to that of the hens; however, there was a wider range in live weight differences at both ages. Strains 3 and 6 both gained significantly more be-
between 20 and 28 weeks than did strains 8 and 10, with better feed conversion and meat yields.

No one strain of turkey best fits the needs of today's market demand. Data from the USU tests have helped Utah's turkey growers choose which strain to raise to be processed at a given age for a particular type of market.

During the past 15 years, the turkey industry has completely converted from the bronze to the white bird because of consumer preferences and the ease of processing the white bird. A strain test conducted the past three years has shown that the bronze birds are slower to mature. Both hens and toms are slightly smaller at 17 and 20 weeks of age than the white birds. When processed at these ages the grade of bronze birds is much lower than that of the white birds. At the older ages they process much better, but black pigment spots are noticed. The bronze are heavier at more mature ages with much better feed conversion. No differences are noted in their resistance to disease, but they do seem to tolerate extreme weather conditions, both hot and cold, better than do the white birds.

**Dust and Temperature Control**

Since the early years, turkey production units have become larger, open range rearing is decreasing and many operations are using large confinement or semi-confinement buildings. With the resultant large concentrations of birds in small areas, housing and management of dust and disease can be crucial.

In Utah, turkey production is seasonal. Studies conducted at Snow Field Station have shown that the growing season can be extended by using large pole barns.

Dust in a poultry confinement building is primarily composed of pulverized fecal material, feed particles as well as feather and epidermal fragments. Airborne particles in turkey confinement buildings can mean an unhealthy environment for both the birds and the producer. Research as early as the 1980s suggested that infections in animals could be caused by inhalation of bacilli-infected dust particles. Recent research has indicated that many turkey disease problems such as *E. coli* infection, aspergillosis and airsacculitis are all transmitted by dust.

Age of birds, bird density, sex of birds, time of day, season of year, humidity, air pressure, temperature, lighting system, and type of ventilation all affect dust levels. Any dust control program must take these factors into consideration.

Studies at Snow Field Station have found that the best way to control dust in confined areas is with a combination of sprinklers and foggers. In our research facility, the location of foggers was limited. One fogging line was placed near the front of each pen over the feeder with two nozzles spraying away from the feeder. A ceiling-mounted sprinkler covers the back half of each pen. Foggers are controlled by a thermostat, while the sprinklers are controlled by a time clock that turns on for five minutes at 3 p.m. each day. We arbitrarily chose 3 p.m., as that is the hottest part of the day, and the sprinkling produces some cooling effect. In our large pole barn the temperature has been reduced 10° to 15° F without the use of fans.

Careful management is very important when water is used for dust control. Too much water can increase disease problems due to a rapid buildup of mold, fungus, bacteria, etc. There is also an increase in flies when levels of moisture in the litter exceed 35 percent.

The initial system design for a commercial operation should consider condensation on water lines, overlapping of foggers and sprinklers, and proper installation so that wet spots do not occur. Foggging and sprinkling equipment must be cleaned regularly. Where water is hard, fogger nozzles should be cleaned more often, using a cloth dampened with mild acid or vinegar.

In a test conducted at Snow Field Station in 1980, Oriopp 91 hens grown in confinement pens for 118 days weighed 14.12 lbs. with a 2.59 feed conversion. Similar birds on the open range weighed 14.21 lbs. with a 2.63 conversion. For the last two weeks of this test, high and low temperatures were 96° and 50° F. Temperatures inside the building were maintained between 60° and 80° F.

**Housing and Shelter**

In 1979, a study was conducted from June through September, when the weather conditions were not extremely cold and rainfall was about 1 inch below the long-time average. Four types of shelters were studied: (1) portable range shelter with a single-slope roof and raised 2" x 4" perches; (2) a double-sloped roof, portable shelter with 4" x 6" perches near ground level; (3) a permanent, flat-roofed shelter with no perches; and (4) confinement in a pole barn with wood shavings for litter.

No difference in bird weight and feed efficiency were noted between shelter types 1, 2, or 3. Turkeys in the confinement building were smaller but had better feed conversion. With improved management, we have achieved equal or better weights in the confinement barn in 1982 and 1983 studies.

There was a wide variation in the number of A grade birds and in the reasons for downgrading with all shelter types (Table 5). The lowest grade was noted in shelter type 1. This was due primarily to the high number of birds that developed breast blisters. Grade for type 2 birds was significantly better than for type 1, but was lower than for types 3 and 4 birds. Grades for types 3 and 4 birds were similar, but there were differences in the reasons for downgrading. Type 4 birds had a higher incidence of body trimming and wings off, while type 3 birds had more legs off and breast blisters. Other research has shown that downgrading due to body trimming can be reduced by toe trimming and/or light control.

**Solar Building**

In 1983 a new solar facility was completed at the Snow Field Station. The new solar building is being used in brooding studies with turkeys. Its concept, however, could be used with a wide variety of agricultural projects. The unique feature of the building is the passive solar collector. The collector is a reinforced concrete wall painted black. The wall is 12 inches thick, 10 feet high and 120 feet long, with sealed double glass panels in front of the wall. The building is 40 feet wide and 120 feet long, covered exteriorly with plywood and galvanized sheet metal and internally with either waterproof plaster board or plywood. Fiberglass insulation rated R-19 was used in the exterior walls and blown in cellulose (R-40) in the roof area. The heat passing through the solar wall is circulated within the building with "casablanca" type fans (with variable speed controls) mounted 18 inches below the ceiling. Air exchange is accomplished with four reversible type fans mounted in the ceiling and eight louvered openings in the north wall. These fans are controlled.
warwick, Robert E. Effect of midwest and Utah diets on considerably reduced propane usage. Started June 21st. The building has been modified in the building on March 3, 1983. We were very pleased with the building is divided into three rooms, two brooding rooms 40 x 50 feet and a service room 20 x 40 feet. The brooding rooms are divided into eight pens for nutritional type studies. Each room can be operated completely independent of the others. All external energy sources are metered separately. Each room is equipped with a gas furnace and duct work such that hot-house brooding trials can be conducted or compared with a wide variety of conventional type brooders. The building is not a total blackout type, but its lights can be adjusted as to intensity and time. The initial flock of birds were placed in the building on March 3, 1983. We had some problems with equipment, etc. but were very pleased with the overall results. The second flock of poulters were started June 21st. The building has proved very energy efficient and has considerably reduced propane usage.

REFERENCES

ABOUT THE AUTHOR
Robert E. Warnick is a research assistant professor in the Animal, Dairy & Veterinary Science Department, USU, and director of the Snow Field Station, Ephraim. He received his PhD from USU in poultry nutrition and is working with the turkey industry to improve commercial turkey production through breeding and specialized feeding programs.

TABLE 1. Turkey feeding schedules.

<table>
<thead>
<tr>
<th>Age Weeks</th>
<th>Early</th>
<th>Mid</th>
<th>Late</th>
</tr>
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<tbody>
<tr>
<td>0-2</td>
<td>28%</td>
<td>28%</td>
<td>28%</td>
</tr>
<tr>
<td>2-8</td>
<td>26%</td>
<td>26%</td>
<td>26%</td>
</tr>
<tr>
<td>8-10</td>
<td>24%</td>
<td>24%</td>
<td>24%</td>
</tr>
<tr>
<td>10-12</td>
<td>22%</td>
<td>22%</td>
<td>24%</td>
</tr>
<tr>
<td>12-14</td>
<td>22%</td>
<td>22%</td>
<td>18%</td>
</tr>
<tr>
<td>14-16</td>
<td>20%</td>
<td>20%</td>
<td>18%</td>
</tr>
<tr>
<td>16-19</td>
<td>20%</td>
<td>20%</td>
<td>16%</td>
</tr>
<tr>
<td>19-22</td>
<td>18%</td>
<td>16%</td>
<td>16%</td>
</tr>
<tr>
<td>22-25</td>
<td>16%</td>
<td>16%</td>
<td>14%</td>
</tr>
<tr>
<td>Over 25</td>
<td>16%</td>
<td>14%</td>
<td>12%</td>
</tr>
</tbody>
</table>

TABLE 2. Turkey performance with added dietary fat.

<table>
<thead>
<tr>
<th>Fat Level</th>
<th>Live Weight lbs.</th>
<th>Grade</th>
<th>Feed Conversion</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>2%</td>
<td>35.80</td>
<td>77.15</td>
<td>3.53</td>
<td>9.32</td>
</tr>
<tr>
<td>4%</td>
<td>34.94</td>
<td>70.14</td>
<td>3.65</td>
<td>9.31</td>
</tr>
</tbody>
</table>

TABLE 3. Comparison of strains of hen turkeys at two ages.

<table>
<thead>
<tr>
<th>Strain</th>
<th>Age Weeks</th>
<th>Live Weight lbs</th>
<th>Feed/Grain Ratio</th>
<th>White Meat %</th>
<th>Thigh Meat %</th>
<th>Total Meat Parts %</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>17</td>
<td>13.60</td>
<td>2.62</td>
<td>29.5</td>
<td>10.6</td>
<td>62.0</td>
</tr>
<tr>
<td>3</td>
<td>23</td>
<td>18.65</td>
<td>3.17</td>
<td>32.1</td>
<td>11.9</td>
<td>65.2</td>
</tr>
<tr>
<td>6</td>
<td>17</td>
<td>14.51</td>
<td>2.72</td>
<td>28.3</td>
<td>10.4</td>
<td>61.0</td>
</tr>
<tr>
<td>6</td>
<td>23</td>
<td>20.00</td>
<td>3.14</td>
<td>32.5</td>
<td>11.2</td>
<td>64.7</td>
</tr>
<tr>
<td>8</td>
<td>17</td>
<td>13.76</td>
<td>2.78</td>
<td>29.1</td>
<td>10.3</td>
<td>61.3</td>
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<td>8</td>
<td>23</td>
<td>18.65</td>
<td>3.31</td>
<td>30.4</td>
<td>11.4</td>
<td>63.0</td>
</tr>
<tr>
<td>10</td>
<td>17</td>
<td>15.17</td>
<td>2.82</td>
<td>27.0</td>
<td>10.2</td>
<td>59.5</td>
</tr>
<tr>
<td>10</td>
<td>23</td>
<td>20.06</td>
<td>3.39</td>
<td>28.5</td>
<td>12.4</td>
<td>62.8</td>
</tr>
<tr>
<td>Ave.</td>
<td>17</td>
<td>14.26</td>
<td>2.74</td>
<td>28.5</td>
<td>10.4</td>
<td>61.0</td>
</tr>
<tr>
<td>Ave.</td>
<td>23</td>
<td>19.34</td>
<td>3.25</td>
<td>30.9</td>
<td>11.7</td>
<td>63.9</td>
</tr>
</tbody>
</table>

TABLE 4. Comparison of strains of tom turkeys at two ages.

