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24th Central Alfalfa Improvement Conference: June 18-20, 1995, Ramada Inn, Spearfish, South Dakota

K. D. Kephart

B 721

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Agricultural Experiment Station
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Cover photo: *Niels Ebbesen Hansen (1866–1950) once said, "You don't get very far if you keep to the sure safe road all the time." On his eight trips to Europe and Asia, there sometimes were no roads at all. And he encountered both debilitating drought and blizzards, exposure, and bandits in his many months of searching for alfalfas to bring home to the central prairies of the United States. Germplasm from the plants and seeds that this first "Agricultural Explorer" for the U.S. Department of Agriculture brought back are in the lines of many of our best alfalfas now growing in this country.*

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24TH CENTRAL ALFALFA IMPROVEMENT CONFERENCE
SPEARFISH, SOUTH DAKOTA
JUNE 18-20, 1995

SUNDAY, JUNE 18

7:00-9:30 p.m. Registration and Reception, Poolside

MONDAY, JUNE 19

7:00 - 8:00 Registration and Continental Breakfast

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Lanny Rhodes, Chairman, CAIC.
Kevin Kephart, Local Arrangements Chairman, CAIC.
David Bryant, Dean, College of Agriculture and Biological Sciences, South Dakota State University.
Jim Gerwing, Soils Extension Specialist, South Dakota State University.

8:45 - 10:00 N. E. HANSEN COMMEMORATION
Historical Perspective. Kevin Kephart.
N. E. Hansen, the Man. David Gilkerson.
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10:40 - 11:00 **Winterhardiness and physiology of alfalfa germplasms developed with contrasting fall dormancy**. S.M. Cunningham, J.J. Volenec, and L.R. Teuber. Purdue University.

11:00 - 11:20 **Two cycles of divergent selection for root architecture in alfalfa**. J.F.S. Lamb, D.K. Barnes, and K.I. Henjum. University of Minnesota.

11:20 - 11:40 **Breeding yellow-flowered alfalfa for wildlife habitat and forage**. Arvid Boe, R. Bortnem, K.D. Kephart, and S. Selman. South Dakota State University.

11:40 - 12:00 **The effect of cultivars and environment on alfalfa seed quality characteristics**. S.R. Smith, Jr., R. Gjuric, and F.M. Katepa-Mupondura. University of Manitoba.

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1:00 - 1:20 **Effect of temperature and duration of leaf wetness on two components of aggressiveness in the alfalfa - alfalfa rust pathosystem**. Douglas Webb and Forrest W. Nutter, Jr. Iowa State University.

1:20 - 1:40 **Current procedures for field testing alfalfa cultivars for resistance to Sclerotinia Crown and Stem Rot**. L. H. Rhodes and R. M. Sulc. Ohio State University.

1:40 - 2:00 **Conclusions about alfalfa persistence, 1925 - 1995**. E.T. Bingham. University of Wisconsin.

2:00 - 2:20 **How good are alfalfa variety trials? A question of ethics and accuracy**. T. H. Busbice. Great Plains Research.

2:20 - 3:20 **DISCUSSION SESSION: Open forum for discussion of alfalfa variety testing issues**. Moderator, Bruce Anderson, University of Nebraska.

3:20 - 4:00 REFRESHMENT BREAK AND POSTER VIEWING

3:20 - 4:00 POSTER SESSION

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Root organic reserve accumulation and defoliation tolerance of alfalfa seedlings. N.E. Kalengamaliro, J.J. Volenec, S.M. Cunningham, and B.C. Joern. Purdue University.

Performance of alfalfa clones in crude-oil contaminated soils. W.L. Rooney, C.C. Wiltsie, Z. Chen, A.P. Schwab, and M.K. Banks. Kansas State University.

Influence of growth conditions on alfalfa protein degradability. D.Z. Skinner, I.E.O. Abdelgadir, T.K. Fish, and R.C. Cochrane. USDA and Kansas State University.

Detecting yield differences from alfalfa plots of various widths. Bruce Anderson, Greg Cuomo, and Mike Trammell. University of Nebraska.

Stem branch morphology of alfalfa cultivars. Kevin D. Kephart, Arvid Boe, R. Bortnem, and S. Selman. South Dakota State University.

4:00 - 5:00 CAIC Business Meeting.

6:00 Load busses for CAIC Dinner at Latchstring Inn, Spearfish Canyon (cash bar).

TUESDAY, JUNE 20

7:45 a.m. Load busses for Conference Tour:
Surface Gold Mine Reclamation
Mt. Rushmore (lunch on your own at the concessions)
near Rapid City, alfalfa tillage research
Return to Spearfish

CONFERENCE ENDS

SDSU

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Examining the Use of Various Factors to Predict Alfalfa Persistence in Wisconsin

Mark McCaslin and Wayne Helming
Forage Genetics

For several years alfalfa breeders have selected for improved winterhardiness within fall dormancy classes. This has caused increasing concern about the reliability of fall dormancy as a predictor of alfalfa winterhardiness. A standard test for the evaluation of winter survival per se was developed to address this concern. The experiments reported here were designed to examine the relationship between fall dormancy and winter survival indices generated from standard tests with winter injury in multiple location alfalfa trials after a "test winter".

The winter of 1994-95 caused significant winter injury in much of Wisconsin and Minnesota. This winter injury resulted in significant differences in persistence among several commercial and experimental alfalfa varieties planted in FG trials. These persistence differences were consistent over four tests in three Wisconsin locations seeded in 1992 and 1993. These same 42 varieties were also evaluated for fall dormancy and winter survival in nurseries established at West Salem, Wisconsin in 1993 and 1994. A persistence index was calculated based on survival and spring vigor after the second winter in the 1993 winter survival nursery.

Regression analysis was used to compare the relationship between mean % stand in the four yield trials (stand) with fall dormancy (FD), winter survival (WH), and persistence index (PI). The correlation coefficients describing these relationships are summarized in Table 1. Fall dormancy and winter survival scores were based on two year means, the persistence index was measured only in 1995.

Table 1. Relationships between various factors used to predict persistence

traits	r
FD/WH	0.01
WH/PI	-0.58**
FD/stand	0.03
WH/stand	-0.65**
PI/stand	0.78**

Thirty five FG experimentals and seven check cultivars were used in these trials. The check cultivars represent new varieties from each of the major alfalfa breeding companies. These data suggest that fall dormancy is ineffective in predicting winterhardiness in modern alfalfa germplasm. The winter survival index is a more effective predictor of winterhardiness, but appears to have some limitations. The persistence index, measured after the second winter in winter survival nurseries, appears to have the closest association with winterhardiness. This data was collected only in Wisconsin. Additional trials are being conducted to test these conclusions in other northern alfalfa production areas.

