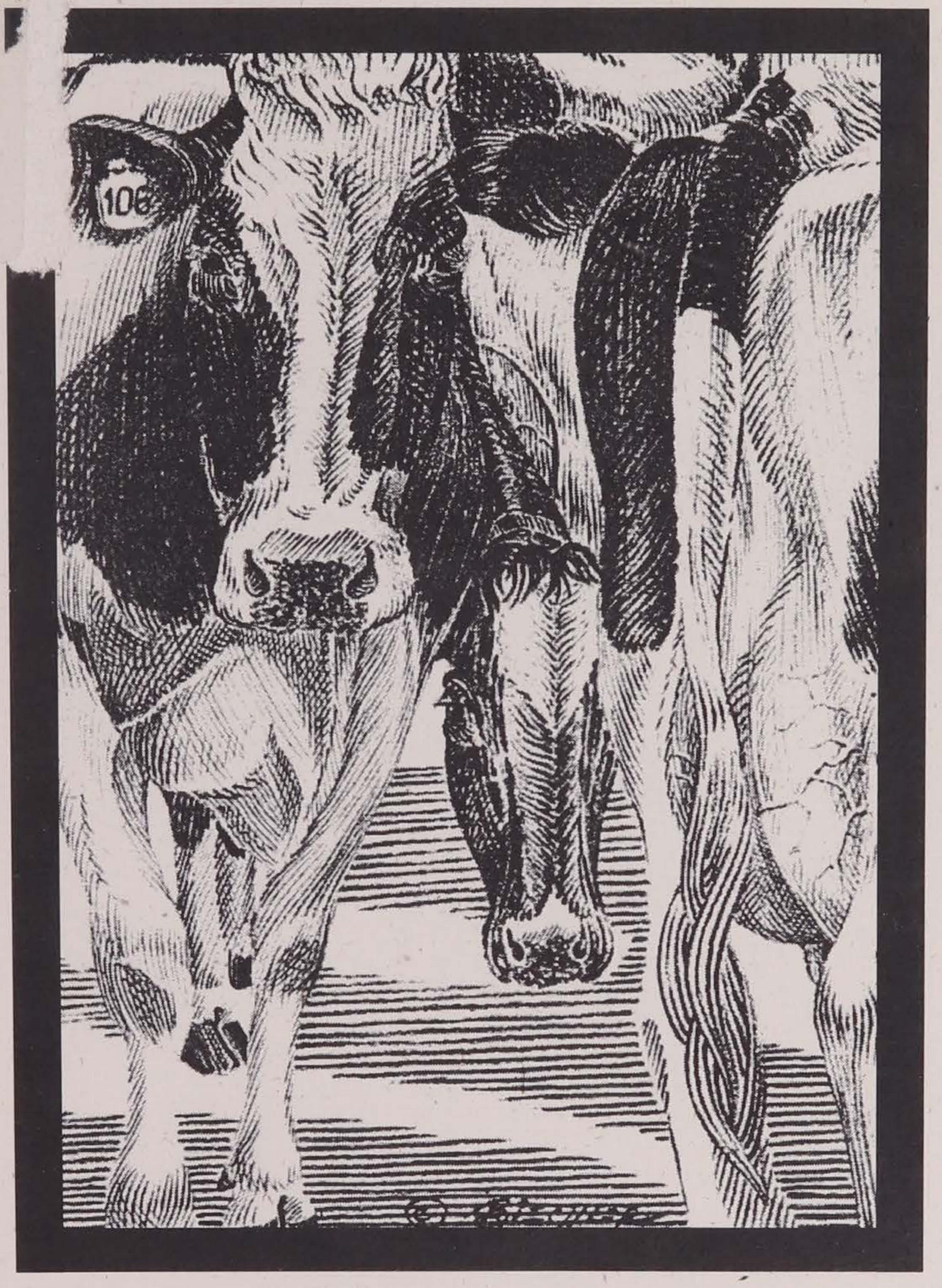


1997 Dairy Report

IOWA STATE UNIVERSITY

University Extension

Iowa State University Dairy Staff

Campus

DAIRY TEACHING
123 Kildee Hall
Fax: (515) 294-0018
M. Douglas Kenealy, (515) 294-6022, dkenealy@iastate.edu
Howard D. Tyler, (515) 294-6434, htyler@iastate.edu
William Wunder, (515) 294-6513, wwunder@iastate.edu

DAIRY FARM RESEARCH STATION SUPT.
Mortensen Road, Ames, IA 50011
Cindy Achen, (515) 292-4731,

AG. RESEARCH STATION SUPT.
801 SW Magazine Road, Ankeny, IA 50021
Fax: (515) 295-2401
Nathan J. Beck, (515) 964-1343

DAIRY EXTENSION
123 Kildee Hall
Phone: (515) 294-2116, Fax: (515) 294-2393
Lee Kilmer, x1kilmer@exnet.iastate.edu
Leo Timms, x1timms@exnet.iastate.edu
Marj Faust, x1faust@esnet.iastate.edu

DAIRY PRODUCTS
Food Sciences Building, Fax: (515) 294-8181
E.G. Hammond, professor, (515) 294-5799
hammond@iastate.edu
B.A. Glatz, professor, (515) 294-3970, bglatz@iastate.edu
William LaGrange, professor, (515) 294-3156
foodsci@iastate.edu

ANIMAL BREEDING
239 Kildee Hall, Fax: (515) 294-2401
A.E. Freeman, professor, (515) 294-3352
Charles F. Curtiss Distinguished Professor in Agriculture
genef@iastate.edu
P.J. Berger, professor, (515) 294-3435, pjberger@iastate.edu
David H. Kelley, livestock research specialist, (515) 294-3810
dhkelley@iastate.edu

NUTRITIONAL PHYSIOLOGY
313 Kildee Hall, Fax: (515) 294-6445
Jerry W. Young, professor, (515) 294-5889,
jwyoung@iastate.edu
Donald C. Beitz, distinguished professor, (515) 294-5626
dcbeitz@iastate.edu
Gary L. Lindberg, assistant professor, (515) 294-5759
gll@iastate.edu

COLLEGE OF VETERINARY MEDICINE
Dr. Nolan Hartwig, (515) 294-8790, nhartwig@iastate.edu
Dr. Mark Kirkpatrick (515) 294-7905, kirmar@iastate.edu

NATIONAL ANIMAL DISEASE CENTER
USDA, ARS, National Animal Disease Center
PO Box 70, 2300 Dayton Ave., Ames, IA 50010-0070
Fax: (515) 239-8458
Dr. Ronald L. Horst, (515) 239-8312
Dr. Jesse P. Goff, (515) 239-8578
Dr. James A. Harp, (515) 239-8533
Dr. Marcus E. Kehrli, Jr., (515) 239-8462
Dr. Brian J. Nonnecke, (515) 239-8311
Dr. Timothy A. Reinhardt, (515) 239-8200

Field

DAIRY FIELD SPECIALISTS
Dale Thoreson, livestock, (319) 882-4275
Bremer Co. Extension Office
Box 49, 100 1/2 1st N.W.
Tripoli, IA 50676
Tony Harvey, livestock, (319) 583-6496
Dubuque Co. Extension Office
2600 Dodge St. Plaza 20
Dubuque, IA 52003
Paul Brown, farm management, (515) 394-2174
Chickasaw Co. Extension Office, 104 E. Main
New Hampton, IA 50659
Brian Lang, crops, (319) 382-2949
Winneshiek Co. Extension Office, 911 S. Mill
Decorah, IA 52101
Dan Meyer, agricultural engineer, (319) 425-3331
Fayette Co. Extension Office, 201 E. Clark
Fayette, IA 52142