<table>
<thead>
<tr>
<th>Strain</th>
<th>Age Weeks</th>
<th>Live Weight lbs</th>
<th>Feed/Grain Ratio</th>
<th>White Meat %</th>
<th>Thigh Meat %</th>
<th>Total Meat Parts %</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>20</td>
<td>23.28</td>
<td>2.78</td>
<td>30.6</td>
<td>11.4</td>
<td>66.3</td>
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<td>28</td>
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<td>3.58</td>
<td>32.6</td>
<td>11.1</td>
<td>65.5</td>
</tr>
</tbody>
</table>

TABLE 5. Causes of downgrading as affected by shelter type.

<table>
<thead>
<tr>
<th>Shelter Type</th>
<th>Trim %</th>
<th>Wings Off %</th>
<th>Legs Off %</th>
<th>Breast Blister %</th>
<th>Other %</th>
<th>A Grade %</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>9.07</td>
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<td>1.78</td>
<td>36.64</td>
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<td>45.3</td>
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<td>2</td>
<td>6.91</td>
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<td>1.38</td>
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<td>5.52</td>
<td>3.37</td>
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<td>68.7</td>
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<tr>
<td>4</td>
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<td>7.85</td>
<td>1.71</td>
<td>5.33</td>
<td>6.30</td>
<td>67.3</td>
</tr>
</tbody>
</table>

1. Portable, raised perches
2. Portable, low perches
3. Permanent shed, bare ground
4. Pole barn, wood shavings
FIGURE 3. Cross-section of Russian olive leaf showing the localization of chloroplasts in palisade parenchyma cells. Chloroplasts appear as elliptical green objects at the periphery of the cell. Photo courtesy of Dr. Richard Mueller.

FIGURE 4. Electron micrographs of *Spiraea vanhouttei* leaf mesophyll tissue. a) Normal green tissue chloroplasts. Note grana consists of up to 20 thylakoids. b) Iron chlorotic tissue chloroplasts. Note lack of developed grana. c) and d) Higher magnifications of normal and iron chlorotic grana respectively. Bar equals 1 um in a) and b), and 0.1 um in c) and d). Photo courtesy of John Manwaring and originally appeared in Pushnik et al 1983.

FIGURE 8. Tomato variety difference in chlorosis sensitivity. (Floradel, resistant* and T3820, sensitive).

FIGURE 9. Turf grass displayed greenery after iron-sol application. Photo courtesy of Dr. David James.

FIGURE 11. Effect of foliar sprays (iron-metalosates) on raspberry. Plants show chlorosis on older leaves and greening on new leaves after foliar sprays. Photo courtesy of Dr. LeGrande Shupe.
Nutrition of plants is primarily a question of minerals. Unlike animals, plants are autotrophic, capable of self-nourishment based on light and inorganic chemicals. Through the process of photosynthesis, plants are able in the presence of carbon dioxide (CO₂) and light, to synthesize all the organic components (e.g., vitamins, fats, amino acids, etc.) they require for growth and development. The 16 essential (acquired from the environment) mineral nutrients are listed in Table 1.

Function of Nutrients

Three of the essential elements (carbon, hydrogen and oxygen) are taken primarily from the air and water. The other 13 are normally absorbed from the soil by plant roots. None of these elements is more essential than any of the others, regardless of the amounts required. Growth and development of the plant would not be completed if any one of the 16 essential mineral elements were lacking.

Sodium may have a beneficial effect on all plants when potassium is limited. It also may be required for growth by certain halophytes such as Halogeton glomeratus and Atriplex vesicaria. Selenium may be needed by certain species of Astragalus that accumulate this element. Cobalt is part of vitamin B₁₂, which is required for nitrogen fixation. Consequently, legumes must have cobalt when there is no “fixed” nitrogen available. Cobalt is nonessential when “fixed” nitrogen is available.

Nitrogen, phosphorus and potassium are the three fertilizer elements most often added to the soil for crop production. Farmers annually use over 13 million tons of actual nitrogen, 7 million tons of phosphorus and 6 million tons of potassium as fertilizer in the United States. Nitrogen is a component of all amino acids, which are necessary for the making of protein. Phosphorus is required for the formation of phosphate intermediates that are the principal agents in which the energy of cellular oxidation is made available to other cellular processes and reaction. Potassium stimulates processes such as the opening and closing of stomata (pores in the leaves that allow water vapor to leave and carbon dioxide, so critical to photosynthesis, to enter). It is also essential for the functioning of many enzymes.

Iron: A Micronutrient Requirement

Iron functions in a plant in many ways, but a lack of iron is most often evidenced by a yellowing of the leaves commonly referred to as iron-chlorosis. Although iron is abundant in nearly every soil, its availability may be so low (particularly in calcareous soils) that plants are unable to absorb enough to sustain normal growth and development. Iron-chlorosis involves a reduction in the chlorophyll content of leaves. The resultant decrease in photosynthesis directly affects plant growth and development and reduces productivity for economic uses by man.

Iron chlorosis affects the production and well being of many kinds of plants including field crops, nursery stock, large and small fruits, as well as forage and turf plants. In addition, many economically useful species of plants are currently unable to grow in chlorosis-endemic areas.

Photosynthesis

Chlorophyll is necessary for the formation of sugars and energy in the leaves of plants (photosynthesis). These sugars are converted to various compounds such as organic acids, proteins, and fats largely using the energy produced in photosynthesis. Photosynthesis can be divided into dark reactions and light reactions, Figure 1. In the light reaction, bundles of light (quanta) are absorbed by chlorophyll. Electrons are excited from chlorophyll complexes and raised to a higher energy state, then passed through the electron transfer system (Figure 2).

These electrons are excited first at photosystem I (PSI) and then at photosystem II (PSII). The energy from the excited electrons is captured in the energy compound adenosine triphosphate (ATP) and (NADPH). These energy compounds are used to drive the dark reaction. The dark reaction in photosynthesis utilizes carbon dioxide, which provides the carbon for the sugar molecules and ultimately all organic molecules necessary for the growth and development of plants.

Light and carbon dioxide thus are the limiting factors for photosynthesis under normal conditions. Under mineral stress conditions, plants may be unable to absorb sufficient quantities of minerals such as iron (e.g. in calcareous soils). Chlorophyll may then become limiting, with its concentration correlated directly to the amount of “functional” iron available. Iron: chlorophyll: photosynthesis: growth and development are intimately related and dependent on each other.

Iron Stress Effects on Chloroplasts

Photosynthesis, involving the capture and conversion of light energy to a biologically useful form of energy, is associated with a specific subcellular organelle of leaf tissue, the chloroplast. Light microscopic examination of a cross-sectional view of green leaf tissue reveals that the chloroplasts are primarily localized in a group of columnar cells in a central region of the leaf, the palisade parenchyma (Figure 3). The chloroplasts appear as elliptical objects around the periphery of the cell, measuring 5 to 10 micrometers in length and 2 to 4 micrometers in diameter.

An electron micrograph from our laboratory details the basic structure of chloroplasts.
the chloroplast and provides a view of the framework within which photosynthesis takes place (Figure 4). This organelle is surrounded by an outer, double-membrane envelope. Within the envelope is a series of parallel green membranes (thylakoids). Regions of the thylakoid membrane system are associated in stacks, resembling a stack of coins. These stacked areas are known as grana. They concentrate the chlorophyll and proteins that are necessary for photosynthesis into the most efficient arrangement. Suspending the internal membrane system is a colorless matrix (stroma). The stroma is an organically rich soup containing the enzymes responsible for CO₂ fixation (dark reaction in photosynthesis), as well as chloroplastic DNA coding for some of the proteins found in the thylakoids.

When light energy is trapped by an antenna chlorophyll molecule of photosystem II, it is transferred to others within the same cluster (photosystem) until it reaches a special pair of chlorophyll molecules (reaction center) where the energy is used to move an electron to a higher energy molecule in the electron transport scheme (Figure 2). At this point the electron flows energetically “down hill” through a series of electron transport molecules until it eventually arrives at the reaction center of photosystem I, where a second photochemical event energetically elevates the electron to the final membrane bound carrier, ferredoxin (Fd).

Ferredoxin is reduced by the electron transport chain, and can then contribute the electron to an enzyme (ferredoxin-NADP reductase) that is capable of catalyzing the biologic storage of the light energy as reducing power (NADPH). In addition to reducing power, another biologically rich energy form is chemiosmotically generated during the passage of electron, ATP. Both ATP and NADPH are critical to the dark reactions of photosynthesis that fix atmospheric CO₂ as sugar and are dependent on efficient light capture and electron transport. The photosynthetic system resupplies itself with electrons by enzymatically splitting water, which results in the release of oxygen.

Iron-stressed plants present a different picture in terms of photosynthetic capacity. Examination of iron-chlorotic tissue shows that the chloroplasts have undergone structural modification, principally a reduction in the number of grana (Figure 4). This characteristic would result in a decrease in the efficiency of photosynthesis. Associated with this decline in granal stacking are reductions in chloroplast protein and (most significantly) a reduction in the chlorophyll content. The loss of these compounds may be responsible for the structural changes.

The loss of chlorophyll content reduces the overall quantity of light energy that can be conserved. As a consequence of this reduction in light-driven energy conservation, the plant is unable to maximally generate sugar from CO₂. This reduction in photosynthetic sugar affects all aspects of growth and results in substantial yield losses.