References

1. Barnes,D.K., D.M. Smith, L.R. Tueber, and M.A. Peterson. 1991. Fall dormancy. *In* Standard Tests to Characterize Alfalfa Cultivars, C.C. Fox (ed). North American Alfalfa Improvement Conference, Beltsville, MD.

2. McCaslin, M.H., W.T.W Woodward, and C.C. Fox, 1991. Winter survival. *In* Standard Tests to Characterize Alfalfa Cultivars, C.C. Fox (ed). North American Alfalfa Improvement Conference, Beltsville, MD.

3. McCaslin, M.H., Don Brown, H. Deery and D. Miller. 1990. Fall dormancy and winter survival in alfalfa: variation within and between populations. Proc. 32nd NAAIC. Pasco, WA.

Winter Hardiness and Changes in Bud and Taproot Physiology of Alfalfa Cultivars Selected for Contrasting Fall Dormancy

S.M. Cunningham[1], J.J. Volenec[1], and L.R. Teuber[2]

[1]Department of Agronomy, Purdue University, West Lafayette IN 47907,

[2]Department of Agronomy and Range Science, University of California - Davis CA 95616

Winter hardiness is essential for alfalfa (*Medicago sativa* L.) grown in northern latitudes of the U.S. Winter hardiness has long been associated with fall dormancy, the reduction in shoot growth in autumn. Understanding the physiological and biochemical bases for genetic differences in fall dormancy and freezing tolerance could provide opportunities to genetically enhance winter hardiness of alfalfa. Our objectives were to 1) determine how divergent selection for fall dormancy influenced alfalfa winter survival; and 2) examine if changes in winter survival were associated with altered metabolism of sugars, starch, and protein in taproots and crown buds. Cultivars used as parents in the selection scheme were 'Wadi-Qurayat' (frost-sensitive, fall nondormant), 'CUF 101' (nondormant), 'Lahontan' (semi-dormant), and 'Norseman' (fall dormant). The third cycles from selection for enhanced fall dormancy (less shoot growth) and less fall dormancy (greater shoot growth) and their respective parent cultivars were established in the field in two successive years. Plants were harvested in Sept., Oct., Nov., Dec., and March. Taproots were separated from crowns, and divided into the top 2.5 cm of the taproot, and the remaining root tissue. White buds and green buds (those containing chlorophyll) were removed from crowns and analyzed separately.

Selection for less fall dormancy did not increase fall height of 'Wadi-Qurayat' or 'CUF 101', whereas, fall height of 'Norseman' and 'Lahontan' were increased by selection for less fall dormancy. Selection for greater fall dormancy reduced fall height of 'Lahontan' and 'CUF 101', but not 'Norseman' or 'Wadi-Qurayat'. Wadi-Qurayat and selections from it did not survive winter. Selection for greater fall dormancy improved winter survival of 'CUF 101' from 1% in original 'CUF 101' plants to over 90% in 'CUF 101' plants selected for greater fall dormancy. Winter survival of other cultivars was not affected by selection.

Buffer-soluble proteins in root tops of parental cultivars and selections that survived winter increased from Sept. to Dec., and were about 20% higher in Dec. than that of plants that winter killed. Sugar levels increased 3.5 fold from Sept. to Dec. in 'Norseman' root tops. Starch levels remained highest throughout autumn in the root tops of 'Wadi-Qurayat.' Sugar levels in green and white buds increased throughout autumn in all cultivars, but reached higher levels by Dec. in buds of plants that survived winter. In contrast white and green buds of more winter hardy cultivars contained more starch in late fall than the buds of the less winter hardy cultivars.

CUF 101 and its selections provide opportunity to characterize the physiological and biochemical bases for fall dormancy and its relationship to winter hardiness in alfalfa. Further studies will include HPLC analysis of sugars and qualitative analysis of membrane and buffer-soluble proteins from root tops and white buds throughout autumn to elucidate specific changes occurring in alfalfa as it hardens for winter.

Two Cycles of Divergent Selection for Root Architecture in Alfalfa

J.F.S. Lamb, D.K. Barnes, and K.I Henjum

Fertilizer N is the single most expensive input in most crop productions systems and has been implicated in declining ground water quality due to nitrate contamination. Improvements in N cycling and efficient use of symbiotically-fixed N can have a marked effects on the economic and environmental impact of agricultural systems. A major limitation to better management of symbiotically-fixed N from alfalfa is the lack of varieties with specific characteristics that influence the N cycle. Johnson (1992) showed that modern alfalfa varieties (released after 1980) had little variability for root morphological traits. Most alfalfa varieties were tap rooted with a few secondary roots and a small amount of fibrous roots. Older varieties (released before 1970) and Plant Introductions showed a large amount of variability for the number of secondary roots, the amount of fibrous root mass, and taproot diameter. The objective of this study was to evaluate experimental alfalfa germplasms that have undergone two cycles of divergent selection for root morphological traits for the number of secondary roots, amount of fibrous root mass and taproot diameter.

Four experimental alfalfa germplasm sources were evaluated after two cycles of divergent selection for root architecture. MN NDRN (dormancy= 8), and MN FLEM (dormancy=5) were selected for few (T, taproot) vs. many (B, branched) secondary roots. MN NCPL and MN MWNC (dormancy=3) underwent two cycles of selection for no or few (LF, low fibrous) vs. many (HF, high fibrous) fibrous roots and one cycle of selection for few (T) vs. many (B) secondary roots. Selected and unselected populations were established in two experiments at both Becker, and Rosemount, MN in May 1994. The experimental design was eight replications of a randomized complete block with a split plot arrangement of the treatments, with fertilizer rates (0 kg N ha-1 and 200 kg N ha-1) as whole plots and alfalfa populations as subplots. One experiment at each location was dug in October 1994 and evaluated for taproot diameter (measured in mm), number of secondary roots (scored, 1= few, 5= many), and amount of fibrous roots (scored, 1= few, 5=many). The remaining experiment at each location will be evaluated in 1995.