LIVESTOCK SPECIALISTS
Dennis DeWitt, (712) 336-3488
Dickinson Co. Extension Office
1610A 18th St.
Spirit Lake, IA 51360
Denise Schwab, (515) 484-2703
Tama Co. Extension Office
PO Box 308, USDA Building
203 W. High Street
Toledo, IA 52342
Darrell Busby, (712) 482-6449
E. Pottawattamie Extension Office, 120 N. Main
Oakland, IA 51560
Joe Sellers, (515) 774-2016
Lucas Co. Extension Office, R.R. 5, Box 91
Chariton, IA 50049
Beth Doran, (712) 472-2576
Lyon Co. Extension Office, 301 1/2 1st. St. #202
Rock Rapids, IA 51246
Gary Fike, (712) 263-4697
Crawford Co. Extension Office, 35 S. Main
Denison, IA 51442
Education Director.
Ida Co. Extension Office, 211 Main Street, PO Box 167
Ida Grove, IA 51445
Russ BreDahl, (515) 782-8426
Union Co. Extension Office, 105 W. Adams Suite B
Creston, IA 50801
Ron Irvin, (319) 398-1272
ISU Outreach Center, Box 2068
Cedar Rapids, IA 52406
Byron Leu, (515) 472-4166
Jefferson Co. Extension Office,
Box 445, 2606 W. Burlington
Fairfield, IA 52556
Russ Euken, (515) 648-4850
Hardin Co. Extension Office, 524 Lawler St.
Iowa Falls, IA 50126
Carl Neifert, (515) 462-1001
Madison Co. Extension Office
117 N. 1st Street
Winterset, IA 50273
Roger Brummett, (712) 523-2137
Taylor Co. Extension Office, 312 Main Street
Bedford, IA 50833

Table of Contents
1997 Dairy Report

Welcome to the 1997 Iowa State University Dairy Report

We are pleased to present this summary of the dairy teaching, research, and extension programs in the College of Agriculture, College of Veterinary Medicine, and College of Family and Consumer Sciences, and collaboration from researchers at the National Animal Disease Laboratory. This report is a multidisciplinary effort and represents the wide-ranging efforts of the team of faculty, staff, and students who work to enhance the opportunities for the dairy industry in Iowa.

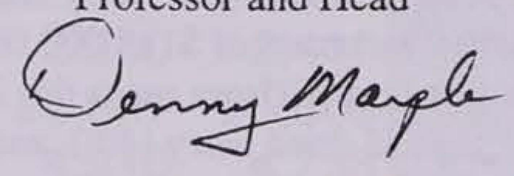
Just as change is taking place in the dairy industry, there has been change in the dairy programs on campus. The construction project in Kildee Hall has resulted in the dairy extension group moving from room 4 to other locations in the building. Likewise, we have experienced the sounds of progress as jack hammers have transformed areas in Kildee Hall into modern classrooms and laboratories to support both teaching and research programs. The old Meat Laboratory is undergoing renovation to become a modern classroom and teaching amphitheater that will allow students to gain hands-on experience with livestock as well as provide an opportunity for us to host youth events on campus that involve livestock activities.

The shell of the new addition to Kildee Hall is nearly complete and the entire project is expected to be complete by September 1998. The new addition will provide space to conduct metabolism studies on cattle, including lactating dairy cattle, to address the nutrient requirements of high producing cows. It also will provide a modern meeting area to address the changing needs of a rapidly changing livestock industry. Laboratories will be constructed to address questions of animal behavior and physiology of reproduction, and to develop new ways to use molecular genetics to further improve selection strategies. Another major component of the new laboratory space will be new facilities to address issues of food safety through use of irradiation of foods.

The dairy group in Animal Science is in the process of developing a long range plan to identify needs and goals of the dairy program in the department. This is important as we try to anticipate the needs of the dairy industry and our students, and will include recommendations for hiring future faculty members as well as identifying needs for new facilities. Discussions continue on strategies to combine the two dairy herds at a modern production facility. Our goal is to construct a new dairy farm to address questions in nutrition and management of lactating cows and heifer calves from birth to production and to provide data to enhance selection methods. Input from producers will be important to the success of this planning process, and we look forward to these efforts.

The dairy industry in Iowa is experiencing many of the changes observed in other livestock industries. As new issues arise, please know we are here to provide assistance so that the dairy industry can continue to be an important part of the economic structure of Iowa. We hope you find the information in this report useful, and we encourage you to call on us if we may be of service to you and your programs.

Sincerely,
Dennis N. Marple
Professor and Head



The Iowa Dairy Industry

Lee H. Kilmer, professor
Department of Animal Science

DSL-106

The Iowa dairy industry, like much of American agriculture, is undergoing a major transition. Dairy cow numbers continue to decline (Table 1) since peaking around 1950. Certain parts of the country, however, have increased cow numbers in recent years. Consequently, traditional dairy states in the upper Midwest have lost dairy business to these expanding states. The decline of dairy cow numbers during the 1990s has been less in Iowa than in neighboring states. Overall, U.S. production of milk has continued to increase at a modest rate (Table 2). Total milk production in Iowa has remained around 4 billion pounds throughout the 1990s, while average production per cow has remained around 15,000 lb (Table 3). Currently, Iowa ranks 11th in both total number of dairy cows and in total milk production. Although the number of dairy cows has declined, the number of dairy farms has declined even faster (Table 4). Iowa has lost approximately 10% of its dairy herds annually since 1990.

Dairying still represents a significant component of Iowa agriculture, with the sale of milk, cull cows, and calves contributing more than $561,000,000 to the economy of the state. This amounts to a $1.4 million daily cash flow contribution for raw milk supplies alone.

Dairy is a value added industry in that dairy cattle annually consume more than 37 million bushels of corn, 2.5 million bushels of soybeans, 1.2 millions tons of hay, and 2.25 million tons of corn silage. Many Iowa producers also have cropping enterprises, growing their own forages and, in many cases, grains as well. In addition, most producers also have other livestock, usually hogs. The corn grain and soybeans that are fed to dairy animals are worth a combined total in excess of $115,000,000. Processing of milk into cheese (Iowa ranks 6th in the nation), dry milk (5th) and dried whey (4th), and marketing of these products adds to the economy of Iowa as well.

Dairy is more labor intensive than other agricultural enterprises, providing productive work for 10,000 people on-farms and 3,600 in hauling and processing plants. Total annual economic contribution of dairy, including the value of labor, support services and materials, to the Iowa economy is in excess of $1.5 billion. A recent study done by individuals at the New York State Department of Labor and Cornell University showed economic multipliers of 2.29 for dairy farming and 2.61 for dairy manufacturing, whereas the highest non-farm economic multiplier (construction industry) was 1.66. Thus, every dollar spent by a dairy farm generates $2.29 in additional economic activity within the local community.

Due to the heavy geographic concentration in northeast Iowa (Table 5), dairy represents the primary economic force in that portion of Iowa. However, the industry is often overlooked within Iowa because of swine, beef, corn, and soybeans. Livestock generates 60% and cash crops 40% of Iowa's agricultural income, with hogs accounting for approximately 50% of the gross livestock dollars, beef 40%, and dairy 10%.

Herds participating in the Dairy Herd Improvement Association (DHIA) production records program can serve as a barometer for monitoring production changes. The number of herds making significant improvements in their average production per cow has increased during the past several years. The top DHIA herd continues to have nearly double the production per cow compared with the average Iowa cow.

The Iowa dairy industry has and continues to change, but its importance to the state is still significant. Dairying is both labor intensive and capital intensive, and thus is able to use many of Iowa's abundant resources. Finally, given the concerns about soil conservation, expansion of the dairy industry can provide a marketing (value added) outlet for forages, which can be grown on the more highly erodable soils in Iowa.

Table 1. Number of cows in Iowa and selected states.

	1990	1991	1992	1993	1994	1995	1996	% of 1990
				thousand head				
Illinois	174	171	169	167	165	163	154	88.5
Iowa	**280**	**275**	**268**	**264**	**265**	**251**	**250**	**89.3**
Minnesota	710	681	653	635	609	599	598	84.2
Missouri	223	213	208	209	197	190	179	80.3
Nebraska	97	92	88	82	77	74	69	71.1
South Dakota	140	136	130	125	120	118	112	80.0
Wisconsin	1,731	1,681	1,618	1,543	1,494	1,490	1,449	83.7
Arizona	69	95	98	102	116	114	120	173.9
California	1,135	1,155	1,180	1,210	1,235	1,254	1,264	111.4
Idaho	179	178	183	189	208	232	256	143.0
New Mexico	81	98	111	136	165	191	195	240.7
Oregon	99	100	102	100	100	97	93	93.9
Texas	386	386	386	394	402	401	398	103.1
Washington	237	237	249	257	261	266	264	111.4
U.S.Total	9,993	9,826	9,688	9,589	9,525	9,461	9,351	93.6

Table 2. Pounds of milk marketed in Iowa and selected states.