Biochemical Sites of Iron Involvement

Almost all of a plant’s total iron is housed in its chloroplasts, the plant organelles responsible for photosynthesis. Chloroplastic iron is found in a number of different forms: phytoferretin, a non-toxic proteinaceous iron storage compound; various cytochromes, essential for electron transport; iron-sulfur proteins (ferredoxin), also necessary in electron transport. The remainder of a plant’s accumulated iron is distributed in the cytoplasm and other organelles which contain additional heme and/or iron-sulfur proteins.

Correlation between chlorophyll content and leaf iron has been subjected to a number of investigations. Examination of iron chlorotic leaves has revealed that often the total iron levels were equal to or higher than those of green plants (Table 2). These observations lead to the concept of a “chemically active” iron fraction required for chlorophyll biosynthesis.

Chlorophyll content is adversely affected by iron stress as shown in Table 2. Observations of reduced amounts of chlorophylls and carotenoids have been reported in iron-stressed tissues (Shetty and Miller, 1966).

Current knowledge of how iron functions in the biosynthesis or maintenance of chlorophyll is somewhat limited. It is known that no precursor for chlorophyll formation in the porphyrin biosynthesis pathway contains iron, but the synthesis of one or more of these compounds may require iron. The biosynthetic pathway for chlorophyll (Figure 5) indicates particular reactions where iron may be involved.

Iron has been shown to be a factor in the coproporphyrinogenase reaction. Hsu and Miller (1970) were the first to successfully isolate coproporphyrinogenase from higher plants. This enzyme was found to be inhibited by α-phenanthroline and EDTA, both are iron chelators. Maximal activity of the purified enzyme was shown to require 0.5 mM ferrous iron (Figure 6).

Participation of inorganic iron in the conversion of Mg-protoporphyrin IX monomethyl ester has been supported by various researchers. These reactions are shown in Figure 5.

Accumulating evidence supports the hypothesis that 5-aminolevulinic acid (ALA) formation in higher plants is formed primarily via a 5-carbon pathway, using as substrates glutamate or α-ketoglutarate (Beale, 1974) (Figure 5). ALA is an initial precursor for the biosynthesis of chlorophyll and iron is apparently involved in its formation.

Iron is promotive in the biosynthesis of light-induced ALA, as shown by an inhibition of its formation by specific ferrous-iron chelators, which ultimately form chlorophyll (Table 3) and by an increase in its formation in iron-stressed leaves by their preincubation in iron (Figure 7) (Miller et al., 1982). The requirement for iron in ALA formation was suggested by earlier research by Marsh (1963) involving 14C-citrate, succinate, α-ketoglutarate and ALA. In iron-deficient plants, only ALA from these substrates was incorporated normally into protoporphyrin and chlorophyll. The reduction of chlorophyll in iron-stressed plants may be a result then of ALA not being synthesized.

This synthesis may be related to the iron-containing intermediate in the electron transfer chain, ferredoxin. It may mediate the reduction and activation of enzyme(s) responsible for ALA synthesis (Miller et al., 1983). Under conditions of iron stress, ferredoxin would be expected to be limiting and thus not function in the activation of ALA synthesis. Table 4 indicates limiting ferredoxin in chlorotic tissue. Chloroplasts from chlorotic tissue had much less oxygen evolution capacity than those from control tissue. Addition of ferredoxin increased their oxygen evolution capacity, indicating that this iron-containing compound was limiting.
FIGURE 1. The process photosynthesis, during which light energy is converted into chemical energy intermediates (light reactions). These intermediates are necessary for the synthesis of sugar from water and the release of carbon dioxide (dark reactions).

FIGURE 2. Schematic representation of the light driven electron transport sequence. Electrons, ultimately derived from the splitting of water, are moved by two photochemical events through a series of compounds. During this passage of electrons, the light energy of the photochemical events is harvested into two chemical energy forms (ATP, NADPH).

FIGURE 5. Chlorophyll biosynthesis pathway depicting reactions where iron (Fe) has been demonstrated to be involved.

FIGURE 6. Effect of ferrous iron on the activation of coproporphyrinogenase isolated from tobacco. (Hsu and Miller, 1969).

FIGURE 7. Effects of iron on ALA (a chlorophyll precursor) formation in leaves from barley grown under low iron conditions. Preincubation treatments were: △, 10 ppm iron (EDDHA) in light; ○, water control in light; and □, 10 ppm iron (EDDHA) in dark. Light treatments were 4 hours at 110 μE m^-2 s^-1. (Taken from Miller, et al., 1982).

FIGURE 10. Effect of the addition of iron chelate on leaf pigments of chlorotic tobacco (Nicotiana tabacum). Iron stressed plants were supplied with 89 μM EDDHA and sampled subsequently at 24 hour intervals (Shetty, 1966).
Iron Chlorosis Correction Practices in Agriculture

Iron chlorosis commonly occurs in arid and semi-arid regions where the soils contain high natural concentrations of calcium carbonate (lime). Although iron is present in these soils, it is not readily available to plants. Lime soils have high pH values, causing the iron to be found as ferric oxide and other unavailable forms. This widespread condition necessitates effective and inexpensive corrective practices.

In an attempt to adapt the plant to the naturally occurring conditions, plant breeders are developing strains that are capable of efficiently extracting iron under stress conditions. Such plants are able to acidify the soil in contact with the root zone and thereby change the solubility of iron.

It has been known for many years that sensitivity to chlorosis may vary among species of the same genera and among varieties within a particular plant species. Wann (1941) found that Concord and many native American (labrusca) grapes are susceptible to chlorosis. He also found that many European (vinifera) grapes exhibited a high degree of resistance to chlorosis. Grafting American grapes on European root stocks provided a mechanism for control of chlorosis and enabled the development of grapes for chlorosis-endemic areas.

Species and varietal differences in the ability of plants to absorb and utilize iron have been shown in corn, tomatoes, oats, soybeans and other crop plants. Figure 8 compares Floradel, an iron-efficient tomato, to T-3820, an inefficient variety. Both varieties were subjected to the same iron-stress conditions. Floradel remained green and healthy, whereas the T-3820 variety was noticeably yellow.

Soil amendments have been employed to increase iron availability to plants. On a commercial scale, the prime considerations must be cost, duration and effectiveness. Direct acid application to the soil to lower the pH and subsequently increase iron solubility has proven effective. The major drawback to this process is that on a commercial agricultural scale the high buffering capacity of a calcareous soil requires large amounts of acid application, which can be quite expensive.

Iron salts and chelated iron complexes have been shown, to varying degrees, to improve iron availability. Iron-containing salts added to the soil encounter the same solubility problems as natural soil iron. Salts that are capable of modifying the soil pH are most effective (e.g., ferric ammonium sulfate). Additional benefit can be derived from soil manipulations that combine acid-iron salt treatments (Figure 9). The duration of effectiveness varies with the iron salt and existing soil conditions. Correction of iron-chlorosis by adding chelated iron to soil is widely recognized. An EDDHA addition has visible effects on leaf pigmentation (Figure 10).

Chelates derive their effectiveness by maintaining iron in the usable form despite high pH conditions. Unfortunately, the high cost of effective chelated iron complexes are as widely recognized as their corrective properties. This, coupled with the necessity of repeated application, makes their use less than appealing.

An alternate approach has been to bypass the soil solubility problem and supply iron directly to the aboveground parts of the plant. The most direct of these has been foliar application. Foliar sprays of iron compounds have increased the greening of the tissue (Figure 11). A limitation with this procedure is how well the iron penetrates from the leaf surface into the chloroplast. The inclusion of a wetting agent and/or a carrying compound in the spray, to increase surface dispersion of spray droplets and enhance absorption will minimize the green island spotting commonly observed with foliar iron application. This procedure requires repeated applications throughout the season since new growth requires iron supplementation. Urea and amino acid derivatives have been shown to effectively increase foliar nutrient uptake.

Corrective Practices in Arboriculture

In addition to the soil amendments and foliar sprays just discussed, lime-induced chlorosis in trees has been successfully treated by introducing iron compounds directly into the tree trunk. There are two currently accepted methods for accomplishing this iron introduction. The first and least expensive of the two requires the implantation of encapsulated iron into shallow borings of the trunk of the chlorotic tree. A number of forms of iron have been incorporated into such capsules, with ferric ammonium citrate being one of the most effective. The iron is slowly solubilized from the capsule and moves to the foliage in the transpirational stream. In oaks and white pine, beneficial effects have been noted for two or more years after the initial treatment. Capsules containing ferric ammonium citrate are commercially available under several trade names.

Pressure injection is another way to introduce iron into the transpiration stream. This technique entails embedding injection screws into the recently formed xylem directly below the bark, where water moves most rapidly via the transpiration stream.

In fruit trees, 1 percent to 2 percent ferric sulfate (and iron chelates to a lesser extent), have been successfully injected using pressures of 100-200 psi. In standard-sized fruit trees, positive responses have been observed for three or more years after a single injection. This procedure requires experience in inject rates, which varies with tree sizes, species, and time of year as phototoxic symptoms are commonly produced.

Future of Iron Chlorosis

The worldwide nature of the problem of lime-induced chlorosis has resulted in vast human efforts to reduce the agricultural impacts. Primarily this research has taken two compatible routes of inquiry, remedial and avoidance.

The ultimate solution to iron chlorosis is avoidance of the problem through the development and use of genetically resistant plants. Substantial progress in the breeding of economically important crops has occurred in recent years. The use of these resistant varieties would certainly do much to eliminate the iron-deficiency problem in several geographic areas. Unfortunately, resistant varieties are not yet developed for all crops or climatic regions. Further genetic improvement of a number of crops is limited by a lack of basic information on the role played by iron in the plants' biochemistry (e.g., chlorophyll biosynthesis, uptake and translocation of iron).

