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ESTIMATION OF SOME ENVIRONMENTAL AND GENETIC SOURCES
OF VARIATION AFFECTING PROTEIN QUANTITY IN SORGHUM
(SORGHUM BICOLOR (L.) MOENCH) GRAIN.

The University of Nebraska - Lincoln, Ph.D., 1973
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ESTIMATION OF SOME ENVIRONMENTAL AND GENETIC SOURCES
OF VARIATION AFFECTING PROTEIN QUANTITY IN SORGHUM
(Sorghum bicolor (L.) Moench) GRAIN

by

Richard F. Koenig

A DISSERTATION

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In Partial Fulfillment of Requirements
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Department of Agronomy

Under the Supervision of Professor C. O. Gardner

Lincoln, Nebraska

May, 1973

TITLE

ESTIMATION OF SOME ENVIRONMENTAL AND GENETIC SOURCES
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INTRODUCTION

Sorghum is used primarily for livestock feed in the United States and a few other countries, but in many countries sorghum is an important food in the human diet. Where sorghum is used for human consumption, the diet consists primarily of cereals which are generally low in protein. One way to increase the amount and quality of protein available to such people is through plant breeding and improved agronomic practices. In view of the exponential increase in the world's population, we must constantly search for superior genotypes and develop better farming practices to meet the need for abundant, high-quality foods. Even where sorghum is used for livestock feeding operations, there is a need for higher protein in the grain to reduce the amount of protein supplement which must be added to balance the diet.

To breed higher protein sorghums, breeders must know as much as possible about protein inheritance and the effects of environment on the character. Protein content of most grains is quite variable. Almost any environmental factor that will affect the plant also influences protein content of the grain. Soil fertility, water availability, amount of sunlight that the plant is able to intercept; and of course,

the genetics of the plant are only a few of the variables upon which protein content of the grain will depend.

The nature of the inheritance of quantity and quality of protein must be understood in order to initiate an efficient breeding program for their improvement. The breeder must know whether genetic variation for protein quantity or quality is largely due to additive gene effects or to dominance and epistatic effects, which lead to heterosis.

Protein quality is an important consideration in cereal crops. Quality of protein may be even more important than quantity. In this study most of the protein determinations were made using the Udy technique which indirectly measures protein quality as well as protein content. The Udy method measures the amount of basic amino acids, one of which is lysine, an essential and very limited amino acid in most plant proteins. Protein of low quality is of little value; therefore increasing the quantity of protein may or may not be of importance.

The objective of this research was to study environmental and genetic sources of variation affecting protein quantity in grain sorghum to provide information that would be useful in planning plant breeding programs to increase protein quantity in sorghum. Information is badly needed on variation in protein content in different parts of the same head, between different plants of the same genotype, and between different plots of the same genotype because such variations contribute to errors of estimation of genotype

means. If we are going to study genetic variation and develop breeding systems for increasing protein content, we need the best possible estimates of genotype means.

PREVIEW

REVIEW OF LITERATURE

Need to Improve Protein Content of Cereals

Is there a need to increase protein quantity through the production of cereals, or should cereal plant breeders work toward maximum production without regard for protein content and hope that people can get the needed protein from other plant and animal foods in their diet? According to findings of the United Nations Protein Advisory Group, from 40 to 70 percent of the protein consumed in developing countries comes from cereals (Sigurbjornsson, 1970). The cereals account for 71 percent of the world's harvested crop area (Brown, 1965). These two facts lead us to believe that cereals are, and most likely will remain, the most important portion of the diet of people throughout the world.

The world's population is increasing at a tremendous rate; and, even though food production has increased, we are not doing enough to keep up with the population increase. The world food production increased by 46 percent between the years 1952-56 and 1967; however, due to the growth in population, the per capita increase was only 13 percent (Jalil and Tahir, 1970). The difference between developed and developing countries was not great in the increase in total food production; but, due to disparities in population

growth, the per capita increase in food-crop production was 25 percent in the developed regions and only 6 percent in the developing countries (Jalil and Tahir, 1970).

Probably the most serious problem that faces mankind is the production of enough high-quality food to feed the ever-increasing numbers of people. One of the most important factors of food quality is the quantity and quality of protein contained in these food products. In this paper the author will be most concerned about quantity of protein; however, we must not overlook the fact that the quality is just as important to the well-being of mankind. Protein requirements range between 51 and 83 grams per person per day (Autret, 1970). This range depends upon age and sex of the individual. Some countries possess a considerable margin of safety; however, in 43 of the 88 countries studied by Autret, the average person had a mean consumption level below the estimated needs. These 43 countries represent a population of 900 million people, or 50 percent of the total number of inhabitants of the countries studied. In view of the predicted population increase by 1975 and 1985, an increase of 50 and 90 percent, respectively, in the total amount of protein available will be necessary (Autret, 1970).