MN NDRNB$_{C2}$ and MN FLEMB$_{C2}$ had more secondary roots while MN NDRNT$_{C2}$ and MN FLEMT$_{C2}$ had fewer secondary roots than their unselected parent populations. The branched selections had more fibrous roots than the unselected population in the MN NDRN source, but there was no difference in fibrous root score among the MN FLEM populations. MN MWNCHF$_{C2}$B and MN NCPLHF$_{C2}$B had more fibrous and secondary roots when compared to there unselected parent populations. MN MWNCLF$_{C2}$T was not significantly different from the unselected parent population for fibrous or secondary root score. MN NCPLLF$_{C2}$T had the same amount of fibrous root but fewer secondary roots when compared to its unselected parent population. Preliminary results indicate that two cycles of divergent selection for root morphological traits in all four germplasm sources produced populations that were significantly different in root architecture.

References:

Johnson, L.D. 1992. Morphology and genetics of root types in alfalfa. Ph.D. Thesis. University of Minnesota.

Breeding Alfalfa for Combined Wildlife Habitat and Forage Purposes

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There is expanding interest in the northern Great Plains in the use of alfalfa for both wildlife habitat and forage purposes. Pheasants and ducks utilize alfalfa fields as nesting sites in spring and early summer in our region. However, haying procedures in early June often destroy nests, eggs, and incubating hens (McCabe et al. 1956). Yellow-flowered alfalfa (*Medicago sativa* ssp. *falcata*) has several traits that lead us to believe it offers promise for this purpose. It has high levels of winter hardiness and drought tolerance, prolonged flowering, and more tolerance than common hay and pasture types to potato leafhopper yellowing (Boe et al. 1994, Bortnem et al. 1993, 1994).

The objectives of our research are: (1) compare yellow-flowered germplasms to standard hay- and pasture-type alfalfas for yield and quality in a system where forage is stockpiled until late July to enhance gamebird production, and (2) develop a new cultivar with the morphological and forage quality characteristics that make it especially suitable for combined nesting cover and stockpiled forage purposes.

Forage yield and quality and insect resistance data for yellow-flowered alfalfa germplasms compared to standard hay- and pasture-type cultivars were collected from replicated seeded and spaced-plant trials during 1992-1994 at 3 locations in eastern South Dakota.

Forage production of alfalfa stockpiled until mid or late July was highest for entries that had yellow-flowered alfalfa as their sole or primary source of parental genotypes. However, our preliminary data suggest that germplasms with high levels of yellow-flowered alfalfa in their pedigrees were of poorer quality than many standard hay or pasture types under this delayed first-harvest system when potato leafhopper infestations were not severe. We feel that if alfalfa is intended to be stockpiled until mid July that tolerance to potato leafhopper yellowing is crucial. Timing and level of potato leafhopper infestation are difficult to predict, but if the insects arrive early in the growing season they can cause significant reductions in yield and quality of susceptible cultivars if the forage is stockpiled until mid July. In the northern Great Plains, it is common to obtain only one harvest of alfalfa in a growing season. Germplasms with high levels of yellow-flowered alfalfa in their parentage appear promising for 1-cut systems that will provide nesting habitat for gamebirds in the spring and abundant forage after young gamebirds have fledged. Since forage yields of yellow-flowered germplasms exceeded those of standard and hay- and pasture-type cultivars in this delayed-harvest system (Boe et al. 1994), future work will focus on selecting for improved quality in yellow-flowered alfalfa. We have selected about 30 genotypes from yellow-flowered accessions from the former Soviet Union for tolerance to potato leafhopper yellowing, prolonged flowering, vigor, and leaf retention. Seed of this selected germplasm was increased under isolation in Washington in 1993 and 1994. Hopefully, data obtained from trials established in 1994 and 1995 in South and North Dakota will provide support for the release of a new yellow-flowered cultivar for combined wildlife habitat and forage purposes in the late 1990's.

References

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The Effect of Cultivar and Environment on Alfalfa Seed Quality Characteristics

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Development of an establishment year seed production system would allow contract multiplication of non-winterhardy cultivars in western Canada, but may have a negative effect on seed quality as these cultivars are removed from their area of adaptation. The objective of this research was to study the effect of cultivar and environment on the following alfalfa seed quality characteristics: seed weight/size, seed colour, percentage germination, hard and non-viable seed. Nine alfalfa cultivars covering the whole range of fall dormancy classes were included in a series of field experiments at three locations in Manitoba over the period 1992-1994. Differential success in establishment and winter survival resulted in seed being harvested in only seven location-years (four establishment year stands and three second year stands). Seed harvested from these experiments were tested for seed quality using standard germination tests and by using a computer Digital Image Analysis (DIA). Measurements of plant seasonal development showed that the most fall dormant cultivars, 'Rangelander' and 'Algonquin', had slower early spring growth and development, but these differences diminished later in the season.

Variable environmental conditions (cool temperatures and high rainfall) negatively influenced pollinator activity and seed development during all years of this study. Over the range of location-years the cultivars 'Arrow', 'Algonquin', Cimmaron VR', 'Rangelander' and 'Florida 77' produced higher seed yields than 'Wilson', 'Moapa 69' 'Nitro' and 'CUF101'. The top five cultivars also showed a consistent, though non-significant, ranking for seed yield even though yields ranged from 1 to 350 kg ha^{-1} over all location-years. All seed quality characteristics were under a strong influence of the environment. There were also significant cultivar differences for seed weight and percentage hard seed. Even the non-dormant cultivars showed hard seed development above 45% in 5 out of 7 location years.

The most fall dormant cultivar, Rangelander, consistently developed the highest percentage hard seed and the lowest seed weight. The measurements of seed characteristics through DIA (size and colour) were strongly correlated with measurements of seed quality through standard tests and showed potential for development of alternative or supplementary methods to measure seed quality.

In conclusion, the potential for seed production during the establishment year is unpredictable in western Canada, but successful winter survival suggests that contract multiplication may be possible of medium dormancy cultivars (2 to 3 year stands). Hard seed content was controlled by both genotype and environmental conditions, but the environment had the strongest influence on percentage hard seed.

Effect of Temperature and Duration of Leaf Wetness on Two Disease Components of Alfalfa Rust

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The Standard Tests used to Characterize Alfalfa (*Medicago sativa* L.) Cultivars published by the North American Alfalfa Improvement Conference recommends a constant temperature of 25 C for all phases of the alfalfa rust (*Uromyces striatus* Shroet.) monocycle when testing alfalfa genotypes for resistance to this pathogen. Following inoculation, a 24-hour leaf wetness period under darkness at 25 C has been recommended to facilitate the infection process (germination and penetration). During the latent and infectious periods of a monocycle, it is recommended that plants be maintained at 25 C with a 16 hour photoperiod. While the methods recommended in the Standard Tests have proven useful in differentiating the resistance and susceptibility of alfalfa genotypes based on the size and number of pustules, further information is warranted on the environmental conditions necessary to optimize individual disease components such as infection efficiency and latent period. Therefore, the objectives of this study were to (i) quantify the effects of leaf wetness duration and temperature on infection efficiency and (ii) quantify the effect of temperature during the latent period of infection efficiency, latent period and the rate of pustule appearance.