The only alternative in many geographic areas where iron-chlorosis-resistant crops are unsuitable or unavailable is to treat the symptoms through soil amendment and foliar sprays. In the long run, these corrective practices are expensive and time consuming. These characteristics of soil amendments in themselves may become yield limiting.
TABLE 1. Essential mineral nutrients of higher plants

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Absolute Requirement or Beneficial</th>
</tr>
</thead>
<tbody>
<tr>
<td>MACRO</td>
<td>MICRO</td>
</tr>
<tr>
<td>Oxygen</td>
<td>Iron</td>
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<td>Carbon</td>
<td>Manganese</td>
</tr>
<tr>
<td>Hydrogen</td>
<td>Copper</td>
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<tr>
<td>Calcium</td>
<td>Zinc</td>
</tr>
<tr>
<td>Magnesium</td>
<td>Molybdenum</td>
</tr>
<tr>
<td>Sulfur</td>
<td>Chlorine</td>
</tr>
<tr>
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<td>Boron</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>Sodium</td>
</tr>
<tr>
<td>Potassium</td>
<td>Cobalt</td>
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TABLE 2. Chlorophyll and iron contents from chloroplasts of normal and chlorotic tobacco.

<table>
<thead>
<tr>
<th></th>
<th>Chlorotic Tissue</th>
<th>Intermediate</th>
<th>Control</th>
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</thead>
<tbody>
<tr>
<td>Chl/g fr wt n moles</td>
<td>a</td>
<td>b</td>
<td>a/b</td>
</tr>
<tr>
<td>Active Iron n moles/g fr wt</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chlorotic Tissue</td>
<td>14</td>
<td>5</td>
<td>2.82</td>
</tr>
<tr>
<td>Intermediate</td>
<td>22</td>
<td>8</td>
<td>2.75</td>
</tr>
<tr>
<td>Control</td>
<td>124</td>
<td>45</td>
<td>2.77</td>
</tr>
</tbody>
</table>

Chloroplasts were extracted from tobacco plant at indicated stages of iron-induced chlorosis. Chlorophyll contents were determined by dimethyl formamide extraction (Moran, 1982). The iron concentration of isolated plastids were extracted by 1 N HCl for 6 hrs. and determined by atomic absorption spectroscopy.

ACKNOWLEDGMENTS

The authors would like to express thanks to Kathleen Warnick and John Manwaring for continuous input during the preparation of this manuscript. We would like to also thank Drs. Richard Mueller, David James, and John Manwaring for contributing photographs to complete the publication and Vivian Johnson for typing and editorial assistance.

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Gene W. Miller is a professor and Head of the Department of Biology. He received his BS and MS degree in Soil Chemistry from USU and PhD in Plant Biochemistry from North Carolina State University. Much of his work at USU has been on the role of iron in porphyrin and chlorophyll biosynthesis. His other area of emphasis has been the effects of HF air pollution on vegetation.

James C. Pushnik is a postdoctoral fellow working on the role of iron in the establishment of normal chloroplast photosynthetic activities. He received his BA in Biology and MS in biology/genetics from Humboldt State University and PhD in biology/plant biochemistry from USU.

TABLE 3. Effect of metal chelators on light and dark-induced ALA formation in etiolated barley.

<table>
<thead>
<tr>
<th>Chelator</th>
<th>Concentration mM</th>
<th>n moles ALA Formed /g Fresh Weight</th>
<th>% Inhibition</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Light</td>
<td>Dark</td>
<td>Light</td>
</tr>
<tr>
<td>O-Phenanthroline</td>
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<td></td>
<td>500</td>
</tr>
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<td></td>
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<td></td>
<td>418</td>
</tr>
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</tr>
<tr>
<td>α, α'-Dipyridyl</td>
<td>0</td>
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<td></td>
<td>1.0</td>
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</tr>
<tr>
<td></td>
<td>5.0</td>
<td></td>
<td>50</td>
</tr>
<tr>
<td>BPDS</td>
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<td>500</td>
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<tr>
<td>(Batho-phenanthroline-dimethyl-sulfonate)</td>
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<td>305</td>
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<tr>
<td></td>
<td>1.0</td>
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<td>255</td>
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<td></td>
<td>5.0</td>
<td></td>
<td>27</td>
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</table>

Leaves from etiolated 8-day-old barley plants were infiltrated with the chelator indicated for 18 hours in the dark. They were treated with 75 mM LA and light where indicated at 110 E m⁻²s⁻¹ for 4 hours. Leaves were analyzed for ALA as indicated in Materials and Methods. (Taken from Miller et al., 1982.)

REFERENCES


TABLE 4. Measure of Ferredoxin as related to O₂ evolution in chloroplasts.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>No Ferredoxin Addition</th>
<th>Ferredoxin Added 2.6 n moles/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorotic</td>
<td>1.0</td>
<td>1.5</td>
</tr>
<tr>
<td>Intermediate</td>
<td>2.3</td>
<td>5.6</td>
</tr>
<tr>
<td>Control</td>
<td>8.9</td>
<td>9.2</td>
</tr>
</tbody>
</table>

Biologic oxygen evolution of isolated plastids was determined by Clark oxygen electrode according to the procedures of Haslett and Commack (1973). Ferredoxin additions were made using purified commercial ferredoxin (Sigma St. Louis).
Agricultural Implement Industry in Utah

This study was completed in 1981 and will be up-dated in 1985. Analysis of the findings of the study indicated a 21.61 percent increase in full-time jobs in this industry in the two years prior to the study (1979-81). It was predicted by those surveyed that in two years, the number of full-time employees would increase by 20.35 percent. The most important technical competency was found to be the ability to perform preventive maintenance on agricultural equipment systems (e.g., electrical, fuel intake, exhaust, lubrication, cooling, hydraulics and brake). The least important competency as defined in this study was the ability to use surveying equipment and do surveying. Sixty-six skills and categories of knowledge were prioritized for use by curriculum planners.

Personal qualities such as honesty, sincerity, willingness to work and learn, positive attitude, following directions, and neat appearance, received the highest rankings as desirable interpersonal skills.

Knowing about advertising and promoting the sale of equipment received the lowest ranking of the interpersonal skills, but, nevertheless, this skill was considered important by those surveyed.

The results of this study could be used to help: vocational instructors and others update their curriculum in agricultural mechanics, in program articulation, or anyone interested in pursuing a career in agricultural mechanics.

1. There are 398 full-time and 53 part-time employees employed by 49 of 50 farm implement dealers in Utah.

2. Numbers of employees grew between 1979 and 1981 by 20.35 percent for full-time employees and 15.87 percent for part-time employees. This is in addition to the 3 to 5 percent needed to fill jobs made available by retirement, employees leaving the industry, etc.

3. Fifty-seven percent of the farm implement dealers in Utah were aware of USU’s Agricultural Equipment/Machinery Technology Program to prepare employees for this industry.

4. Ninety-four percent of the respondents stated a preference for vocationally trained workers, but only 11.8 percent of the last 51 who had been hired belonged to this group.

5. The average, standard, shop-labor rate for the state of Utah is $15.47 per hour.

6. Farm Implement Dealerships in Utah have existed an average of 22.72 years.

7. The average tenure of industry employees in Utah is 8.25 years.

8. According to dealership management, individuals primarily quit jobs in the industry because they can make more money elsewhere.

Ornamental Horticulture

In 1980, Utah’s 576 ornamental horticulture enterprises were categorized into seven areas of specialization: Arborists (4.5 percent), Maintenance of Landscapes (10.8 percent), Landscape Construction (18.6 percent), Golf Courses (13.5 percent), Greenhouse Production (5.4 percent), Florist-Plant Shops (30.9 percent), and Garden Center Nurseries (16.3 percent). One hundred and sixteen such enterprises were surveyed.

Employee competencies perceived to be the most important by industry managers were in: human relations and communications, pest identification and control, record keeping, turf care, marketing, and safety and first aid.
In addition, highly rated individual competencies categorized by major areas of specialization included:

1. Arborists—mix and handle chemicals safely, turf establishment and maintenance, removal of trees and shrubs, and pruning techniques.
2. Maintenance of landscape—mix and handle chemicals safely, apply fertilizer materials, and operate and maintain spray equipment.
3. Landscape construction—plant and transplant, turf establishment and maintenance, and sell products and services.
4. Golf courses—apply fertilizer materials, maintain golf greens and turf, and apply knowledge of irrigation and watering methods.
5. Greenhouse production—apply knowledge of irrigation and watering methods, sell products and services, and operate a cash register and handle money.
6. Florist—design and construct floral pieces, dish gardens, and terrariums; operate a cash register and handle money, and design displays.
7. Garden center nurseries—operate a cash register and handle money, sell products and services, and select plant materials according to growth habits and uses.

The three individual competencies considered most important for all major areas of specialization were from the skill grouping titled "Human Relations and Communications." These were: 1) maintain good relations with customers, employers, and fellow employees; 2) recognize what needs to be done; and 3) stimulate others to work effectively. Pest identification and control, record keeping, and safety techniques were other highly rated competencies.

The number of employees anticipated in the future is growing at a 16 percent yearly rate. The greatest expansion of full-time employees is anticipated in the areas of landscape construction (63 percent) arboriculture (35 percent) and greenhouse production (32 percent). Members of owners' families compose 17 percent of the industry's employees.

Feed, Seed and Grain Industry

In 1979, manpower demands in the Utah feed, grain, and seed industry and in the Idaho seed industry were increasing. In Utah, 493 more persons were employed from 1975 to 1977, with 699 persons expected to be employed by 1979. Employees in Idaho increased by 133 between 1975 and 1977, with a projection of 446 more persons to be employed by 1979. In the Feed, Seed, Grain and Feeding industry, individuals having the following job titles needed particular training as determined by those surveyed (listed in order as needing the most training): salesperson, department managers, clerks, fieldmen, branch managers, warehousemen, truck drivers, general managers, mill men, secretaries and general laborers. Several other positions were listed, but by fewer respondents than the above titles.

A projected increase of 699 additional persons employed by 1979 should justify a more concerted effort by Utah vocational agricultural educators to teach the competencies needed in this industry. However, the economic downturn prevented realization of these projections.