	1990	1991	1992	1993	1994	1995	1996	% of 1990
				million pounds				
Illinois	2,559	2,554	2,525	2,553	2,549	2,545	2,335	91.2
Iowa	**4,233**	**4,151**	**4,006**	**4,054**	**3,962**	**4,038**	**3,826**	**90.4**
Minnesota	10,030	9,775	9,858	9,693	9,342	9,442	9,440	94.1
Missouri	3,040	2,865	2,971	2,840	2,720	2,690	2,440	80.3
Nebraska	1,345	1,280	1,230	1,125	1,110	1,095	1,050	78.1
South Dakota	1,716	1,674	1,660	1,619	1,589	1,591	1,474	85.9
Wisconsin	24,187	23,770	23,844	22,844	22,412	22,942	22,376	92.5
Arizona	1,645	1,713	1,787	1,877	2,134	2,230	2,410	146.5
California	20,947	21,407	22,092	22,927	25,019	25,327	25,859	123.4
Idaho	2,949	2,919	3,138	3,229	3,754	4,210	4,735	160.6
New Mexico	1,524	1,917	2,174	2,621	3,325	3,623	3,748	245.9
Oregon	1,611	1,659	1,712	1,692	1,714	1,677	1,608	99.8
Texas	5,539	5,418	5,590	5,910	6,225	6,113	6,120	110.5
Washington	4,392	4,459	4,836	4,980	5,203	5,302	5,279	120.2
U.S. Total	147,721	147,697	150,885	150,582	153,622	155,644	154,268	104.4

Table 3. Average milk per cow for Iowa and selected states.

	1990	1991	1992	1993	1994	1995	1996	% of 1990
Illinois	14,707	14,936	14,941	15,287	15,448	14,857	15,162	3.1
Iowa	**15,118**	**15,095**	**14,948**	**15,356**	**14,951**	**16,135**	**15,304**	**1.2**
Minnesota	14,127	14,354	15,096	15,265	15,340	15,708	15,786	11.7
Missouri	13,632	13,451	14,284	13,589	13,807	14,158	13,631	0.0
Nebraska	13,866	13,913	13,977	13,720	14,416	14,797	15,217	9.7
South Dakota	12,257	12,309	12,769	12,952	13,242	13,398	13,161	7.4
Wisconsin	13,973	14,140	14,737	14,805	15,001	15,397	15,442	10.5
Arizona	17,500	18,032	18,235	18,402	18,397	19,561	20,083	14.8
California	18,456	18,534	18,722	18,948	20,258	20,211	20,458	10.8
Idaho	16,475	16,399	17,148	17,085	18,048	18,147	18,496	12.3
New Mexico	18,815	19,561	19,586	19,272	20,152	18,969	19,221	2.2
Oregon	16,273	16,590	16,784	16,920	17,140	17,289	17,290	6.2
Texas	14,350	14,036	14,482	15,000	15,485	15,244	15,377	7.2
Washington	18,532	18,814	19,422	19,377	19,935	19,932	19,996	7.9
U.S. Total	14,782	15,031	15,574	15,704	16,128	16,433	16,498	11.6

Table 4. Number of dairy farms in Iowa and selected states.

	1990[a]	1991[a]	1992[a]	1993[a]	1994[b]	1995[b]	1996[b]	% of 1990
Illinois	3,700	3,000	3,000	2,800	2,322	2,171	2,027	54.8
Iowa	**7,800**	**7,000**	**6,600**	**5,600**	**4,754**	**4,469**	**4,390**	**56.3**
Minnesota	15,500	15,000	14,000	13,500	12,626	11,817	10,970	70.8
Missouri	7,000	6,900	6,800	7,500	3,539	3,377	3,308	47.3
Nebraska	3,000	2,700	2,500	2,900	1,187	1,078	995	33.2
South Dakota	3,600	3,300	3,000	2,800	1,916	1,724	1,523	42.3
Wisconsin	34,000	33,000	32,000	30,000	28,323	26,887	25,526	75.1
Arizona	500	500	500	500	105	135	139	27.8
California	4,500	4,200	4,200	4,200	2,426	2,383	2,178	48.4
Idaho	2,200	1,900	1,900	1,700	1,179	1,156	1,111	50.5
New Mexico	1,200	1,300	1,200	1,100	154	151	151	12.6
Oregon	2,100	1,900	1,500	1,500	553	522	500	23.8
Texas	5,700	5,300	5,300	5,000	1,960	1,880	1,667	29.2
Washington	3,000	3,000	3,000	3,000	1,043	962	919	30.6
U.S. Total	192,660	180,640	170,500	162,450	117,732	111,932	106,045	55.0

[a]Data from Census of Agriculture and Crop Reporting Service (estimated numbers).
[b]Data from Farm Bureau and various state Department of Agriculture agencies (actual number of commercial farms).

Table 5. Number of Grade A and Grade B milk producers in Iowa, October 1997.

County	Grade A	Grade B	Total
Adair	8	2	10
Adams	3	0	3
Allamakee	214	21	235
Appanoose	6	6	12
Audubon	3	2	5
Benton	26	5	31
Black Hawk	14	4	18
Boone	5	0	5
Bremer	102	18	120
Buchanan	49	134	183
Buena Vista	3	1	4
Butler	32	17	49
Calhoun	0	0	0
Carroll	4	3	7
Cass	6	1	7
Cedar	17	4	21
Cerro Gordo	4	4	8
Cherokee	11	5	16
Chickasaw	72	12	84
Clarke	2	3	5
Clay	3	2	5
Clayton	330	66	396
Clinton	26	3	29
Crawford	4	2	6
Dallas	3	0	3
Davis	9	46	55
Decatur	5	0	5
Delaware	230	17	247
Des Moines	5	2	7
Dickenson	2	1	3
Dubuque	474	27	501
Emmet	1	0	1
Fayette	189	22	211
Floyd	15	4	19
Franklin	5	4	9
Freemont	3	0	3
Greene	0	0	0
Grundy	3	1	4
Guthrie	5	1	6
Hamilton	2	0	2
Hancock	2	2	4
Hardin	16	1	17
Harrison	0	2	2
Henry	8	2	10
Howard	80	25	105
Humboldt	4	0	4
Ida	4	0	4
Iowa	11	5	16
Jackson	86	20	106
Jasper	16	1	17
Jefferson	6	1	7
Johnson	9	3	12
Jones	44	12	56
Keokuk	1	3	4

County	Grade A	Grade B	Total
Keokuk	1	3	4
Kossuth	12	5	17
Lee	16	1	17
Linn	35	8	43
Louisa	1	0	1
Lucas	2	7	9
Lyon	44	12	56
Madison	1	0	1
Mahaska	17	2	19
Marion	12	4	16
Marshall	8	0	8
Mills	1	0	1
Mitchell	53	47	100
Monona	0	2	2
Monroe	8	1	9
Montgomery	3	0	3
Muscatine	13	0	13
O'Brien	15	2	17
Osceola	18	4	22
Page	1	0	1
Palo Alto	6	4	10
Plymouth	17	4	21
Pocohontas	5	3	8
Polk	5	0	5
Pottawattamie	1	3	4
Poweshiek	5	2	7
Ringgold	0	2	2
Sac	9	2	11
Scott	19	2	21
Shelby	5	1	6
Sioux	89	14	103
Story	5	0	5
Tama	13	5	18
Taylor	3	0	3
Union	1	0	1
Van Buren	12	24	36
Wapello	2	2	4
Warren	9	1	10
Washington	41	67	108
Wayne	6	3	9
Webster	4	0	4
Winnebago	6	7	13
Winneshiek	291	35	326
Woodbury	3	1	4
Worth	6	2	8
Wright	2	2	4
Total for Iowa	3,005	791	3,796

Iowa's Dairy Foods Processing Industry

W. S. LaGrange, professor of food science and human nutrition

DSL-107

Iowa's dairy foods processing industry contributes significantly to Iowa's economy both from the standpoint of numbers of employees, nearly 3,500, and in the value of dairy foods manufactured and marketed, nearly $2 billion annually. In 1995, the latest data available, farm cash receipts for milk sold to dairy foods plants totaled $400.2 million, representing 5% of total farm receipts in Iowa.