Alfalfa plants were inoculated with urediospores of *Uromyces striatus* and then subjected to leaf wetness durations of 4, 8, 12, 16, 24 or 32 hours at 20 C. There was a significant positive, linear relationship between hours of leaf wetness and infection efficiency (pustules/leaf) from 4 to 24 hours ($P \leq 0.001$, $r^2 = 0.96$). To determine the effect of temperature during a 24-hour period of leaf wetness following inoculation, inoculated plants were subjected to temperatures of 17, 19, 22, 25 or 28 C. There was a significant negative and linear relationship between temperatures during the leaf wetness period and infection efficiency ($P \leq 0.001$, $r^2 = 0.98$). Infection efficiency was more than twenty times higher at 17 C than at 28 C. Constant temperatures of 15, 18, 21, 24, 27, or 30 C after the initial 24-hour leaf wetness period did not effect infection efficiency (pustules/cm^2), but did effect latent period (the time from inoculation to the time when 50% of the pustules were visible). There was a significant negative and linear relationship ($P \leq 0.001$, $r^2 = 0.94$) between temperature and latent period ($\ln T_{50}$). Thus, temperature during the initial 24-hour leaf wetness period is more critical and has a greater impact on infection efficiency than post-infection temperatures. The rate of pustule appearance increased as temperature increased and the rate of pustule appearance (as affected by temperature) was best described by the Gompertz population growth model.

Current Procedures for Field Testing Alfalfa Cultivars for Resistance to Sclerotinia Crown and Stem Rot

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Sclerotinia crown and stem rot (SCSR), caused by Sclerotinia trifoliorum, is a serious disease of late-summer seeded alfalfa. Although differences in resistance to SCSR among alfalfa cultivars have been reported, all cultivars presently available may be severely damaged when inoculcum concentration is high and environmental conditions are favorable for disease development. Identification of cultivars or experimental lines with commercially acceptable levels of resistance to SCSR would be a major advance in controlling this disease. In 1991, a program of field testing of alfalfa cultivars and experimental lines was begun at the Ohio State University Waterman Farm, Columbus, Ohio. Entries are submitted by alfalfa seed companies and evaluated under field conditions. Although some modification of procedures has occurred since the beginning of the testing program, the following procedures are currently being employed.

A red-clover/orchardgrass sod is seeded at least one year prior to the establishment of the alfalfa trial. Grain inoculum, consisting of 1 part wheat to 1 part oats colonized by S. trifoliorum, is spread uniformly throughout the red clover/orchardgrass sod during the fall or spring. Red clover plants become infected from mycelium growing from the colonized grain and sclerotia are produced on stems and crowns of infected plants. During the spring and summer, sclerotia fall to the soil surface and gradually become buried in the upper 1 cm of soil.

Alfalfa entries are seeded in late August or early September using no-till methods. Irrigation is supplied as necessary to insure stand establishment and sclerotial germination. Typically, apothecia emerge in October or November and release ascospores which infect alfalfa plants. The primary infection period usually continues until mid-December. To achieve a disease-free control treatment, Vinclozolin (Ronilan 50 DF) is applied to plots of Armor alfalfa at 2 lb. formulated product per acre in mid-September, mid-October, mid-November, and mid-March. Unsprayed Armor serves as a susceptible check cultivar.

Plots are monitored from February through May for symptoms of SCSR. Data on disease development (percent of plot area affected by Sclerotinia) are recorded from late winter through spring as needed. Beginning in late May, plots are harvested 4 times at approximately 35-day intervals, and dry matter yields are determined for each entry. In October, plots are undercut and industry representatives are invited to select plants for breeding programs.

Conclusions About Alfalfa Persistence, 1925 - 1995

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Conclusions about alfalfa persistence are based on project experiments, and the writings and communications of R.A. Brink, L.F. Graber, Dale Smith, R.P. Murphy, Pat Palmer, Derek Woodfield, and R.B. Ipson. The list is not complete and is continuously refined.

- Persistence is essentially adaptation.
- Adaptation is the most fundamental trait in the perenniality of alfalfa, and perenniality requires persistence.
- Persistence has its own genetic base. The base is not necessarily the same for all materials with equivalent persistence. And, there is much interaction with specific traits.
- The genetic base is not fixed in most materials and requires selection to maintain it.
- Persistence usually decreases over sexual generations for other traits.
- The more intervening sexual generations for other traits, the greater the potential for loss in persistence.
- The loss in persistence over generations fits classical population genetic theory and may be due to genetic drift, negative genetic linkages with other traits being selected, and/or more complicated factors associated with lack of selection pressure.
- Winter hardy plant materials may be fall dormant or relatively non-fall dormant.
- Winter hardy plant materials are not always persistent. Hence, some materials may overwinter one or two winters just fine, and then crash due to lack of persistence.
- Persistent plant materials may be fall dormant or relatively non-fall dormant.
- The benefits of pest resistance are best obtained in persistent alfalfa. High levels of multiple pest resistance do not necessarily ensure persistence.
- The best method to ensure persistence is to select for persistence. But, selection of plants persisting in depleted stands with little or no competition is often ineffective. Selection of plants persisting in competitive stands is most efficient.
- Selection for persistence lengthens the breeding cycle. Nonetheless, if a new cycle is started every year there is only one lag-phase and eventually a new product can be selected every year.

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How Good are Alfalfa Variety Trials? A Question of Ethics and Accuracy

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Recommendations are made, alfalfa varieties are chosen, and millions of pounds of alfalfa seed are sold based on alfalfa trial data of questionable value. Alfalfa trials are subject to fraud and often contain large experimental errors.

Fraud in variety testing can occur when unbiased sampling is not practiced. Clever ways of rigging alfalfa trials are as follows:

(1) Entering selected sample (F1 hybrid or early generation) that is known to give better performance than the commercial variety that the sample represents.
(2) Sizing and polishing seed to improve initial stand and seedling vigor.
(3) Entering a variety under an experimental number or several numbers. This is effective when experimental numbers are changed to variety names after the fact. That is, data are given a variety name only when the experimental yields high in the test. Multiple entry of the same variety under several numbers insures success.