Sigurbjornsson (1970) states that there are three methods which we might use as promising approaches to "bridging the protein gap": (1) Persuade plant-eating peoples to change their diet so that the proportion of such high protein foods as meat and fish is increased, and make

such foods available in sufficient quantities; (2) fortify the plant diet with added protein, possibly in the form of protein concentrates, synthetic amino acids, or some form of edible and palatable protein made of microorganisms, algae, fishmeal, etc; and (3) alter the composition of the plant so that, through the normal intake of staple plant food, the consumer is provided with adequate or nearly adequate amounts of nutritionally balanced protein.

He then indicated why he thinks these methods will not work. Persuading a vegetarian to eat meat is easier said than done; and, in many cases, it would take a new generation to bring about such a change. The protein additives would provide an attractive solution in theory but most likely would be faced with many formidable obstacles, especially in those areas where the problem is most acute. Such proteins or amino acids would be needed regularly and continuously, and transportation from the place of manufacture to the local market and from there to the consumer would be only one of the problems. Consumers would need to know the value of such foods and need to have something to pay or trade for protein concentrates.

Sigurbjornsson suggested that, although the first two methods have faults, it would be unfair if one did not concede that there is a place for all of the approaches. The last solution he states "offers the simplest and most lasting solution to the problem as faced by the majority of those suffering from protein malnutrition."

The total world protein production from plant sources for direct consumption in 1968 was 153.85 million metric tons, and the per capita production was 43.04 kilograms per year (Jalil and Tahir, 1970). The United Nations Protein Advisory Group concludes that increased production of cereal is the principle solution to the problems of world hunger and protein-calorie malnutrition and has encouraged research toward the improvement of cereal protein content and quality (Sigurbjornsson, 1970).

Sorghum Utilization

In 1966 sorghum grain ranked fifth in acreage of the crops in the world following wheat, rice, maize, and barley. It is grown in areas where the average summer temperature exceeds 20° C and the frost-free season is 125 days or more. In Europe and in parts of North America, sorghum is one of the least known of the leading world crops; however, in many warm-climate countries of the world, sorghum is a major source of food and feed. Between the years 1940 and 1968 the acreage of sorghum more than doubled, and the yields per acre increased more than three times (Martin, 1970).

The composition of sorghum grain is comparable to that of other grains grown extensively for food and feed. Sorghum grain is low in fiber and ash like corn and wheat. The oil content of sorghum is higher than rice, wheat or barley but lower than corn and oats. In amount of total available energy, sorghum ranks next to corn among cereal

grains. Sorghum grain has a slightly higher protein level than corn or rice (Wall and Blessin, 1970).

Environmental Factors which Influence Protein Content

The effect of environmental conditions is an important factor to be considered in any genetic study. The environmental factors that influence the character to be studied, in this case protein content of the grain, need to be known and need to be controlled as much as possible.

Doty et al. (1943) reported seasonal variations were responsible for variations in the protein and fat content of corn grain. The protein content was also affected by soil type and location. They found no significant differences in the chemical composition of more than 40 commercial hybrids and open-pollinated varieties of dent corn. Tanksley and Lyman (Quisenberry and Tanksley, 1970) analyzed 10 seedstock varieties of sorghum whose protein content ranged from 10.1 percent (Early Hegari) to 13.2 percent (Spur Feterita) at one location. Martin, RS 608, and RS 610 were also sampled from 20 locations in 12 states. When the samples were analyzed, the protein content varied from 8.5 to 15.9 percent in Martin, from 8.5 to 14.6 percent in RS 608, and from 7.8 to 14.1 percent in RS 610. Apparently season, location, variety or hybrid grown, soil fertility, soil type, and yield level can all have a very marked influence on the protein percentages of grain sorghum.

Nitrogen fertilization has been shown to increase protein content of grain sorghum (Burleson et al., 1956; Miller et al., 1964; Waggle et al., 1967; Campbell and Pickett, 1968) and wheat (Haunold et al., 1962). With low soil nitrogen conditions, Haunold et al. (1962) could not find significantly more protein in a known high-protein line. If soil nitrogen was not limiting, however, this line did produce a significantly higher percentage of protein.