In 1997 there are six fluid milk plants, including four that manufacture cultured dairy foods such as cottage cheese and yogurts. There are four plants that manufacture ice cream and other frozen desserts; seven plants that manufacture cured type cheeses; 11 plants that are involved in drying skim milk, whey, and whey by-products; and one small creamery that makes butter for their local trade.

USDA data indicate that Iowa's seven cured cheese manufacturing plants made and marketed a total of 241 million pounds of cheese in 1995. This quantity ranked Iowa 7th nationally in 1995. These cheeses included several popular types such as blue, Swiss, cream, mozzarella, and American that includes cheddar, colby, Monterey/Jack. Cottage cheese production in Iowa in 1995 totaled over 12 million pounds made by three Iowa fluid milk and cultured-products plants. Nonfat dry milk powder production ranks Iowa high nationally each year and 4th in 1995 as four plants manufacture the majority of the nearly 55 million pounds produced. Iowa dairy processors also rank within the top 10 states in the production of frozen desserts (four plants) and dried whey products (five plants) such as lactose, and whey protein concentrate.

Iowa's dairy foods plants rank high nationally not only in quality and quantity of dairy foods manufactured but also in income from sales. The July 1997 *Dairy Foods* ranked the top 60 companies in the United States by dairy food sales. Wells' Dairy, Inc., LeMars, Iowa ranked number 17 in the nation with total dairy sales of $500 million in 1996. They manufacture and market fluid milk products, including culture products and frozen desserts and many frozen novelty dairy items. They market not only throughout the United States but also in several other countries, including Canada, Mexico, Japan, and Russia. Because Wells' Dairy's two large frozen dessert plants combined manufacture more frozen desserts than any other in the world, LeMars has been named the Ice Cream Capital of the World.

Dairy Foods also ranked Anderson Erickson Dairy Company of Des Moines 53rd nationally in 1996 with sales valued at $127 million. Their fluid milk plant also manufactures several cultured products and their ice cream plant produces packaged ice creams.

Among dairy food cooperatives, *Dairy Foods* placed Swiss Valley Farms Company 17th nationally in sales of finished dairy products with $222 million in 1996. This cooperative has five plants, including a cultured products plant in Cedar Rapids, a fluid milk and juice plant in Dubuque, a cheese (Swiss, cream Neufchatel, baby Swiss) plant in Luana, and a dried milk and cream plant in Maquoketa.

A 1997 issue of *Dairy Field* presented a listing of the top 100 dairy foods organizations in the United States by sales dollars. Included on this list was Wells' Dairy Incorporated ranked number 218 nationally, Swiss Valley Farms Company ranked 48, and Anderson Erickson Dairy Company 84. Other nationally ranked dairy foods plants that own manufacturing plants in Iowa include number seven Associated Milk Producers Incorporated with a nonfat dry milk plant in Arlington and Sibley, an instant dry milk plant in Mason City and a cheddar cheese plant in Sanforn. Foremost Farms USA, ranked nationally number 11 with headquarters in Baraboo, Wisconsin with a whey protein concentrate plant in Cresco, a cured cheese and whey processing plant in Decorah, and a whey protein concentrate plant in Waukon. Number 15 Borden/Meadow Gold Dairies Incorporated with 27 plants in the United States and headquarters in Ogden, Utah, has a frozen ice cream plant in Des Moines and a frozen novelty plant in Perry. Nestle USA of Glendale, California, ranked 29th in the United States in dollar sales has a large dry milk products plant, for instant beverages, in Waverly. Roberts Dairy's fluid milk plants in Des Moines and Iowa City are the result of joint ventures between number 4 ranked Mid-American Dairymen Incorporated, Spring- field, Missouri, and number 9 Prairie Farms Dairy Incorporated of Carlinville, Illinois.

The Iowa dairy industry is technically well positioned to move successfully into the 21st century. Foreign markets will continued to provide opportunities for growth in marketing Iowa's high-quality dairy foods.

Executive Summary from Dairy Teaching Group

M. Douglas Kenealy, professor in charge,
dairy science, Iowa State University

DSL-108

The dairy science curriculum at Iowa State University is found within the Department of Animal Science. The current dairy science teaching team includes:

M. Douglas Kenealy, professor in charge
William W. Wunder, professor
Howard Tyler, associate professor
Cindy Achen, dairy farm superintendent
Ilene Carlson, secretary

Enrollment

Fall 1997 undergraduate enrollment figures were:

Dairy Science majors	**57**
Animal Science majors	597
General Pre-Vet students	55
Total, department	709
Total, College of Agriculture	2,812

Note: Majors in Animal or Dairy Science may declare pre-veterinary medicine, but the department also manages the undeclared pre-veterinary medicine program (General Pre-Vet).

Graduate student enrollment for fall 1997 was approximately 100, with 85 funded students pursuing Master's or Ph.D. degrees. Ten postdoctoral fellows also were employed by the department.

Curriculum Update

The 1996 executive summary described the background for curricular changes developed for the 1997-1999 catalog biennium. Those changes for Animal Science and Dairy Science curricula were based upon "outcomes." In brief, the faculty spent 18 months in an organized process to determine the skills necessary for graduates to succeed in the career markets of the early 21st century. These skills were translated into outcomes statements, and outcomes were grouped into categories that eventually became courses. An example of an outcome statement is: "After completion of this curriculum/course a student will be able to: *critically evaluate a long-range breeding program for a dairy producer.*"

The ultimate goal was to create curricula that would prepare graduates to be competitive in the immediate post-graduation job market as well as give them skills to meet the needs of evolving career markets.

The immediate result of the curriculum revitalization process was the catalog materials and course requirements that became effective for new students in fall semester 1997. One measure of the enthusiasm of faculty and students for the new Animal Science and Dairy Science core requirements was the move of upper-class students to move from their old catalog requirements and adopt the new requirements. The ultimate test will be the success of graduates in placement and career development.

Placement

Placement rate and salary offerings for 1997 continued to show strength for the livestock industry. Placement rate for bachelor's graduates was between 80 and 90% for 1997. Starting salary average for graduates was approximately $26,000.

Strongest areas of placement for 1997 were feed and livestock product sales; food industry, including quality assurance; and livestock management. Other significant areas of opportunity were animal health industry, breed associations, bull studs, and promotion/public relations.

Dairy Science Club

The Dairy Science Club was successful in competition to host the Midwest regional conference for the 1998 American Dairy Science Association Student Affiliate. The ADSA conference will be held in February 1998. Hosting the conference will be a prestigious event for the club, but has involved extensive planning activity and considerable financial obligation.

Dairy Science Club members hope to bring over 400 dairy science students to ISU. Student affiliates come from colleges and universities throughout the upper Midwest, ranging geographically from Michigan State University to Kansas State University. The ISU Dairy Science club's extensive list of successful, ongoing activities and solid planning from their leaders helped to seal the bid for this opportunity. The ISU faculty continue to be impressed with the quality of leadership displayed by Dairy Science majors.

Building Update

Progress on the Kildee addition and associated remodeling is slow but sure. Remodeling of several facilities in Kildee is significant, not just new desks and chairs. The remodeling includes two completely new Iowa Communications Network (ICN or fiber optics) capable classrooms on the first floor. These classrooms will replace the administrative office complex on the west side of Kildee.

Two existing classrooms will be remodeled into a multimedia, classroom support center for teaching and student drop-in use. Remaining classroom facilities on the first floor of Kildee Hall will receive minor remodeling. Two new "wet laboratory" spaces will be added to Kildee basement: a physiology laboratory and a nutrition laboratory.

One of the impacts and opportunities of remodeling was the chance to review use of all spaces in Kildee. A result was at least temporary officing for dairy extension faculty. Room 123 Kildee, the home of the Dairy Science curriculum since the building of Kildee in 1964, also has become home for a portion of the Dairy Science extension team, specifically Drs. Marjorie Faust and Leo Timms, and Ms. Ardella Krull. Dairy Science extension leader Dr. Lee Kilmer is temporarily housed with Animal Science extension. This grouping has been a positive for group interaction for faculty in 123 Kildee. Post-remodeling plans may allow the option of combined housing for all dairy extension and teaching group members.

Remodeling is to be completed in phases, but final work should parallel completion of the Kildee addition. The twostory, 74,000 square foot addition is to be completed in fall of 1998. The addition will house facilities for research, teaching and outreach for the animal sciences. The construction will incorporate 19,000 square feet of remodeled spaces for teaching in the old meat laboratory, including a small pavilion area. The facility will include a short-term holding area for livestock used in on-campus teaching.