Alfalfa yield trials are particularly subject to fraud because early generations may yield much better than the commercial generation of a variety.

Effect of generation on yield of a six-clone alfalfa variety (1)

Generation	Forage Yield % of Check
Best F1 Hybrid	150
Syn 1 (Breeder Seed)	132
Syn 2 (Foundation Seed)	116
Syn 3 (Commercial Seed)	109

Fraud can be prevented by testing only commercial seed obtained by the scientist from the local market.

Large experiment errors occur when too many entries are included in a test, making it impossible to get uniform conditions within an experimental block. Such error can be reduced by limiting the number of entries to, perhaps, no more than 36. Such error can be further reduced by reducing plot size, perhaps to 60 sq. ft. maximum.

Many tests have inadequate replication. Steel & Torrie (2) provides a formula for estimating the replication required, based on past experience. Experience teaches that more than four replications are often needed.

The value of an alfalfa variety trial can be estimated by the modified coefficient of variation. M.C.V. = 100 (LSD)/test mean. The LSD expressed as a fraction of the mean will estimate the percentage difference between varieties that can be detected at a stated confidence level. A value less than 10% is required for a yield test to have practical value. Forage yields of adapted commercial alfalfa varieties are not expected to differ by more than 10%.

The proper sizing of trials and careful attention to experimental techniques can effectively control error.

Reference

(1) Evaluating Parents and Predicting Performance of Synthetic Alfalfa Varieties. 1976. ARS-S-130.
(2) Steel and Torrie. 1960. Principles and Procedures of Statistics. Page 154.

Interaction of Nitrogen and Potassium Nutrition on Growth and Root Physiology of Alfalfa

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Potassium (K) deficiency reduces alfalfa (*Medicago sativa* L.) shoot growth as well as root protein and carbohydrate concentrations. Nitrogen (N) fertilizer may replace N normally acquired via N_2-fixation, a process that is severely reduced by K deficiency. Our objective was to determine if N fertilization alters shoot growth, and root carbohydrate and protein accumulation in alfalfa plants receiving varied levels of K. 'Resistar' alfalfa plants were defoliated and transplanted into quartz sand where they were provided Hoagland's solutions containing 10 mM N as NO_3^- or NH_4^+ in 0, 3, and 6 mM K for 90 days. Plants were defoliated at 30-day intervals. Plants were sampled immediately after the third defoliation, and at 7-day intervals thereafter during 28 days of regrowth. At sampling, plants were separated to shoots, roots, and crowns. Roots were washed free of sand, lyophilized, and milled to pass a 1-mm screen. Root tissues were analyzed for total N, K, sugar, starch, and buffer-soluble protein.

Root K concentrations increased as K levels of the nutrient solution increased. Root K concentrations were similar for all N treatments when plants were supplied 0 and 3 mM K, whereas, root K concentration was reduced in NH_4^+-treated plants receiving 6 mM K. Root N concentrations were low at 0 mM K, and increased with K nutrition. Addition of N increased root N concentrations only in plants receving 0 mM K. The NH_4^+-treated plants accumulated slightly higher root N levels at 3 and 6 mM K when compared to NO_3^--treated and control plants. Survival of NH_4^+-treated plants was reduced, but survival improved as solution K levels increased. Shoots per plant increased as solution K concentration was raised from 0 to 3 mM. For control and NO_3^--treated plants, mass per shoot increased markedly as K was raised from 0 to 3 mM, whereas, 6 mM K was required to increase mass per shoot of NH_4^+-treated plants. Root mass per plant was reduced by NH_4^+ application. Root starch concentrations increased significantly as solution K concentration increased from 0 to 6 mM, and were severely reduced in NH_4^+-treated plants. Root sugar concentrations were unaffected by K nutrition. Root protein concentrations were not affected by N application, but increased as solution K concentration increased from 0 to 3 mM.

Application of N as NO_3^- had little impact on growth or taproot physiology of alfalfa irrespective of K nutrition. Alfalfa persistence, shoot growth, and taproot starch were reduced by NH_4^+ applications, especially in plants receiving 0 mM K.

Root Organic Reserve Accumulation and Defoliation Stress Tolerance of Alfalfa (*Medicago sativa* L.) Seedlings

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Previous results have suggested that defoliation tolerance of alfalfa (*Medicago sativa* L.) depends on root organic reserves. Our seedling development studies have shown that deposition of starch and vegetative storage proteins (VSP) in alfalfa roots begins approximately 40 days after planting (DAP). We hypothesized that seedlings defoliated prior to 40 DAP would have impaired regrowth. Our objectives were: (1) to determine if combined inorganic N would stimulate early VSP deposition in alfalfa roots; and (2) to determine if defoliation tolerance of seedlings depends on root organic reserve accumulation. In Exp. 1, normal and ineffective-nodulating Saranac were grown in coarse sand with and without combined N. Roots were sampled 15, 25, 39, 55, and 67 DAP and analyzed for sugars, starch, and VSPs. Root starch and VSP concentrations were low 15 and 25 DAP. There was rapid accumulation of root starch and VSPs between 25 and 55 DAP in Saranac without N and Saranac and ineffective-nodulating Saranac with N. Roots of ineffective Saranac without N did not accumulate starch or VSPs during this period. Combined N increased herbage growth, but did not stimulate early deposition of starch or VSPs. This suggested that initial VSP deposition may be controlled by factors other than N availability.

In Exp. 2 seedlings were completely defoliated 14, 28, 42, and 56 DAP. Results showed that 78% of seedlings survived complete defoliation 14 DAP. At this time root starch levels were very low (20 µg/mg dry wt.) and VSPs could not be detected using immunoblotting. In Harvest 3 and 4, when root starch and VSP accumulation began, 80% of the seedlings survived. However, at Harvest 2 (28 DAP) only about 50% of seedlings survived complete defoliation. This low survival rate was associated with the largest defoliation-induced declines in root protein and sugar concentrations. This suggests that there may be threshold sugar and protein concentrations in roots required for seedling survival.

The results from these experiments show that starch and VSPs were not abundant in roots of young alfalfa seedlings (<30 DAP). In addition, VSP accumulation was coordinated with root starch deposition. The onset of starch and VSP accumulation coincided with declines in buffer-soluble protein and sugar concentrations in roots. Addition of fertilizer N increased herbage growth but did not lead to early VSP deposition. High levels of starch and VSPs may not be an absolute necessity for defoliation tolerance of young alfalfa seedlings.