The amount of water supplied to plants will also affect the protein content of seed (Fraps, 1931; Widtsoe, 1903). Increased water available to the plant tends to increase starch content relative to protein content; and, therefore, lowers the percent of protein. Fraps (1931) found a correlation of protein with rainfall of -0.576 ± 0.072 .

The effect of yield on protein content has been studied rather extensively in several of the cereals. In almost every case the percent protein was negatively correlated with yield. In sorghum, grain yield has been reported to be negatively correlated with protein percentage by several authors (Campbell and Pickett, 1968; Collins and Pickett, 1972; Liang, 1967; Liang et al., 1969). Protein content in maize has been studied for many years, and in each case grain yield has been reported to be negatively correlated (Dudley et al., 1971; East and Jones, 1920). Hayes and Garber (1919) reported a negative correlation between number of seeds produced per ear and protein content in corn. The same negative correlation has been found in wheat (Berg,

1941; Clark, 1926; Clark and Quisenberry, 1929; Grant and McCalla, 1949; Malloch and Newton, 1934; Neatby and McCalla, 1938; Waldron, 1933; Whiteside, 1936), barley (Grant and McCalla, 1949; Neatby and McCalla, 1938), and soybeans (Johnson et al., 1955). In two cases high protein content was reported not to be associated with low yields (Clark et al., 1928; Middleton et al., 1954). However, in almost every instance these authors stated that, even with the highly significant negative correlation, breeding for higher protein content and increased yield should be possible. Under some conditions, yield and crude-protein may even be unassociated or correlated positively as frequently as negatively. One reason for this might be low soil nitrogen availability in their studies. Haunold et al. (1962) found that percent protein was negatively correlated with yield at low levels of nitrogen. However, if nitrogen in the soil was sufficient, this was not true.

Locations, seasons, soil fertility, etc. may be major causes of variation in percent protein in cereal grains. If we are going to use a sampling procedure to evaluate genotype differences to produce high-protein varieties, we must be aware of the variation that exists between plots in a field, between plants within a plot, between heads of the same plant, and between sections within a head. Levi and Anderson (1950) studied this type of variation in wheat plants. They found protein contents of individual kernels of wheat, representing random samples taken from two plots

of 0.1 and two plots of 0.23 acres, were distributed within samples in an approximately normal manner with a range of at least 6 percentage units with a standard deviation of 1.4 units. They then sampled 68 Thatcher wheat plants from a 10-foot, 1-row plot and found a range of 2.7 percent and a standard deviation of 0.6 percent in protein content of those plants. Among heads of the same plant, the average range was 1.7 percent. Protein determinations were also made on each kernel in three plants. Mean values for individual spikelets were found to be normally distributed over a range of 5.1 percent with a standard deviation of 1.1 percent. In the top one-third of the head, the protein content of the spikelets tended to decrease towards the tip. Within spikelets containing three kernels, the middle kernel was slightly higher in protein content (15.9 percent) than the bottom kernel (15.7 percent). The top kernel was decidedly lower (14.7 percent). The protein contents of individual kernels within a plant were normally distributed over a range of about 6 percent with a standard deviation of 1.2 percent.

The same type of experiment by Ali et al. (1969) showed the bottom third of the spike to have the highest protein content (15.50 percent) followed by the middle portion (15.17 percent) and the top portion of the spike which had the least protein (14.83 percent). All differences were significant at the 5 percent level. Other research (McNeal and Davis, 1954; Knyaginichev, 1936; Stuber et al., 1962) has shown approximately the same results. Two of these authors (McNeal

and Davis, 1954; Knyaginichev, 1936) found the middle of the spike to be the highest in protein content while the other (Stuber et al., 1962) found the lowest third of the head equal to the center and the top third lowest in percent protein.

Another source of variation that needs to be considered is the effect of bagging sorghum heads to control pollination in breeding programs. Both yield and protein content may be affected. A study reported by Pickett (1969) was conducted to determine the effect of bagging and flag-leaf removal--both normal artificial pollinating procedures in sorghum--on grain yield, protein content, and seed size. Bagging and flag-leaf removal independently reduced grain yield, and a combination of both further reduced yield. Percent protein showed a slight, but significant, increase in both procedures. Seed weight did not change significantly.

Genetic Variance in Protein Content

Probably the most well-known selection experiment that has been continued through many cycles of selection is the Illinois high- and low-oil and high- and low-protein study. The Illinois study was started by Dr. Cyril G. Hopkins (1899) in 1896 with the Burr White variety of corn. The same study has been continued almost every year until the present time. The original variety had a mean oil content of 4.70 percent and a mean protein content of 10.92 percent.