Summary

For the Dairy Science program, calendar year 1997 started and finished as a period of dramatic change, especially in curriculum and in the physical surroundings of Kildee Hall. Change brings with it opportunity. Students and faculty have embraced these opportunities as vehicles to improve their position in the educational and career markets for the turn of the century.

Iowa State University Teaching and Research Farm: Management Practices and Research, Teaching, and Outreach Activities

Cindy Achen, superintendent, ISU Dairy Farm

DSL-109

Introduction

The Iowa State University (ISU) Dairy Teaching and Research Farm at Ames, Iowa, is used by dairy faculty to conduct research that will benefit dairy farmers and the related industry here in Iowa and the Midwest. The ISU Dairy Farm also provides work experience for students, a laboratory for Animal Science and College of Veterinary Medicine courses, and a location for dairy extension faculty to conduct outreach programs for 4-H and other dairy interest groups.

General Farm Management

Cows at the ISU Dairy Farm are milked three times daily in a single eight herringbone style parlor by using automatic takeoffs. Production by breed is summarized in Table 1.

Tie stalls for 110 cows house the early lactation cows, and late lactation cows are housed in a 48-cow, free-stall barn. Cows are fed according to three groups: first lactation; high mature Holstein; and a late lactation group. The forage portion of the total mixed rations (TMRs) consist of corn silage, alfalfa haylage, and chopped dry hay. Concentrate feeds used include whole roasted soybeans, high moisture shell corn, wet corn gluten feed, whole cottonseed, and a mineral/vitamin mix.

This past year included some needed farm improvements. We began by rebuilding fences to help give the farm a neater appearance. Also, the classroom, heifer barn, silo shed, goat barn, and main barn were painted. The feed mangers in the stall barn were also cleaned and coated with an epoxy liner to facilitate cleanup and give the cows a bunk free of rotting debris that collects in pitted concrete.

Work has begun on developing a total farm protocol handbook for cow and calf ailments and a whole herd recordkeeping system. These developments will help with research projects as well as a system to analyze all farm procedures.

Research Activities

Research activities at ISU are diverse; animals are used in projects by staff and students in the Animal Science Department, National Animal Disease Center, and the College of Veterinary Medicine. Many of these projects are summarized in separate articles in this publication. These projects include investigating dry cow nutrition, barrier teat dips, Mycoplassma bovis research, and fresh cow metabolic problems.

Teaching Activities

A variety of courses taught in the Animal Science Department and the College of Veterinary Medicine meet at the ISU Dairy Farm. These courses use the animals and facilities in different teaching situations for students. Specific dairy course work includes: dairy cattle performance, dairy cattle selection, intercollegiate judging training, and competition, and dairy enterprise planning. Other animal science courses are more general for all species but the the farm and dairy animals serve a very important role in broad training of all animal science students.

Table 1. ISU rolling herd averages, September 1997.

Breed	Producing Females	Standardized 150-day milk	Milk	Fat	% Fat	Protein	% Protein
Ayrshire	14	78.6	16,896	711	4.5	538	3.3
Brown Swiss	16	76.0	19,415	845	4.6	645	3.6
Guernsey	6	57.3	16,966	848	6.1	574	3.9
Holstein	112	96.6	25,768	1001	3.8	767	3.2
Jersey	29	67.5	17,283	852	4.8	611	3.6
M. Shorthorn	9	66.4	17,299	722	4.3	551	3.3

The ISU Dairy Farm provides part-time labor and training for 20 to 25 student employees each year. These students vary in background, and train to perform and carry out much of the needed labor to operate the farm.

The College of Veterinary Medicine also uses animals and facilities at the ISU Dairy Farm. Senior students visit the farm with the attending veterinarian when herd health is done and also receive instruction on production medicine and public health.

Outreach

The ISU Dairy Farm has the opportunity to give a positive image of dairying to the hundreds of school-age children and families from the city of Ames who come to visit the dairy farm each year. Major effort has been made to make the premises neat and clean to convey this positive image. The dairy farm also has hosted tours from the former Soviet Union, South America, and Japan.

Several 4-H activities are conducted at the farm each year as well. In June the Animal Science 4-H Roundup was held with a 2-day dairy section conducted at the farm. The annual youth dairy judging contest conducted by the ISU Dairy Science Club was held this past fall, and the "I Milked A Cow" event was successful for the club at VEISHEA this past spring.

This past summer a very successful Share-a-Heifer program was conducted with 14 Story County 4-H'ers. This program gives young people the opportunity to care for an animal and exhibit at the local fair. Plans are being made so that the project can be expanded to include more project meetings to enable these young people to learn more about the dairy industry and the total life cycle of the dairy cow.

The Future

The ISU Dairy Farm continues to promote the dairy industry with emphasis on education and outreach. The dairy industry, as every other production animal industry, is rapidly evolving. The ISU Dairy Teaching and Research Farm must position itself to provide adequate research to assist producers with questions on new management and feeding strategies.

Acknowledgments

The teaching and outreach programs at the ISU Dairy Farm would not be possible without a dedicated and qualified full-time staff. These people are: Paul Amundson, assistant manager; John Kent, animal caretaker II; Tom Golden, animal caretaker II; and Ron Nyman and Jennifer Sickles, milkers.

A New Dairy Nutrition Course

G. L. Lindberg, assistant professor; and
R. Orth,[a] professor
Department of Animal Science
Iowa State University

DSL-110

Summary

A dairy nutrition short course was offered during January 3-5, 1997, at Iowa State University, Ames. The course featured lecture and laboratory topics and was attended by 36 students from Iowa, Wisconsin, North Dakota, Illinois, Michigan, and Minnesota. The enrollees consisted of undergraduate students, postgraduate students, and dairy-related industry personnel. Plans have been made to offer a similar course with expanded enrollment by ICN (Iowa Communications Network) among three sites in Iowa during January 5-7, 1998.

Introduction

Iowa State University is one of several land grant universities in the upper Midwest that is located in a state with significant numbers of dairy cattle and that offers an undergraduate major in Dairy Science. Specialized courses in dairy-related subjects, even within large universities, are often taught infrequently because only limited numbers of students usually express interest in specialized courses. If specialized courses are taught at times that are feasible and convenient to students, then courses with specialized content may attract significant numbers of students from other universities and from allied industries. Our goal was to initiate a specialized dairy nutrition short course in 1997 that would attract enrollees from Iowa and from locations outside Iowa, and to determine interest by students in such a course.

Course Description and Organization

The nutrition course was organized as a 3-day series of lectures and laboratories that was held on consecutive days in January 1997. Lecture topics were diverse and generally given by faculty with special expertise in the respective area (Table 1). Laboratory sections consisted of computer laboratories and farm laboratories and included computer diet formulation, body condition scoring, and particle size evaluation. The course was advertised by notices sent to area extension offices and to extension offices in neighboring states. Ten faculty members accepted the invitation to participate in lectures or laboratories, and each delivered one or more lectures in their area of expertise (Table 1).

Table 1 Faculty and topics for the dairy nutrition course.

Faculty Member	Topic
R. Orth	Summary of dairy industry
G. Lindberg	Principles of dairy nutrition
W. Mahanna	Ensiled forages
H. Tyler	Calf and heifer nutrition
J. Goff	Milk fever/anion balance
J. Young	Ketosis and fatty liver
K. Nelson	Nutritional management
W. Wunder	Financial management
D. Beitz	Fat supplementation
J. Schroeder	Protein nutrition

Results

Thirty-six students from Iowa, Wisconsin, North Dakota, Illinois, Michigan, and Minnesota enrolled for the course. Attendees included approximately equal numbers of undergraduate students, veterinary and other professional students, and dairy industry personnel. The course was of considerable interest to attendees for two major reasons: (1) the subject matter was of interest to them in their business or educational goals, and (2) the course was available for their attendance because it was held during a short period between semesters when industry personnel, professional students, and undergraduate students from other institutions could attend.