Performance of Alfalfa Clones in Crude Oil Contaminated Soils

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Phytoremediation of crude oil contaminated soils may be the most economical means of cleaning contaminated soils. In studies to compare the phytoremediation potential of different species, alfalfa was effective in the remediation of crude oil contaminated soil. Given the highly heterozygous nature of alfalfa, it is likely that variability exists for performance of individual alfalfa plants in crude oil contaminated soil. The objectives of this study were to determine if agronomic varies in contaminated soil between selected alfalfa clones, and (2) to determine if differences exist between clones for TPH degradation rates in the soil. In a twelve month greenhouse experiment, agronomic and soil contaminant data were collected for twenty alfalfa genotypes from the cultivar 'Riley'. Significant variability was detected between genotypes grown in contaminated soil for forage yield, plant height and maturity at clipping. Forage yield in the contaminated soil was only 33% of the forage yield of the same clones in uncontaminated soil. Total petroleum hydrocarbon degradation was highly correlated with total forage yield and TPH levels were lower in vegetated pots than in unvegetated controls. The results indicate that overall growth and vigor are reduced in contaminated soil, but that suitable variability exists among genotypes in contaminated soil that selection for improved performance is feasible. High correlations between plant growth and TPH degradation rates indicate that selection for enhanced degradation may be accomplished by selection of the plants with the highest forage yields.

Influence of Growth Conditions on Alfalfa Protein Degradability

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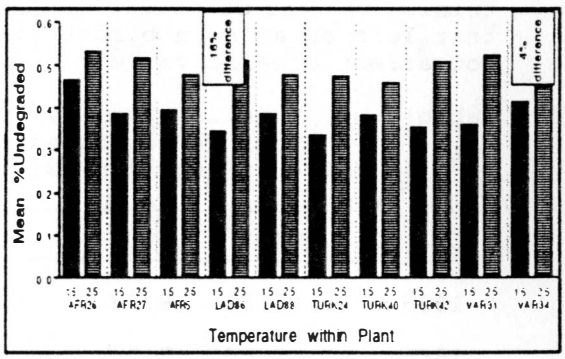
The shortcomings of alfalfa (*Medicago sativa* L.) as a high quality forage include extensive ruminal degradation of the forage protein, sometimes with concomitant loss through ammonia formation. Previous research has shown that the degradability of total alfalfa protein varies among individual plants, and among harvests of the same plants (Skinner *et al.*, 1994). In this report, the effect of environmental conditions during the growth of alfalfa plants was investigated as a source of significant variation in protein degradability. Ten plants of diverse origins were vegetatively propagated and genetically identical ramets were grown in rigidly-maintained environments differing in temperature and humidity [25C and 32% average relative humidity (rh) vs. 15C and 61% rh) . Some hay quality characteristics related to protein differed among plants grown under the same conditions (example in Table 1). Total protein from plants grown at 25C was consistently more resistant to degradation than the protein from the same plants grown at 15C (Fig. 1). Differences among degradable fractions between the environments were not consistent across genotypes (Fig. 1). These results suggest that genotypic effects for quality characteristics expressed under rigidly-maintained environments exist, and that environmentally stable genotypes may exist. Selection for reduced degradability among environmentally stable genotypes likely will result in populations with reduced degradability expressed over a broad range of growth conditions.

Table 1. Genotypic Effects on Quality Characteristics of Alfalfa Hay Grown at 15C and 61% rh.

	Plant		
Item	Varia-34	Ladak-86	σ_m
N, %of DM	2.09	2.39	0.12
ADF, %of DM	26.70	26.90	0.23
NDF, %of DM	34.50	35.90	1.43
Ash, %of DM	8.75	8.80	0.33
ADIN, %of total N	5.05	5.10	0.74
NDIN, %of total N	10.25	7.45	0.46
PoolA[1], %of total N	32.67	38.20	4.32
Pool B, %of total N	56.23	47.95	3.91
PoolC, %of total N	11.10	13.85	0.44
UIP[2]	23.30	21.34	0.96

[1]Pool A=nonprotein nitrogen; pool c=48h residual nitrogen from *Streptomyces griseus* protease in vitro method; pool B=potentially available nitrogen.
[2]UIP= Undegradable intake protein.

Figure 1. Degradation of protein from 10 plants grown under different conditions.



Reference:Skinner, D. Z., J. O. Fritz, and L. L. Klocke. 1994. Protein degradability in a diverse array of alfalfa germplasm sources. Crop Sci. 34:1396-1399.

Detecting Yield Differences from Alfalfa Plots of Various Widths

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Alfalfa variety trials currently are conducted with 2.5 to 8 foot plot widths and yield collected from 38 to 100 percent of the plot. Border effects influence yields of adjacent plots in some plot studies. Many investigators assume these effects are negligible in alfalfa trials, possibly because plots are harvested at nearly the same time and characteristics like plant height do not appear to favor one population over another. Thus, many alfalfa variety trials have no side borders for their plots, resulting in harvest of the entire plot for yield. We examined the effect of various plot widths on alfalfa variety yields and the statistical ability to detect differences among these varieties.

'Spredor II', 'Dawson', and 'Wrangler' alfalfa were seeded in 1988 near Mead, NE in plots containing 5, 10, or 15 rows spaced six inches apart with twelve inches between plots. Plot length was twelve feet with a three foot border of alfalfa on each end. Plots were arranged as a split-plot in a RCBD with six replicates. A three-foot wide strip was harvested from the center of each plot using a flail harvester from nineteen total harvests during five years. Thus, entire 5-row plots were harvested and alfalfa borders remained on 10-row and 15-row plots.

Spredor II, Dawson, and Wrangler yields averaged 5.40, 5.51, and 5.94 tons/acre annually, similar to historical yields at this location. Plot width did not affect yields even though six or seven rows were harvested from 10- and 15-row plots in contrast to just five rows harvested from the 5-row plots. Experiment wide variation (C.V.) and ability to detect differences in variety yields (F values) differed little among plot widths (Table 1). Plots with borders (10- and 15-rows) tended to have higher F values when several years of data were combined. This should result in greater ease in detecting significant differences in yield, but sensitivity analyses indicated there were no technique differences ($P > 0.05$). Interactions of plot width and variety were insignificant (Table 2), suggesting that yield comparisons among varieties did not change due to plot width.

To conclude, plots with 5 rows spaced 6 inches apart and harvested with a three-foot wide flail-type plot harvester (commonly used for many alfalfa variety yield trials) were as effective at comparing yields of alfalfa varieties as wider plots that left an alfalfa border following harvest, indicating that plot width does not affect alfalfa variety trials.