Production Agriculture

One-third of all respondents in this study terminated their formal education in 1980 with a high school diploma. The vocational agricultural instructor thus provides the last formal training that many farmers receive. Over 40 percent of surveyed farmers indicated training was desired in their employees in 47 of 114 competency areas. Animal health, maintaining equipment and vehicles, soil preparation, planting, irrigation and harvesting as well as observing legal practices and following general safety precautions were the four competency areas considered most important by the farmers in production agriculture. These data provide a sound basis for implementing education short courses to be taught by extension specialists. They can also give direction to existing young-farmer training programs.

The findings of this study also support these additional conclusions:

1. It is difficult to differentiate between large and small farmers who have general-production operations.
2. Many areas of production agriculture have overlapping areas of needed skills and competencies.

3. There is a commonality of competencies needed by all who enter the production agriculture industry.
4. The average number of full-time family employees fluctuated by 9.1 percent (up or down) over a 5-year period.
5. The number of part-time family employees tended to increase approximately 5 percent over a 5-year period.
6. The number of full-time, non-family employees is projected to increase by 10 percent over a 5-year period.
7. The number of non-family employees working part-time is projected to undergo a marked 18 percent decrease during the projected 5-year period.

The information collected has been used to refine curriculum and to plan program emphasis at Utah State University. State curriculum guides have been developed based upon these industry surveys, and University teacher preparation has been evaluated to measure the relevancy of its curriculum to the industry. This important systematic evaluation is made possible by an up-dating of the data base every four years.

To facilitate local program planning, additional local surveys must be completed to refine the state-wide surveys reported in this article. Previous research conducted by the department indicates that only a continuous effort can keep up with rapidly changing agricultural technology.

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1981—Jim Jensen, USU Thesis Study—"A Study to Determine desirable Employee Competencies and Man-Power Needs for the Utah Farm Implement Industry."

ABOUT THE AUTHORS

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FIGURE 1. Scrapie Suffolk ram with wool rubbed from shoulder, flank and legs.

FIGURE 2. Scrapie Suffolk ewe with bilateral loss of wool from rump and legs.

FIGURE 3. Monitoring vital body signs of anesthetized ewe during surgery.

FIGURE 4. Glass cannula is inserted through wall of uterus and will provide exit port for flushing out embryos.

FIGURE 5. Cavity of uterus is flushed with tissue culture medium and drained into glass collecting dish.

FIGURE 6. Embryos are located in a uterine flush by observation through a microscope.

FIGURE 7. Five-six day old embryos as observed through the microscope.

FIGURE 8. Embryo that was collected from scrapie-exposed donor is placed into the uterus of nonexposed recipient.

FIGURE 9. Embryo transfer Cheviot sheep were transported as embryos from Mission, Texas, and born at Dugway, Utah.
SCRAPIE AND EMBRYO TRANSFER

W. C. FOOTE, W. W. CLARK,
J. W. CALL, T. D. BUNCH, and J. PITCHER
The term "scrapie" is far from being a common household term. In fact, it is even unknown to most people in the livestock industry, including many who are concerned with sheep, the animals to whom it applies directly. Scrapie, a virus-like infection of the central nervous system, is the name of a formidable disease that affects sheep and less commonly goats. There is no method for early detection or treatment once the disease is demonstrated clinically, and no animals survive scrapie.

Scrapie has been observed in most countries of the world with only two, Australia and New Zealand, being recognized as scrapie free. It is most commonly known among purebred sheep producers and particularly those producing Suffolks, Cheviots or Hampshire breeds, because the occurrence is much higher among animals of these breeds than other breeds. In fact, except under experimental conditions, the disease occurs predominantly in Suffolks and to a lesser degree Cheviots and Hampshires in the United States. Because of the close relationship of the occurrence of scrapie to these breeds, it was considered by some for many years to be of genetic origin—that it was inherited. This was later showed to not be the case and, although the causative organism has not been definitively identified, it is now accepted as being due to an infectious organism with the Suffolk, Cheviot, and Hampshire and goats showing a definite susceptibility to the disease under opportune conditions of exposure.

Incidence in North America

Scrapie has been recognized as a serious threat to the sheep industry in the United States and Canada for about 40 years. The disease apparently entered the United States from Canada. A sustained and largely successful effort has been made by the Veterinary Services of the Animal and Plant Health Inspection Service (APHIS) of the USDA to control scrapie. The goal of APHIS is to eradicate the disease. During the 35-year period from 1947 through Sep-

October 1983, 278 flocks and 391 animals were identified as infected. Of these flocks, 254 were Suffolk, 11 were Cheviot, 8 were Hampshire, 2 were Montadale, and 3 were of mixed breeding. Scrapie has also been diagnosed in the Targhee and Rambouillet breeds of sheep and in Angora, Nubian and Toggenburg breeds of goats which were experimentally exposed in the field to the natural disease. Other breeds have not been experimentally checked. Most of the research on scrapie in the U.S. has been carried out at the USDA, APHIS, Field Trials facilities at Mission, Texas.

Scrapie was diagnosed in one flock of sheep in Northern Utah in 1957. Scrapie has not been reported in Nevada, Arizona or New Mexico.

The name "scrapie" comes from one of its early clinical symptoms. Pruritis (to itch), most apparent around the tailhead of afflicted animals, causes them to scrape against solid objects causing loss of wool and abrasions to the skin. Animals may also scratch or dig at their head or sides with their hind legs.

The biology of scrapie is described in terms of clinical and pathohistological symptoms, some methods of transmission and incidence under experimental conditions. The first indication of this insidious, degenerative disease of the central nervous system includes a change in temperament; the animal becomes apprehensive and easily excited. As the disease progresses, the early nervous and scraping symptoms intensify and the animal develops a characteristic, uncoordinated gait. Muscle tremors involving the head may also develop. Because of the nervous system disorders, they eventually are unable to stand or to chew or swallow feed. Death inevitably occurs within a few weeks or months after the first clinical signs are observed.

Control Potentials

For several reasons scrapie is an extremely difficult disease to effectively control.

First, the causative agent has not been specifically identified. It is classified as a slow virus, so called because of the very long period from the time of exposure to when the disease can be clinically observed which may be 60 months or even longer. Actually, referring to the scrapie agent as a virus is a misnomer because it is now generally accepted as being subviral (T. O. Diener, 1983). One group of subviral agents, all of which are pathogens of higher plants, are known as viroids. The scrapie agent, which has been studied for more than 30 years, appears to represent a second class of subviral pathogens, smaller than the smallest viroids with a molecular weight of 50,000 or less. This class of subviral agents has been referred to as prions (Diener, 1972; Gajdusek, 1977; Prusiner, 1982). This agent is highly resistant to heating, incapable of eliciting an immune response, and is resistant to ionizing and ultraviolet radiations. Physical and chemical properties of the agent are still largely unknown.

Approximately 5 percent of clean (non-exposed) animals first exposed after weaning at 3-6 months of age develop scrapie, whereas 15-22 percent develop the disease if exposed from the time of birth in an infected flock. This situation makes it impossible to know the status of the disease in a population that may have been exposed. Most breeding animals would demonstrate the disease during their period of greatest production. Others may have died before the disease is demonstrated clinically, yet have already exposed their own offspring or other flock members.

Other diseases also identified as being caused by slow viruses are visna-maedi in sheep, Transmissible Mink Encephalopathy in mink, and Creutz-
Scrapie is a very costly disease. The costs to the Federal government for surveillance and for indemnity in destruction of exposed animals, are high. Also, the sheep industry and particularly the purebred producers must absorb, directly or indirectly, the costs resulting from loss of breeding animals and the restriction of international sales that could otherwise result from exports.

An evaluation of what is known about scrapie suggests that a logical method of control is to develop a procedure to interrupt transmission. One logical point would be during vertical transmission, specifically the embryo. If the disease is not transmitted to the embryo at the time of fertilization, the embryo might be transferred from an infected or exposed dam to a known scrapie-free foster dam. This would interrupt the transmission and provide a procedure whereby scrapie-free offspring could be obtained from scrapie exposed of infected flocks.

**Embryo Transfer as a Tool**

In 1979 APHIS invited the International Sheep and Goat Institute, Utah State University, to conduct cooperative studies to determine the route and time of any vertical transmission of scrapie using embryo transfer. To accomplish this, flocks of donor and recipient ewes were created in two locations in the United States. One flock of ewes was established at the Scrapie Field Trial Facilities at Mission, Texas. These were artificially exposed to scrapie. One flock of donor and one flock of recipient ewes were established at the Dugway Proving Grounds, Dugway, Utah. These animals were obtained from known scrapie-free flocks and were not exposed to scrapie. Animals from the Suffolk and Cheviot breeds of sheep are being used in the study. This site was chosen because the experimental sheep could be isolated from any other sheep in the area and thus guarantee protection from any possible spread of the disease. To determine the route and time of suspected vertical transmission, embryos from scrapie-exposed ewes in Texas are being transferred to scrapie-free ewes in Utah. If the animals developing from these embryos do not develop scrapie, it will be assumed that the disease is not transferred from parent to offspring at fertilization. This would provide a method of interrupting the transmission chain.

Embryos from scrapie-free donor ewes in Utah are being transferred to scrapie-exposed ewes in Texas. If these offspring do not develop scrapie it will be assumed that the organism is not transmitted to the embryo during gestation following fertilization. If this were the case, scrapie-free fetuses could be removed from their scrapie-exposed dams by Caesarean section, providing an alternative to embryo transfer. If the disease is transmitted to the offspring during gestation but not at the time of fertilization, embryo transfer would remain a viable alternative.

Appropriate control offspring are also being produced from the scrapie-exposed donor and recipient animals in Utah and from the scrapie-free donor and recipient ewes in Texas. This is necessary to determine the efficacy of transmission of scrapie under the experimental procedures and also the effectiveness of management protocol to protect against non-vertical transmission.