Future Plans

Because of the considerable interest generated, the course will be repeated in Spring semester of 1998 on January 5-7. To fulfill the demand for attendance, the 1998 course consists of lectures to be held on three consecutive mornings at three sites: ISU at Ames, Northeast Iowa Community College (NICC) at Calmar, and Dordt College at Sioux Center. In addition, because all three sites have dairy herds and computing facilities on campus, laboratory sections will be held simultaneously at all three sites on three consecutive afternoons. The rationale for using three sites is: (1) more space for laboratory sections can be generated because of the availability of cattle and

[a] Current address: Iowa Institute for Cooperatives, Ames, Iowa.

computer space at the multiple sites; (2) the distribution of sites will decrease travel time and housing costs for many of the participants; and (3) more participants as lecturers in the course can become involved because area extension specialists and faculty at the other colleges can originate contributing lectures and demonstrations from any of the three sites.

Specialized Dairy Education at the ISU College of Veterinary Medicine

Mark A. Kirkpatrick, food animal specialist, Veterinary Diagnostic & Production Animal Medicine; and Leo Timms, extension dairy specialist, animal science department

DSL-111

The College of Veterinary Medicine is offering a specialized course for students interested in food animal production medicine. It consists of 2-week blocks with emphasis on dairy, beef, swine, and small ruminant topics. The 2-week dairy series is coordinated by Dr. Leo Timms and Dr. Mark Kirkpatrick. Previous versions of this course have been offered in the winter. This year the placement was advanced to late July and August in an effort to train veterinary students in production medicine topics prior to their outside preceptorships in practices throughout Iowa and the Midwest. The initial feedback on this placement has been good and will be repeated next year.

The emphasis of this course is to teach the principles of production and herd medicine, rather than individual animal case management. With this in mind, the course was broken down into topical segments that were taught by both experts within, and outside ISU, as shown below.

A successful part of previous dairy courses was the use of field trips to the dairy areas of Iowa. We elected to repeat this valuable resource. We would like to extend our thanks to the following dairies that opened their doors and operations to us. In Northeast Iowa, these include Mr. and Mrs. Terry Eick of Plainfield, Jon and Jeri Kerns of Oelwein, and Dick Blough of Waterloo. In Northeast Iowa, we visited the Maasen Bros., Hoogland, VanVeldheuizen and Terry Van Maanen herds. These trips provided the opportunity for the students to visualize the principles of feeding, milking, building, and waste management in different scenarios, locations, and conditions.

In addition to the classes mentioned above, pharmaceutical industry representatives were invited to introduce their products to the students. These representatives provided an hour lecture on the science and use of these tools in veterinary medicine. Most of these representatives also were veterinarians that offered additional insight into future professional career choices. This segment was popular with the students and will be expanded next year where possible.

A final objective of this class is to expose students to as many dairy production medicine practitioners as possible, not only from the subject matter, diagnostic, and trouble-shooting areas but also from the lifestyle perspective. How do I: (1) find time to fit this in; (2) find a practice that will allow this; (3) convince my future partners, as well as my family, about the time commitments). These are equally, and sometimes, more important issues facing young practitioners.

Student reviews following the course were very favorable and had good suggestions that we would like to incorporate next year. These included requests for more case studies so students can follow the thought processes that go into diagnosing herd problems. We also would like to add more computer lab time to provide hands-on experience with software tools of the trade such as: the Cornell Univ. Interactive Body Condition Scoring CD, Dairy Comp 305, PC Dart, Spartan Ration Analyzer, and the National Dairy Database. We want to ensure that our students have some concept of what these tools will do for them and their clients, and be exposed to the explosion of information that is available to them through a computer and modem.

We look forward to being a continuing resource for our students that have progressed through this program and are excited about working to improve the next offering.

Day 1	Ron Orth	Expansion Group	DHIA records, Dairy Expansion Strategies
Day 2	Mark Kirkpatrick DVM	ISU College of Veterinary Medicine	Dairy Computer Records Introduction
Day 2	Mark Kirkpatrick DVM	ISU College of Veterinary Medicine	Basics of Dairy Operations
Day 3	Mark Kirkpatrick DVM	ISU College of Veterinary Medicine	Northeast Iowa Dairy Field Trip
Day 4	Lee Kilmer Ph.D.	ISU Dairy Extension	Applied Nutrition
Day 4	Dan Meyer Ph.D.	ISU Extension -- Fayette Iowa	Facilities/Waste Management
Day 5	Lee Kilmer Ph.D.	ISU Dairy Extension	Applied Nutrition
Day 5	Marj Faust Ph.D.	ISU Dairy Extension	Genetics and Breeding Program
Day 6	Steve Bolin DVM Ph.D.	National Animal Disease Center	BVD Virus and Today's Dairy
Day 6	Mark Kirkpatrick DVM	ISU College of Veterinary Medicine	Johnes Disease and Control Measures
Day 6	Marj Faust Ph.D.	ISU Dairy Extension	Reproduction and Culling
Day 7	Veterinary Practitioners		Veterinary Roundtable
Day 8	Ed Kreykes DVM	Private Practice Sanborn, Iowa	Northwest Iowa Dairy Field Trip
Day 9	Leo Timms Ph.D.	ISU Dairy Extension	Milk Quality/Milking Machines
Day 10	Leo Timms Ph.D.	ISU Dairy Extension	Reproduction Strategies
Day 10	Mark Kirkpatrick DVM	ISU College of Veterinary Medicine	Student Consultation Reports on Field Trips

Executive Summary of Dairy Extension Programs

Lee H. Kilmer, professor
Department of Animal Science

DSL-112

Dairy Science Extension serves a diverse clientele that range from dairy producers and their families to milk processing-plant field staff, from veterinarians to financial lenders, and from youth to representatives from agribusinesses. The primary objective of Dairy Science Extension is to provide research-based information that enables clientele to make informed management decisions. This mission of education and transfer of technology for clients is accomplished through a variety of formats, including one-on-one contacts, meetings, workshops, magazine articles, and other publications. The variety of opportunities provided by Dairy Science Extension for client education is almost as diverse as the clientele.

Table 1. Current ISU Extension dairy team.

State Specialists
- M. A. Faust, Breeding and Genetics
- N. R. Hartwig, Ruminant Veterinarian
- L. H. Kilmer, Nutrition and Management
- L. L. Timms, Milk Quality and Reproduction

Agricultural Field Specialists
- P. W. Brown, Farm Management
- T. J. Harvey, Dairy/Beef & Forage
- B. J. Lang, Row Crops & Forages
- D. J. Meyer, Agricultural Engineering
- D. R. Thoreson, Dairy/Beef & Forage

ISU Extension Specialists for Related Areas
- B. A. Berna, Family Life
- E. J. Spurlock, Family Resource Management
- M. S. Holz-Clause, Industry - Value Added Ag.
- W. S. LaGrange, Food Science
- S. M. Scholl, Community Development Spec
- M. R. Willett, Manufacturing Specialist

Dairy Team

Historically, state extension specialists in dairy science and veterinary medicine have worked closely as the "dairy team" to develop cooperative educational programs for dairy producers in Iowa. However, during 1992, Iowa State University Extension (ISUE) was reorganized and "field specialist" positions were created to provide a local source of subject matter expertise and education for clients in multi-county areas. Dairy Science Extension seized the opportunity provided by the reorganization of ISUE to develop a broader-based agricultural "dairy team" (Table 1) for planning, developing, and implementing educational programs. The "dairy team" was developed to serve clients in northeast Iowa primarily, because two-thirds of the state's dairy cows are located in this area. Programs developed by the dairy team serve as a model for dairy programs offered in other parts of Iowa.

The dairy-team approach continues to evolve towards a goal of developing a truly integrated approach for meeting clientele needs. Current interdisciplinary efforts are targeted at developing a series of "Strategic Advantage" workshops that will help producers identify industry trends, the strengths and limitations of their farm business, and then develop a strategic management business plan that will help them focus their efforts and be competitive in the future.

Education Programs for Iowa Clients

Primary delivery of extension education continues to be through extension-sponsored meetings and one-on-one contacts (telephone calls and face-to-face visitation); dairy team members conduct more than 500 individual farm visits annually to assist producers in identifying and solving problems that are limiting production, efficiency, and profitability of their farms. For the majority of farms visited, factors limiting efficiency, productivity, and profitability most are the quality of milk produced and the facilities where animals are housed. Nutrition and other management issues are principal limitations for the remainder of farms visited. Visits to farms usually include ISUE state and field specialists, agribusiness personnel, and ISU students, and, as a result, these visits serve as a unique opportunity to provide hands-on education for clientele. Recently, there has been a dramatic increase in the number of individuals and groups requesting ISUE assistance for planning herd expansion, remodeling existing facilities, or establishing a new dairy operation. Dairy team members have worked with over 20 operations to provide assistance in cash flow and budget projections, and the technical aspects of design construction. In addition, an "*ad hoc* Dairy Task Force" was created in cooperation with industry and governmental agencies to coordinate expansion efforts and promote Iowa as a good place to establish a dairy operation.