Table 1. F values and C.V.'s of alfalfa variety yield tests using 5-row or 15-row plot widths.

	5-row		15-row	
Year	F	C.V.	F	C.V.
1988	8.87	9.12	9.54	8.06
1989	1.84	7.52	0.60	6.72
1990	20.05	5.75	15.58	5.70
1991	2.06	6.29	2.13	7.30
1992	3.77	8.04	9.64	6.37
1989-1992	3.34	4.56	8.40	4.19
1988-1992	4.27	4.25	9.33	3.98

Table 2. Interactions[1] of plot width and variety.

Year	F value	Pr > F
1988	1.74	.16
1989	0.71	.59
1990	0.68	.61
1991	0.50	.74
1992	1.31	.29
1989-1992	0.53	.71
1988-1992	0.73	.58

[1]Varieties differed ($P<0.01$) except 1989; width never differed ($P>0.10$) except 1988.

Stem Branch Morphology of Alfalfa Cultivars

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Most attempts to improve alfalfa forage quality have increased leaf-to-stem ratio (LSR). Yet, increased LSR may not result in improved quality because of leaf losses. Increased stem quality may be a more stable improvement, but little is known about difference in stem morphology. The objective of this research is to determine genetic differences in stem morphology for diverse alfalfa cultivars. Alfalfa genotypes have been divergently selected for the mass ratio of branches to main stems (BMR). Six high- and six low-BMR genotypes were selected from two populations. Half-sib seed was gathered from four divergent genotypes in 1991.

A single-row nursery was established in 1992. The alfalfa entries included: (i) progeny of lines selected for high or low BMR from both '120' and 'Travois'; (ii) the cultivars Travois, 120, 'Arc', 'Green Genes', 'WL 322 HQ', 'Cimmaron VR'; and (iii) lines selected for high (HL) or low (LL) herbage lignin concentration. The experimental design was a randomized complete block with three replicates. Hand harvest of 1.2 m sections of each plot occurred in June and August 1993 and June 1994.

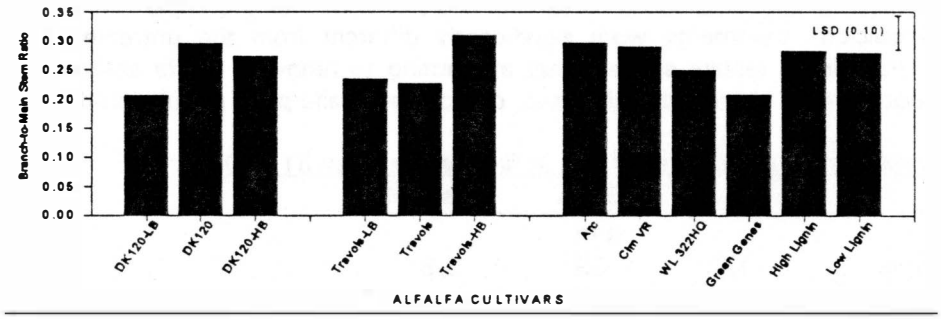

Fig. 1 Branch-to-main stem ratio of twelve alfalfa entries grown in a solid stand. Values are means over three replicates and two harvests in 1993.

Open pollinated progeny from divergent selections for BMR expressed their phenotypic traits (Fig. 1). High-BMR progeny derived from 120 had 29% higher BMR than the low-BMR progeny. For divergent progeny derived from Travois, high-BMR progeny had 35% higher BMR than the low-BMR progeny. Divergent traits were also expressed in June 1994.

Divergent progeny did not differ in concentrations of either neutral detergent fiber (NDF) or acid detergent lignin (ADL) in any of the stem fractions. Also, divergent progeny did not differ from the cultivar entry from which they were derived, except that branches of high-BMR progeny derived from Travois had 16% lower ADL concentration than Travois. There was a positive relationship between BMR and NDF of main stems ($r = 0.83^{**}$), but BMR and total stem NDF were not significantly correlated. There was a positive correlation between BMR and ADL of branches ($r = 0.71$). The NDF and ADL concentrations of total stems were closely related to NDF and ADL of the main stems. Even in high-BMR progeny, little dilution of the main stem occurred by the presence of branches.

Cell wall composition of the total stem fraction is mostly affected by the main stem. The proportion and composition of branches in the total stem fraction had little effect on NDF and ADL concentration of total stems. Concentrations of NDF in the main stem were positively correlated with BMR.

Yield Response to Cultivation of Established Alfalfa

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As alfalfa (*Medicago sativa* L.) stands deteriorate, invading weeds compete with alfalfa and further reduce alfalfa yield. Some growers in the northern Great Plains region atteResearch was initiated near Rapid City, SD in the fall of 1993 to determine the effects of cultivation on weed control, yield, plant density, and longevity of alfalfa stands. mpt to renovate alfalfa with intensive spring cultivation. Cultivation treatments consisted of spike tooth harrow, spring tooth harrow, tandem disk and Triple K harrow. Herbicide treatments included Metribuzin and Pursuit and were applied 10 November 1993. Cultivation treatments were conducted 13 April 1994 when alfalfa was breaking dormancy. Subsamples were hand separated to determine the composition of alfalfa, grasses, and broadleaf weeds. Total herbage yield in May 1994 ranged from 232 g m^{-2} for the spike tooth harrow treatment to 156 g m^{-2} for the Triple K harrow. The proportion of alfalfa, however, from the Triple K harrow was the highest with 860 g kg^{-1}. Herbicide treatments had 600 and 720 g alfalfa kg^{-1} dry matter for Metribuzin and Pursuit , respectively. In August 1994, total herbage yield ranged from 249 g m^{-2} for the spring tooth harrow to 139 g m^{-2} for the tandem disk treatment. Herbicide treatments contained 920 and 790 g alfalfa kg^{-1} dry matter for Metribuzin and Pursuit, respectively. Alfalfa plant population density ranged from 189 plants m^{-2} for the spike tooth harrow to 120 plants m^{-2} for the Triple K harrow. None of the cultivation or herbicide treatments were significantly different from the untreated control for any measurement. These initial results suggest that attempting to renovate alfalfa stands with cultivation practices does not increase yields, control weeds, or increase alfalfa plant population density.