Initially, the ewes in Texas were experimentally exposed to scrapie through oral and subcutaneous injection of an inocula prepared from brain or brain-plus-spleen homogenate from animals that had died from scrapie. After the estimated "incubation" period under this experimental procedure of approximately eight months in Cheviots and two years in Suffolk, the embryo transfer work was begun. This was accomplished by synchronizing the reproductive cycles (estrus and ovulation) between donor ewes in Utah and recipient ewes in Texas, and also donor ewes in Texas and recipient ewes in Utah. The synchronization and also the induction of super ovulation in the donor ewes was achieved with hormones.

The actual embryo transfer is accomplished during a non-stop period of 30-36 hours during which embryos are recovered from donor ewes in Utah, placed in a portable incubator, flown to Texas and transferred to recipient ewes. After a period of about one day for rest and required preparation, another 30-36
FIGURE 10. Diagramatic illustration of experimental design to determine route and time of vertical transmission of scrapie. (The symbol for sheep exposed to scrapie is +; sheep not exposed −).

10A. Test for transmission of scrapie to embryo at time of fertilization.

SCRAPIE-EXPOSED PARENTS (Donors +)

SCRAPIE-FREE FOSTER MOTHER (Recipient −)

OFFSPRING

UTERUS (−)

(+ ) if embryo is infected

EMBRYO

10B. Test for transmission of scrapie to embryo of fetus via the uterus during pregnancy after embryo transfer. Birth is by caesarean section.

SCRAPIE-FREE PARENTS (Donors −)

SCRAPIE-EXPOSED FOSTER MOTHER (Recipient +)

OFFSPRING

UTERUS (+)

(+ ) if scrapie exposure is from the uterus

EMBRYO
The hour work program is undertaken during which embryos are recovered from donor ewes in Texas, transported to Utah and transferred to recipient ewes. The last of these transfers was conducted in October 1983. These embryo transfers involve a team of five persons that travel between the two locations plus additional persons at each location. These transfers may have set a record for the number of sheep (in embryo form) transported in a passenger airplane.

**Potential Gains**

The use of embryo transfer in our studies with scrapie has provided a model and encouraged this approach in studies of other diseases.

This is a long-term project. Four to five years will be spent preparing the donor and recipient ewes and obtaining the embryos. Another 4 to 5 years will be invested in observing the offspring for clinical symptoms of scrapie to determine conditions under which the disease is transmitted.

The results will yield information on if and how scrapie is transmitted vertically and possibly a method to effectively interrupt its transmission. Scrapie-free sheep might then be obtained from scrapie-exposed or infected flocks. The procedure might also provide a way to more adequately control and eradicate the disease in the United States while facilitating the import and export of sheep to further strengthen the United States sheep industry.

A total of approximately 360 donor and recipient ewes and 35 rams were obtained for the study. A significant proportion of these that were exposed to scrapie could not be used in the production of offspring through embryo transfer because they developed scrapie before the embryo recoveries or subsequent pregnancies following transfer could be completed.

A major part of the study consists of a waiting game since as many as five years (or even more) may be required for the clinical demonstration of scrapie. For this reason, data, other than the numbers of offspring produced, are scarce at this point in the study. A diagramatic presentation of the experimental design is shown in Figure 1. Actually 478 embryos have been transferred in the study resulting in 142 live offspring. These live offspring represents 30 percent of the embryos transferred. This survivability rate compares favorably with other reports where embryos have been cultured for comparable periods. The incidence of embryo loss does appear to be increased, however, by some factors related to transport. The numbers of offspring produced to date for each aspect of the study are shown in Table 1.

Some events are already being observed, however, which provide at least a basis for answers to the questions asked in this research. The first case of scrapie in the project has been clinically diagnosed in a 36-month old Cheviot offspring from scrapie-exposed donor and recipient animals. This demonstrates that scrapie is being transmitted under conditions of the experiment. The primary interest now in our scrapie study is directed to the offspring resulting from the reciprocal embryo transfers between scrapie-free and scrapie-exposed donor and recipients.

**TABLE 1. Number of offspring produced by embryo transfer for each comparison in the study.**

<table>
<thead>
<tr>
<th>Donor</th>
<th>Recipient</th>
<th>Test for scrapie Transmission to embryo/fetus</th>
<th>Number of offspring</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scrapie-exposed</td>
<td>Scrapie-free</td>
<td>Transmission via embryo at fertilization</td>
<td>80</td>
</tr>
<tr>
<td>Scrapie-free</td>
<td>Scrapie-exposed</td>
<td>Transmission via uterus after fertilization.</td>
<td>32</td>
</tr>
<tr>
<td>Scrapie-exposed¹</td>
<td>Scrapie-exposed</td>
<td>Determine occurrence of vertical or lateral transmission (positive control).</td>
<td>39</td>
</tr>
<tr>
<td>Scrapie-free</td>
<td>Scrapie-free</td>
<td>Determine freedom from presence of agent in unexposed animals (negative control).</td>
<td>15</td>
</tr>
</tbody>
</table>

¹One lamb born in this experimental group was clinically diagnosed as having scrapie at 30 months of age.

**LITERATURE CITED**


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*WINTER 1983 111*
The progressive Utah farmer or rancher will generally be at least 50 years old, have five or more children, hold a college degree and have two or more full-time employees. The average Utah agricultural employee loves agriculture, doesn’t want to be tied down to a desk and believes agriculture is important to the nation.

Those profiles are accurate according to a recent survey of 109 Utah farmers and ranchers completed by the Agricultural Education Department at Utah State University. The study was part of an effort to determine what kinds of labor relations exist between farmers and their hired help, and how farm employers keep their workers happy. Names of farmers and ranchers contacted in the study were obtained from county agents and local vocational agricultural teachers. Also, a random sample of the employees of the 109 farmers and ranchers were asked about why they stayed with their current employer.

**Employer Characteristics**

According to the study, ages of Utah farmers and ranchers varied from less than 25 to over 70. Over 80 percent of the respondents, however, were between the ages of 30 and 60, with the average being 47.

Only one of the 109 participants was not married. Of the married respondents, only one reported having fewer than 2 children. Over 50 percent of the responding farmers/ranchers had either four or five children and 30 percent had more than 5 children. About 70 percent of the respondents indicated that they had at least one child currently working on the farm or ranch. Family labor is apparently important to the success of the operation on Utah farms and ranches.

Nearly all (97 percent) of the responding farmers/ranchers had at least a high school education. Four of ten reported having a college degree in fields ranging from art to optometry. Of those having college degrees, the most common were in animal science, education or business.

Over 80 percent of those surveyed were in the cattle business—range operations, feedlots or dairies. Of these producers, nearly two-thirds also raised small grains or forage crops. A wide variety of other agricultural enterprises were also reported, ranging from trout and mink to turf care and vegetable production.

More than 90 percent of the responding producers had operations encompassing at least 100 acres. Nearly 30 percent were involved in production enterprises of over 10,000 acres. Most of the larger operations were range cattle or sheep and many of the smaller ones were poultry or dairy.

The responding Utah farmers and ranchers had from 0 to 32 full-time employees, depending on their type of operation. The average was about two full-time employees per ranch or farm when three large organizations (25, 25, and 32 employees) were deleted from the computation. Most full-time employees were farm or ranch foremen, truck drivers, or milkers.

When asked to report the amount of education required of an employee, most employers indicated that a high school education was adequate. Some said that no education was necessary and a few indicated that they would prefer a college degree. Although little formal education was required for most positions, many of the respondents said they demanded appropriate and extensive work experience.

When asked if they perceived a need for more qualified and better help, 70 percent of the employers answered yes. However, 70 percent also stated they were currently able to find and keep good help, provided they paid enough and treated their employees fairly.

Most Utah producers expect to keep their farms or ranches in the family; only 5 percent of the respondents indi-
cated that they planned to sell their operation at anytime. (When employees were asked if they had been offered a chance to buy into the existing operation, 33 percent said that they had that opportunity.)

The Employees' View

Of the employees who had taken Vo-Ag in high school, 81 percent said that the educational experience they received had prepared them for their current position. Of those, several stated that high school Vo-Ag programs should provide more hands-on experience in the classroom and teach more decision-making skills in farm management.

Tenure among the employees surveyed varied from less than 6 months to over three years. (Nearly 60 percent of the employer respondents indicated that their hired help stayed with them for at least three years. Only about 18 percent reported having hired help that stayed less than one year.) According to Herzberg, Mausner and Synderman, employees gained satisfaction from their jobs through personal achievement, recognition and work itself. Also, according to their study, employees derived dissatisfaction from their jobs through company policy, technical supervision and supervision of human relations. On a scale of 1-7 (1 representing completely satisfied and 7 representing completely dissatisfied), most agricultural employees that were surveyed said they were well satisfied with their current job, with the average being 1.75 on the scale. In responses to the question, "If you were paid enough money to live as comfortably as you would like, would you continue with your current job?", 92 percent of the employees answered yes.

Wages offered to full-time farm or ranch help ranged from $500 to $2,200 per month. This compares to a range of $400 to $700 per month among Montana farm and ranch workers in 1981. The average wage earned by a full-time Utah farm and ranch worker was $1,020 per month. When asked whether they believed their current salary was equivalent to those paid for other similar jobs within the same area, 67 percent of the employees stated yes, 17 percent said no, and 16 percent replied they didn't know. The majority of the employees surveyed also indicated that they were satisfied with the amount of pay they received for the number of hours they put into their jobs. Many of the employees also provided housing valued from $50 to $400 per month as well as a number of other benefits. Of the employees interviewed, 80 percent said the housing provided by their employer was adequate for their family's needs.

Eight out of each ten Utah farmers and ranchers in the study paid into the Social Security program for their employees. Over 50 percent provided liability insurance and about the same number offered health insurance to their employees. Other benefits commonly listed included workmen's compensation, housing (out of the 109 respondents, 31 reported furnished and 20 reported unfurnished), opportunities to raise livestock or grow crops (on a limited basis), garden space, fuel allowance, cash bonuses and provision of meat and milk. Employees cited health insurance, paid vacations, access to farm vehicles for personal use and being able to run personal livestock on the operation as the most important type of benefits.