Currently, agricultural businesses are increasing the services and level of support that they provide to dairy producers, consequently, these service providers are rapidly becoming a prime audience for dairy extension programs. Service providers are able to contact individual herd owners on a regular basis more frequently than are extension personnel, thus service providers can serve to "multiply" the transfer of research-based information from extension to

producers. Two extension programs that have been particularly successful for providing research-based information to service providers are ag-service provider meetings and the Professional Dairy Management Seminar. The ag-service provider meetings were developed so that a diverse group of lenders, veterinarians, nutritionists, sanitarians and artificial insemination (A.I.) personnel could meet together and share ideas on methods for working together to improve profitability for our dairy producer clientele. The 2-day long Professional Dairy Management Seminar is a statewide educational seminar for producers and service providers that has been so successful that it now includes educators and clients from Illinois, Minnesota, and Wisconsin. Other cooperative programs include meetings cosponsored with veterinary clinics, and barn meetings that involve A.I. organizations and dairy breed associations. Topics offered in 1997 included effective fiber, protein feeding, mastitis, milk quality, reproduction, was dairy facilities, labor saving tips, and updates on winter teat lesions, dry period mastitis, and synchronized ovulation. A total of 26 meetings was held in cooperation with 20 different veterinary clinics in northeast Iowa. These partnerships between ISUE and others in the Iowa dairy industry are mutually beneficial, because resources and responsibilities for teaching and audience recruitment can be shared.

Use of production and financial records continues to be vital for success of dairy enterprises, because development of an effective plan for the future requires knowledge of previous and current status for the herd. Extension staff members work closely with DHIA to provide clients with information and decision-aids packages that are useful for herd decision making. Extension personnel, including both state and field specialists, assist with education programs for field technicians and conduct workshops designed to educate producers on use of records to improve the productivity and profitability of their dairy herds by informed management decision making.

In addition, extension staff members work closely with numerous dairy-affiliated organizations such as the Iowa Dairy Products Association, Iowa Purebred Dairy Cattle Council, Dairy Lab Services, various state dairy breed organizations, and the Iowa Farm Bureau.

A comprehensive list of extension education programs for dairy clients is beyond the scope of this report; however, some of the more recent programs have focused on herd expansion, grazing, production efficiency, financial analysis, global markets for dairy genetics, two-generation asset transfer, and decision making.

Cooperative Multistate Education Programs

The four upper midwestern states of Iowa, Illinois, Minnesota, and Wisconsin have a long-standing tradition for meeting annually to share ideas and plan programs. The planning group consists primarily of state and field extension dairy specialists, and has included veterinarians, agricultural engineers, agronomists, and farm management specialists at various times during recent years. Cooperation among dairy extension specialists for these four states evolved because the management styles for dairy operations in these four states were similar, and as a result, educational needs for clients also were similar.

Initially, several jointly authored publications were developed; these included a feeding and nutrition bulletin and two 4-H dairy youth project books. Evidence for the success of this venture may be illustrated best by noting that 22 other states have adopted the 4-H dairy project materials. The first cooperative program was a nutrition and management seminar that was presented once in each state on consecutive days in 1989. A comparable program has been offered almost annually since then and consists of four presentations, each given by an extension staff member from one of the cooperating states. Iowa producers benefit by use of this truly cooperative approach, because, with no significant cost increase to them or to Dairy Science Extension, producers are able to see and hear new ideas and gain access to specialists from other states. Four cooperative programs were planned and presented during the past year: a 2-day expansion workshop that was offered in St. Paul, MN and in Dubuque; a 2-day Personnel Management Workshop and a 2-day Applied Nutrition Conference in LaCrosse, WI; and five 1-day seminars at various locations in the four-state region.

National Involvement

The ISUE staff has been and continues to be active on national committees and projects. Dr. Faust is the past-president of the Mid-West section of the American Dairy Science Association. Other involvement has included active membership for the ADSA Extension and Education Program committee, ES-USDA National Dairy Quality Assurance committee, the ES-USDA National Reproduction Workshop committee, the National Dairy Quality Assurance Program implementation steering committee, and the National Mastitis Council executive committee.

Other Responsibilities

Extension staff are involved in other roles within the department and University as well. ISU specialists frequently serve as guest lecturers in various classes and are able to bring real-world experiences into the classroom. Dr. L. L. Timms was codeveloper of a production medicine elective block for senior veterinary students (VCS 413) and for Lactation Biology (AnS 437). Extension specialists have served as guest lecturers in AnS 434, AnS 436, AnS 519, and in other courses.

From a research perspective, extension specialists are uniquely positioned to identify real needs for applied and basic research; consequently, three staff are members of regional research committees. Dr. Timms contributes to the project NE 112 Resistance to Mastitis, and Drs. Faust and Kilmer contribute to NC 119 Dairy Herd Management

Strategies for Improved Decision Making and Profitability. Specific research thrusts include applying total quality management (TQM) approaches on dairy farms to improve milk and beef quality, determining whether milk urea nitrogen is a useful indicator of dietary protein adequacy, developing new compounds as teat dips for cows as they are dried off, evaluating the efficacy of a commercial electronic heat detection system, and developing methods for evaluating the profitability of dairy herds by using DHIA herd performance measures.

Summary

Dairy Science Extension at Iowa State University plays an integral role in transferring information and technology to dairy clientele, and in identifying research needs for Iowa's dairy industry. As such, extension is the primary link between Iowa's people involved directly with dairy production and processing and those that teach and conduct research on the Iowa State University campus.

Rightsizing in the Iowa Dairy Industry

M. A. Faust, associate professor
of Animal Science

DSL-113

Introduction

The dairy industry in Iowa is experiencing a great deal of change lately, and one yardstick of change is annual data showing an increasing trend for larger herds. The recent increase in herdsize may be attributed partially to advantages that Iowa has over other states for an economic climate that is "agriculture friendly," and favorable processor capacity, feed prices, and milk prices. More importantly, however, changes within the industry are occurring because farm families are assessing critically their farming enterprises, and are identifying their "core" business. Farm families that identify dairying as a core enterprise then consider their long-term business goals, the influence of factors beyond the farm gate, and available and potential resources for their dairy. As a result, many Iowa farm families are making conscious decisions to "rightsize" their dairy operation. For some operations, rightsizing has meant eliminating the dairy; other operations have rightsized by maintaining their current herdsize, and by improving their efficiency and profitability. During the last few years, rightsizing for many dairy operations in Iowa has meant combining of herds and adding cows. Objectives for the Iowa State University Dairy Extension Team have been to develop and conduct workshops, seminars, conferences, and individual consultations for Iowa dairy families and those who service these dairy operations as they proceed through this series of decisions.

Strategic Advantage Dairy Workshops

During 1996-1997, the Dairy Team conducted a series of 3-day workshops entitled, "Strategic Advantage Dairy Workshops." Approximately 40 Iowa dairy farm units attended the dairy related workshops that were held in Sioux Center, Dubuque, and Decorah, Iowa. Workshop participants considered personal, family, farm-related, and external factors that will influence their operations in the future. Also, they set individual, family, and dairy related goals that were used to determine future plans and strategies that they can implement and which will gain a competitive advantage for the operation.

Expansion Conferences and Workshops

A 2-day expansion workshop was held in Dubuque as part of the cooperative 4-State Dairy Extension programs; focus for this workshop was "Growing Dairy Profitability through Strategic Growth." More than 120 dairy producers and representatives from agribusiness participated in workshop sessions that included discussions about facilities, manure and waste handling, managing financial risk, incorporating grazing in the expansion process, herd biosecurity, and business ownership and financing options. For 1998, the 4-State Dairy Extension group is planning expansion conferences that will be held in Madison, Wisconsin on March 31-April 1 and New Ulm, Minnesota on April 1-2.