Effects of cultivation and herbicide treatments at first harvest May 31 1994

Treatment	Alfalfa	Grass	Broadleaf
		g m^{-2}	
Spike tooth harrow	183	39	3.8
Spring tooth harrow	136	59	7.8
Disk	114	38	13.3
Triple K harrow	133	12	10.3
Metribuzin	103	65	6.8
Pursuit	127	46	2.5
Control	126	65	0.5

Effects of cultivation and herbicide treatments at second harvest August 1 1994

Treatment	Alfalfa	Grass	Broadleaf	Plant Density
		g m^{-2}		plants m^{-2}
Spike tooth harrow	177	9	17	189
Spring tooth harrow	213	16	19	140
Disk	96	23	19	168
Triple K harrow	192	9	10	119
Metribuzin	168	9	5	139
Pursuit	167	39	5	145
Control	206	25	11	181

Minutes of the 24th Central Alfalfa Improvement Conference

June 18 - 20, 1995
Spearfish, South Dakota

The conference was called to order at 7:59 am on June 19, 1995 by Chair Lanny Rhodes. Those in attendance were welcomed by Kevin Kephart, site host, and David Bryant, Dean of the College of Agricultural and Biological Sciences at South Dakota State University (SDSU). Jim Gerwing from SDSU presented an overview of forage and alfalfa production in South Dakota. Kevin Kephart chaired a session in honor of N.E. Hansen's 100th anniversary. Kevin presented a history of South Dakota, and how that shaped the framework within which Hansen was to later work at SDSU. David (Hansen's Grandson) and Naomi Gilkerson provided a family perspective of Hansen's exploits. The session closed with Ray Moore, former Research Director at SDSU, summarizing Hansen's scientific contributions to SDSU and the USA.

The scientific program commenced at 10:20 am with a contributed paper session. In mid-afternoon a one hour long general discussion was held focusing on issues related to alfalfa variety testing. This was followed by refreshment break when posters were viewed.

The CAIC business meeting was called to order at 4:00 pm by chair Lanny Rhodes. Mark McCaslin moved that the minutes from the 1994 CAIC conference be approved. The motion was seconded by Clive Holland, and passed unanimously. Old business included election of a new vice-chair to replace the position vacated by Cheryl Fox. Thad Busbise was nominated, and was elected by unanimous vote. Mike Peterson was nominated as incoming secretary, and also was elected by unanimous vote. Craig Sheaffer proposed that procedures used for variety testing at various institutions be carefully evaluated, and standardized procedures put in place. This issue is to be discussed at a future CAIC meeting. New business included site selection for future meetings. The 1996 CAIC will be held in conjunction with the North American Alfalfa Improvement Conference June 16 to 20, 1996 in Oklahoma City OK. The site for the 1997 CAIC will LaCrosse WI, and will be co-hosted by Forage Genetics and Cal-West. The date for the 1997 meeting was not determined, but will work around the International Grasslands Congress June 8 to 18 in Canada. Sharie Nygaard and Jessica Brummer presented the following report from the resolutions committee:

"Be it resolved that the participant of the 24th CAIC held at Spearfish, SD on June 18 to 20, 1995 convey their sincere appreciation to: the CAIC Planning Committee for the planning and coordination of these outstanding meetings; the Local Arrangements Chair, Kevin Kephart, and his colleagues and staff; to Lanny Rhodes, Chair of the CAIC, Kevin Kephart, David Bryant, and Jim Gerwing for their opening remarks and welcomes, and to Kevin Kephart, David and Naomi Gilkerson, and Ray Moore for the well-received commemoration of N.E. Hansen's importance to agricultural and horticultural sciences; the contributing scientists for their informative oral and poster presentations communicating the latest in alfalfa and alfalfa improvement. Also to those contributing information, insights, and opinions in the on-going discussion of alfalfa variety testing

issues; and to the staff of the Spearfish Ramada Inn for their hospitality and service." Respectfully submitted, Sharie Nygaard and Jessica Brummer.

Jeff Volenec moved to approve this resolution, which was seconded by Dan Undersander. Jeff Volenec made the motion to adjourn the meeting. This motion was seconded by Kevin Kephart, and passed unanimously. The meeting was adjourned at 5:00 pm.

Respectfully submitted,

J.J. Volenec
Secretary CAIC

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Previous Officers of the Central Alfalfa Improvement Conference

No.	Year	Location	Chair	Vice-Chair	Secretary
1	1947	Manhattan, KS	C.P. Wilsie	C.O. Grandfield	H.O. Graumann
2	1951	Brookings, SD	C.P. Wilsie	C.O. Grandfield	H.O. Graumann
3	1953	Wooster, OH	H.O. Graumann	L.J. Elling	W.C. Adams
4	1955	E. Lansing, MI	L.J. Elling	R.L. Davis	W.C. Adams
5	1957	Fargo, ND	R.L. Davis	M.W. Adams	C.N. Hittle
6	1959	Columbia, MO	M.W. Adams	W.R. Kehr	C.N. Hittle
7	1961	Chicago, IL	W.R. Kehr	C.N. Hittle	F.I. Frosheiser
8	1963	Chicago, IL	C.N. Hittle	F.I. Frosheiser	D. Srnith
9	1965	Chicago, IL	F.I. Frosheiser	D. Smith	E.L. Pinnell
10	1967	Chicago, IL	D. Smith	E.L. Pinnell	K.L. Larson
11	1969	St. Louis, MO	K.L. Larson	K.L. Larson	E. L. Sorensen
12	1971	St. Louis, MO	E.L. Sorensen	M.D. Rumbaugh	J.D. Axtell
13	1973	Kansas City, KS	M.D. Rumbaugh	J.D. Axtell	D.A. Miller
14	1975	Madison, WI	D.A. Miller	D.K. Bames	E.T. Bingham
15	1977	Ames, IA	D.K. Bames	E.T. Bingham	I.T. Carlson
16	1979	St. Paul, MN	E.T. Bingham	I.T. Carlson	M.B. Tesar
17	1981	E. Lansing, MI	I.T. Carlson	M.B. Tesar	J.W. Miller
18	1983	Manhattan, KS	M.B. Tesar	J.W. Miller	D.L. Stuteville
19	1985	Oklahoma City, OK	D.L. Stuteville	J.L. Caddel	M. McCaslin
20	1987	Urbana, IL	J.L. Caddel	M. McCaslin	C.R. Grau
21	1989	Madison, WI	M. McCaslin	C.R. Grau	W.T.W. Woodward
22	1991	Ames, IA	C.R. Grau	W.T.W. Woodward	L.H. Rhodes
23	1993	Lincoln, NE	W.T.W. Woodward	L.H. Rhodes	C.C. Fox
24	1995	Spearfish, SD	L.H. Rhodes	T. H. Busbice	J.J. Volenec