Only a slim 8 percent of our employer respondents involved their full-time help in a profit-sharing plan. The profit-sharing programs included allowing employees to operate a part of the farm for personal gain using the farm's equipment, and/or to buy small calves and feed them through the winter with the farm providing feed and housing. In some cases, employees received an additional percentage of their current salary if the farm made a profit for that particular year.

All the employees surveyed said they received respect as individuals on and off the job from their employers. Some of the major reasons mentioned for continuing to work for their current employer were: they could take time off when they wanted to, they were treated like family, they weren't nagged 8 hours a day, and they were not tied down to doing the same thing everyday. When the employees were asked if they received compliments from their employer after they finished a job, 89 percent answered yes. One employer said, "My employer and myself have the best working relationship possible, we go bowling together, our families go on picnics and this is why I am still here." Another employee stated, "My employer tells me what he wants done and leaves me with that job to be completed, he doesn't come around and bother me every hour. What kind of other job can you find where your employer isn't on your back everytime you turn around?"

If Utah's food producers are to survive the continuing economic problems of declining exports and cost/price squeeze, they must employ the best quality help possible. Only one in ten is providing profit-sharing incentives—yet seven of ten say they need better help. It is true that Utah farm workers are paid more than those in at least one other western state. Nevertheless, producers might do well to consider offering profit sharing and other kinds of employee benefits as incentives to attract the competent people they need.

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When dealing with complex biological systems involving similar components, scientific progress is often limited by the methodology available for detection and measurement. A basic characteristic of the antibody system of animals is its high specificity. This specificity has been utilized in the past in immunofluorescence and radioimmunoassay and most recently in enzyme-linked immunosorbent assay (ELISA), which has significantly improved detection and measurement potentials for the biological scientist.

Some of the materials that have been measured using ELISA includes: disease causing bacteria, parasites and viruses, hormones, toxins, chemicals, and antibodies. ELISA overcomes the disadvantages of immunofluorescence and radioimmunoassay by using reagents that are relatively easy to prepare, highly stable and can be handled with a high degree of safety. Also, a person can be quite easily trained to run the ELISA test compared with the demands of radioimmunoassay. ELISA’s colorimetric endpoint readings are objective, and the system can be automated for handling large numbers of samples, with the equipment required being much less expensive than that for other procedures.

How Does ELISA Work

In ELISA, the material to be evaluated is bound in various immunological layers to a solid plastic surface or phase. An enzyme is then immunologically linked in proportion to the concentration of the material being tested. A specific, enzyme-mediated, color change reaction is then initiated and colorometrically read. Careful control of pH is necessary throughout the entire procedure for proper binding to occur. The real shortcoming of ELISA is reagent availability. At the present time, few reagents are being commercially prepared; however, this number is increasing monthly. Most laboratories are making their own antibodies for use in the specific ELISA tests they are using in research. Therefore, interlaboratory standardization of reagents is much needed.

Figures 1 and 2 show the steps in the two basic types of ELISA. Many modifications of these procedures are now in use. In all of the ELISA procedures, selection of an appropriate solid phase becomes step one. Some forms of the solid phase include a well with a flat, “V” or “U” bottom or a round sphere or bead in a glass test tube. The material of the solid phase includes such possibilities as polystyrene, polyplyene or polyvinyl. A number of proprietary plastics are also marketed for solid phase use.

The sandwich method (Figure 1) is used to detect exogenous agents (antigens) not normally found in the material being tested such as enteropathic virus in fecal samples. An antibody specific for the antigen is bound to the solid phase, the sample containing the antigen of interest is added to the well and, after a reaction period that allows the antigen to bind to the antibody adhered to the solid phase, the wells are washed. A specific antibody-enzyme conjugate is added to the wells and allowed to react, “sandwich,” with the antigen, if it is still present. After a second wash, in which all unadhered antibody-enzyme conjugate is removed, enzyme reagent is added to the well and a color reaction occurs. After a short period of time, the reaction is quenched and the color intensity is measured using a spectrophotometer.

The indirect ELISA technique is usually used to detect immunoglobulins (antibodies) in serum against a specific antigen (Figure 2). This test begins with the binding of specific antigen for the antibody of interest to the solid phase.

The serum sample containing antibody is then added and an antigen/antibody complex forms. After a wash, species-specific, enzyme-antiglobulin conjugate is added. It adheres to the bound antibody. After a second wash, enzyme reagent is added, allowed to react, quenched and eventually a photometric measurement is made. Many companies are now preparing species-specific enzyme-antiglobulins. Some of the commonly used enzyme systems and enzyme reagents are listed in Table 1.

Uses of ELISA

Nakane and Pierce (Department of Pathology, University of Michigan), in 1967, were the first to report in the literature on the use of enzyme-labelled antibodies for the localization of antigens. Their paper was followed in 1971 by the immunoglobulins paper of Engvall and Perlmann (Department of Immunology, University of Stockholm), which introduced the term ELISA. The paper by Van Weemen and Schurrs (Research Laboratories N.V. Organan, OSS, the Netherlands) on immunoassay using antigen-antibody conjugates was also published in 1971.

In less than two decades, ELISA has been used in a wide variety of applications and, with the advent of monoclonal antibodies, will undoubtedly be applied even more widely. Table 2 tabulates ELISA applications in the area of animal and human disease diagnosis. Table 3 lists some of the other varied applications in which ELISA has been used.

ELISA in Calf Diarrhea Diagnosis

In the mountain west, including Utah, there appears to be several causes of calfhood diarrhea in both dairy and beef herds. These diseases often appear to be very similar clinically, yet prevention requires that a rapid, specific diagnosis
FIGURES 1 and 2. The conical shaped wells are used for testing samples. A particular shape has been assigned to each material to demonstrate its binding and building capacity. In testing for the presence of specific antibodies or antigens these materials are sandwiched in various layers and washed with buffered saline solution between each new addition. Non-binding materials are washed away. The substrate is added last and if the sample undergoes a color change, the antigen or antibody is present.

FIGURE 1. SANDWICH ELISA

POSSITIVE REACTION

NEGATIVE REACTION

FIGURE 2. INDIRECT ELISA

POSSITIVE REACTION

NEGATIVE REACTION


PHOTO 2. Reading the Rotazyme™ test by comparing color in tube with colors on card.

PHOTO 3. ELISA test showing various dilutions.

PHOTO 4. Reading the K-99 E. coli test using the Gilford which is linked to the Apple Computer.

PHOTO 5. Reading an ELISA plate.
TABLE 1. Common enzymes and substrates used in ELISA

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Substrate</th>
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<tr>
<td>Alkaline Phosphatase</td>
<td>p-Nitrophenyl phosphate</td>
</tr>
<tr>
<td>Peroxidase (Horse Radish)</td>
<td>0-Dianisidine (3,3'-Dimethoxybenzene)</td>
</tr>
<tr>
<td></td>
<td>2,2'-Azino-di-(3-ethyl)-Benzothiazoline-6-Sulphonic acid</td>
</tr>
<tr>
<td></td>
<td>4-Aminooantipyrine (4-Aminophenazonate)</td>
</tr>
<tr>
<td></td>
<td>0-Phenylenediamine</td>
</tr>
<tr>
<td></td>
<td>5-Aminosalicylic acid</td>
</tr>
</tbody>
</table>

be made so appropriate measures can be taken. Table 4 tabulates some general information on the common look-a-like diarrhea diseases in calves. It is not uncommon for an animal to be suffering from a combination of diarrheal types. An animal with a single diarrheal type may recover quite rapidly, but, when combination infections are found, the animal is much sicker and often dies. Stress, in the form of cold, heat, dampness, wind, crowding, castration, dehorning, improper feeding, etc., often contributes to the onset and severity of an outbreak. A rapid ELISA method that could identify the presence of diarrhea-causing bacteria, viruses or parasites would be of great value in the diagnostic processes. Furthermore, if a blood sample could be taken from an animal and run through a battery of ELISA tests to determine quantitatively its immunological profile for those agents of calf diarrhea, therapy to correct deficiencies could be initiated.

In the Utah State University Veterinary Diagnostic Laboratory, we have completed three projects that will help to identify the specific causes of calf diarrhea. The first project was to develop an ELISA test for differentiation of enteropathic E. coli which carry the K-99 antigen from the normal E. coli of the intestine in fecal samples. The second project was to use the K-99 ELISA test and a commercially available rotavirus ELISA (Rotazyme™ Abbott Laboratories, Chicago, IL) to test cases of calf diarrhea as they came into the diagnostic laboratory. The third project was to utilize the K-99 ELISA tests as well as the Rotazyme™ test in an epidemiology study of 10 dairies in northern Utah.

Development of the E. coli K-99 ELISA Test
A modified, double-antibody, sandwich ELISA was developed to detect the K-99 pillus antigen of enteropathic E. coli using a "U" bottom polystyrene plate for the solid phase. A mouse-derived monoclonal, K-99 antibody was used as one side of the sandwich, and a rabbit derived, anti K-99 antibody was the other side of the sandwich. A commercially available, anti-rabbit immunoglobulin, conjugated to horseradish peroxidase, was attached to the rabbit-derived, anti K-99 antibody. Orthophenylendiamine (oPD) was used as the substrate for the enzymatic reaction. The test was found to be sensitive and specific for the K-99 antigen. A complete description of the procedure has been submitted for publication.

K-99 E. coli and Rotavirus Submitted to the USU Diagnostic Laboratory
Ninety-two fecal samples were obtained from the digestive tract of calves presented to the Utah State University Veterinary Diagnostic Laboratory for necropsy. These fecal samples were
cultured for bacteria as a part of the routine diagnostic examination. Fecal samples were frozen for later rotavirus and K-99 E. coli ELISA testing. It became apparent, after examining the test results, that the data should be divided into two groups: results of tests from those dairy calves less than five days of age and those five days of age and older. Of the 34 cases in calves less than five days of age, 23 (68 percent) were positive with the K-99 E. coli. Of the 58 calves five days of age or older, only three (5 percent) were positive on Rota-ELISA and one (4 percent) cultured Salmonella. Combination infections of Rota and K-99 were seen in six animals (18 percent). Of the 58 calves five days of age or older, only three (5 percent) were positive on K-99 ELISA, 19 (33 percent) were positive on Rota ELISA and two (3 percent) cultured Salmonella. Only one Rota with K-99 case (2 percent) was observed.

It would appear that, whereas Rotavirus can affect calves over a wide range of ages, E. coli K-99 appears to be primarily a problem in calves up to five days of age.

### Epidemiology Study of Calf Diarrhea

Local veterinary practitioners were contacted and asked for client referrals of individuals who would be interested in cooperating in a study to determine the reservoir host of rotavirus and the K-99 E. coli. In the study, rotaviral infections were demonstrated in nine of ten dairy herds located in Cache County, Utah, and Franklin County, Idaho. A 25 percent incidence in calves under one month of age and a 3.6 percent incidence in animals one to eleven months of age was demonstrated. No rotavirus was detected in feces from animals older than eleven months. Approximately 67 percent of the rotavirus-infected animals were diarrheal and 33 percent were nondiarrheal at the time of fecal collection. No E. coli K-99 was detected. There appeared to be no difference in incidence between males and females.

### About the Authors

**Stanley D. Allen**, Doctor of Veterinary Medicine, is an associate professor in the Department of Animal, Dairy and Veterinary Sciences. He has been a member of the faculty at USU since 1979 and his main interests are in the area of diagnostic medicine and use of laboratory animals in research.

**Randy D. White**, Doctor of Philosophy, is a research assistant professor in the Department of Animal, Dairy and Veterinary Sciences. He has been a member of the faculty at USU since 1982 and his main interests are in the areas of nutritional toxicology and in-vivo toxicological testing.

Appreciation is expressed to Jeff Mitchell, Debbie Holley, Tania Clyde, Marie Lui and Carmen Garza for their technical assistance in the ELISA project, to the Utah State University Agricultural Experiment Stations for their support for Project III, to Dr. Bill Barnett for his timely advice in the development of the K-99 ELISA and to Dr. Ross Smart, director of the USU Diagnostic Laboratory, for his help in sample collection. We also express our appreciation to the dairymen who cooperated in the epidemiology study.

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<thead>
<tr>
<th>Type</th>
<th>Cause</th>
<th>Clinical Appearance</th>
<th>Treatment</th>
<th>Prevention</th>
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<tr>
<td><em>E. coli</em></td>
<td>Bacteria</td>
<td>Primarily seen in animals less than 5 days of age</td>
<td>Fluids with energy source and antibiotics</td>
<td>Vaccination1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Good supportive care2</td>
<td>Good sanitation and management</td>
</tr>
<tr>
<td>Salmonella</td>
<td>Bacteria</td>
<td>Often fatal in a herd; septicemia develops, seldom seen in calves less than 2-3 weeks of age</td>
<td>Fluids with energy source and antibiotics</td>
<td>Vaccination1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Good supportive care2</td>
<td>Good sanitation and management</td>
</tr>
<tr>
<td>Rotavirus</td>
<td>Viral</td>
<td>Diarrhea usually very watery (often yellow), seen in calves most often up to 28 days, but usually in calves 10-15 days of age</td>
<td>Fluids with energy source and antibiotics</td>
<td>Vaccination1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Good supportive care2</td>
<td>Good sanitation and management</td>
</tr>
<tr>
<td>Coronavirus</td>
<td>Viral</td>
<td>Seen in calves up to about 10 days of age</td>
<td>Fluids with energy source and supportive care2</td>
<td>Vaccination1</td>
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<td></td>
<td></td>
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<td>Good sanitation and management</td>
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<tr>
<td>Cryptosporidia</td>
<td>Protozoal</td>
<td>Anorexia, weight loss, nonresponsive to antibiotics</td>
<td>Coccidiostats</td>
<td>Coccidiostats</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>Good supportive care2</td>
<td>Good sanitation and management</td>
</tr>
<tr>
<td>Coccidiosis</td>
<td>Protozoal</td>
<td>Calves 3 weeks to 6 months of age. Rear legs and tail often plastered with feces containing traces of blood</td>
<td>Sulfa drugs and good supportive care2</td>
<td>Good sanitation and management</td>
</tr>
<tr>
<td>Nutritional</td>
<td>Improper diet</td>
<td>Calf has soft stool but is otherwise healthy until dehydration becomes severe</td>
<td>Balance ration with proper nutrients fed at proper intervals and good supportive care2</td>
<td>Proper feed and feeding practices3</td>
</tr>
<tr>
<td>Hypogamma- globulinemia</td>
<td>Lack of colostrum</td>
<td>Diarrhea seen along with other illnesses since little antibody is present</td>
<td>Intravenous gammaglobulin, serum or blood</td>
<td>Colostrom—best to feed at least 2 qt. twice per day for 3 days</td>
</tr>
</tbody>
</table>

1Norden Scour Guard = Rota-Corona-E. coli; Scourvax R = Rota-Corona; Scourvax-Rec = Rota; Ft. Dodge Coligen = E. coli; Connaught Vicogen = E. coli; Cutter PARATYFOL = Salmonella spp. (Listing of specific products does not imply endorsement by the authors or Utah State University).

2Good supportive care might include intestinal protectants and pH adjusters.

3Often, livestockmen feed soy based milk replacers instead of whole milk not realizing that calves can’t digest non-animal proteins until about 4 weeks of age. Often rapid changes in diet result in digestive upset. Calves do much better on multiple feedings per day rather than just one or two.

**TABLE 4. Look-a-like diarrhea in calves. (Mixed infections often occur.)**
PHOTOS

1. Range infested with locoweed (Oxytropis sericea) and close up of individual plant.

2. Cow showing signs of congestive right-heart failure after grazing locoweed at a high elevation (note excess fluid under the jaw and along the brisket).

3. Heart with enlarged right side from a cow with congestive right-heart failure.

4. Fluid also collects in thoracic region as result of congestive right-heart failure.

L. F. JAMES, D. B. NIELSEN, and K. E. PANTER
Young cattle grazing on certain high-altitude ranges infested with the locoweed (Oxytropis spp.) have been observed to have a high incidence of congestive right-sided heart failure. Ranchers report estimated frequencies in calves 1 to 6 months of age to vary from a few percent to nearly 100 percent morbidity, with 10 to 15 percent mortality.

The clinical signs are similar to those characterizing the disease known as High Mountain or Brisket Disease. At necropsy, affected calves show gross lesions of congestive right-sided heart failure (i.e., right ventricular hypertrophy and dilation, subcutaneous edema, ascites, hydrothorax, diarrhea, and chronic passive congestion of the liver). Microscopic examinations of tissues collected from these animals have revealed lesions of both congestive right-sided heart failure and locoweed poisoning.

Locoweed Poisoning
Locoweed poisoning results when livestock graze certain species of the genera Oxytropis and Astragalus. The disease is characterized clinically by emaciation, central nervous system depression, dull and lusterless eyes and coat, difficulty in propiation, and excitability under stress. Abortions may occur in pregnant animals. Fetuses taken from ewes fed the locoweed Astragalus lentiginosus between days 60 and 90 of gestation had subcutaneous edema and the right ventricle of the heart was enlarged suggestive of those observed in calves grazing locoweed on high-altitude ranges.

High Mountain Disease
High Mountain Disease is a term used to identify the syndrome of congestive right-sided heart failure in the bovine species and has historically been associated with residence at high altitude. The underlying genesis of congestive right-sided heart failure is from overwork of the right ventricle of the heart. In High Mountain Disease, the overwork is caused by increased pulmonary vascular resistance. The clinical signs and lesions associated with this condition are: right ventricular hypertrophy and dilation, subcutaneous edema, ascites, hydrothorax, diarrhea, and chronic passive congestion of the liver.

The incidence of High Mountain Disease in cow-calf operations above approximately 7,000 feet in Colorado and Wyoming varies from 0.5 to 2.0 percent. The incidence increases with higher resident elevations and newly imported cattle.

The observations that on certain high elevation ranges infested with locoweed (Oxytropis spp.) the incidence of congestive right-sided heart failure increased markedly, caused us to postulate that locoweed had an enhancing effect on this condition.

Field Observations
Ranges varying in elevation from 7,000 to 10,000 feet and infested with locoweed were visited over a period of about 10 years. It was observed that the incidence of right-heart failure was the highest in the years when locoweed was most prevalent. Calves were affected the most severely, with yearlings less so and mature animals even less. The calves were probably most affected because they receive the locoweed toxin in their milk as well as grazing the plants.

Interviewed ranchers using these areas felt that a relationship might exist between the locoweed and the congestive right-heart failure. After one rancher controlled the locoweed on his range with herbicides, his cattle had no more problems with heart failure.

All affected calves did not respond in the same way. Some swelled due to accumulation of fluid under the jaw and brisket area, others dehydrated, and a few swelled along the side of the jaws so the hair stood straight out. All, however, had diarrhea, became depressed, and developed dull hair and eyes. On postmortem examination, all appeared the same internally. Although these animals had microscopic lesions of both locoweed poisoning and congestive right-heart failure, they did not develop the typical signs expected in locoweed poisoning. Severely ill calves that are removed from locoweed areas and the high elevation generally recover.

Experimental Observations
To test the hypothesis that locoweed can predispose cattle at high altitude to congestive right-heart failure, cow-calf pairs were placed at an elevation of approximately 9,000 feet and fed green locoweed (O. sericea). At the same elevation, we were able to produce the congestive right-heart failure in 12 out of 13 calves fed locoweed while other calves not fed locoweed did not develop the problem. Calves at a much lower elevation, 4,500 feet, fed locoweed, O. sericea, did not develop the right-heart failure condition either.

Conclusions
We have concluded that the locoweed Oxytropis sericea can predispose cattle, especially calves, to congestive right-heart failure. The condition is similar to that described as High Mountain Disease. We do not suggest that High Mountain Disease is caused by the consumption of the locoweed Oxytropis sericea by cattle at high elevation. At least in some instances, however, plants or other factors in addition to high elevation may be involved.

ABOUT THE AUTHOR

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