Dairy Expansion Tours

More than 80 gained an in-depth view of dairy expansions by participating in the 2 tours that were conducted by the Iowa State University Extension Dairy Team. One tour visited dairies in northwestern Iowa, eastern South Dakota, and western Minnesota, and the second one toured dairy operations in eastern Wisconsin. Herdsize for the dairies visited ranged from 150 to 2,500 cows, thus participants were able to view and discuss moderate- to large-scale dairy operations. For many who have participated, these tours have provided an excellent educational experience; since the tours, some participants have begun expansions for own dairies, and others have concluded that an expansion would not be beneficial for their operation currently.

Dairy Personnel Management Conference

When evaluating strengths and weaknesses for their dairy operations currently, dairy herd owners indicate frequently that they need to develop additional skills for managing personnel. Iowa State University Extension as part of the 4-State Dairy Extension group planned and conducted the 1997 4-State Personnel Management Conference. This conference was held in LaCrosse, Wisconsin, and included educational presentations and case studies on designing employee positions, structure and organization of farm personnel, training for employees, and improving communications. The 140 attendees for this year's conference received an extensive notebook of materials that can serve as a future reference.

Iowa Dairy Task Force

Iowa State University Extension has been an active participant in the industry coalition, the Iowa Dairy Task Force. Other Task Force members include representatives from Iowa Farm Bureau, Iowa Area Development Group L. C., Northwest Iowa Power Cooperative (NIPCO), Farm Credit Services, the Iowa Department of Agriculture and Land Stewardship - Agricultural Marketing and Dairy

Products Inspection Bureaus, Iowa Institute of Coops, Iowa Bankers Association, and the Iowa Dairy Products Association. Many of the Task Force members have worked with dairies during expansion projects, and as a result, members are aware of several obstacles for growth within the Iowa dairy industry. These obstacles are discussed during Task Force meetings, and strategies for resolving these are defined.

To date, the group has developed materials that can be used for discussing benefits to communities and dairy herd owners for growing Iowa's dairy industry, and for locating and expanding dairies in the state. Several Task Force members traveled to California and evaluated the implications of large-scale dairies. Also, the coalition has met with representatives from the Iowa Rural Water Association and AgConnect, an organization that aids current farm owners to transition farms to prospective new farmers.

The Iowa Dairy Products Association, in conjunction with the Iowa Dairy Task Force supported an Iowa Dairy Industry Summit that was held in Waterloo. More than 130 community and rural economic development leaders attended this summit to increase awareness and discuss the benefits to local economies from a viable local dairy industry.

ISU Workshop Course - Dairy Facilities

For two consecutive years, the dairy teaching and extension groups at ISU cooperated to offer a special workshop course on dairy facilities and equipment, AnS 493M Dairy Facility and Systems Planning. Forty professionals in the dairy industry, and upper-class veterinary and undergraduate students from ISU and other universities earned ISU course credits for the 1997 workshop, and to date, more than 50 have registered for the January 1998 workshop. Course participants have traveled from Michigan, North Dakota, and Oklahoma to attend this 1-credit workshop. The 1997 workshop was offered on the ISU campus only, and, the 1998 workshop is being offered at ISU and through the ICN in Sioux Center and Calmar/Cresco. Topics discussed include planning for the milking facility, building design and costs, ventilation, and manure handling and storage.

Local and Community Events

Local and community groups inquire frequently about business enterprises that will stimulate local economies, and members of the ISU Extension Dairy Team have conducted several different educational activities about dairying and the dairy industry. Field representatives from a dairy cooperative attended one seminar to discuss issues for dairy expansion. Realtors attended a second seminar entitled, "Why Dairy in the Upper Midwest." ISU Extension cooperated with dairy producers to host two open house tours. These dairy producers had incorporated unique waste handling processes and equipment when they had expanded their herds. One herd owner used a flush system in the freestall barns, and incorporated a solid/liquid manure separator to reduce the amount of solids in the two-stage lagoon. The other herd owner composted solids from the new solid/liquid manure separator, cornstalks, and leaves and paper from a local recycling center; the composted material was used as bedding in the freestall barn. In addition, Dairy Team members have discussed economic implications for communities of dairy enterprises at numerous meetings, seminars, and workshops that were sponsored by economic development and value-added agriculture committees.

Strategic Advantage: Executive Summary

Paul Brown, ISU extension field specialist,
Farm Management

DSL-114

In 1994, ISUE field staff in north central and northeast Iowa began to respond to a growing concern among farm families regarding the long-term profitability and competitiveness of their businesses. Two questions were raised frequently by farm families: (1) How do we take what is happening in agriculture and make profitable and competitive changes in our business for long-term success? (2) How can we continue in agriculture and maintain an acceptable quality of life? Several strategic management workshops were held for swine and dairy producers and their families during 1995 and 1996. During this same time period, ISUE informal and formal needs assessment activities confirm growing farm family concern over the future direction of their businesses.

Through the Strategic Advantage initiative, ISUE will focus on helping farm managers and their families: (1) learn and apply the principles of strategic management to their farm businesses, (2) identify and evaluate several long-term strategies to increase the profitability and competitiveness of their farm businesses and the well-being of their families, and (3) identify and acquire skills to develop and implement ongoing strategic management in their farm businesses and families.

ISUE is in a very strong position to take leadership and use our unique strengths in developing and delivering this program. ISUE has the following critical elements to offer: (1) an inter-disciplinary staff to consider both the farm business and the farm family; (2) the research, teaching, and outreach expertise to meld together management and strategy with the realities of agriculture technology and production practices; and (3) the field staff and resources to provide farm families with follow-up and support to implement new strategies.

During July and August 1996, the Strategic Advantage committee began the process to formalize the concept of strategic management for farm families. A review of existing literature and teaching materials was conducted. Progressive farm families were interviewed and asked to provide input on program delivery methods, learning styles, curriculum development, and strategic management concepts. A subcommittee, called the Program Team, was appointed to develop a program and provide leadership. The Program Team consisted of ISUE staff in economics, farm management, family life, livestock, and communications.

A proposal was prepared asking state commodity groups, farm organizations, agricultural lending organizations and the farm media to join ISUE in developing the Strategic Advantage program. Committee co-chairs conducted personal visits with the leadership in each organization. Each committing organization was asked to name two members and their spouses to serve on a Design Team.

Design Team members meet in Ames on December 3, 10, and 17 with a two-fold purpose. First, to provide input in the development of teaching materials, teaching outline, and workshop format. Their input was critical and would play a key role in marketing the workshop series as "designed by producers for producers." Second, the producer members of the Design Team would actually work through or test the first draft of the teaching materials using the proposed workshop format.

Forty-two dairy operations were represented at Strategic Advantage dairy workshops held in Dubuque, Decorah, and Sioux Center. The Iowa Dairy Products Association served as a co-sponsor. Relationships were forged with these participants that will last a life-time. An interactive teaching approach was used with numerous hands-on activities. Discussions and work activities were very engaging. The farm families came wanting a better sense of business direction and left having taken specific action to develop their own strategies for long-term success. Whether farm families were an FSA borrower with 50 cows or had recently expanded their business to 500 cows, everyone gained from this educational opportunity. Field Specialists are currently providing follow-up assistance with workshop participants.

Twenty-nine pork operations were represented at Strategic Advantage swine workshops held in Iowa City, Iowa Falls, and Griswold. The Iowa Pork Producers Association served as a co-sponsor. Program Team members participated in a teleconference with the IPPA on March 10 to evaluate the workshops and suggest future changes.

The Program Team and first year teaching teams met on April 3 for a debriefing session. Suggestions were made to perfect teaching materials and teaching methodology.

Field Specialists made the commitment to conduct at least two workshop series in each farm management area with 15 to 17 operations per workshop. A state-wide target of reaching 450 to 600 farm businesses was established for the 1997-1998 program year.

A two-hour Strategic Advantage training session for all ISUE staff was broadcast over the ICN on June 9. Fifteen receiving sites were connected with over 125 staff participating.

Teaching materials were finalized in July and August. Top priority in design was to develop materials that would exhibit a high degree of professionalism and shed a positive light on ISUE. New and innovative packaging and printing techniques were incorporated. A "for Staff Only" web site was constructed on the ISUE web page for the Strategic Advantage Project in August.

An oak-frame display was designed, printed, and made available to each farm management specialist. Second, a promotional brochure and acrylic-frame display poster were printed and distributed throughout the system.

A six month follow-up survey was conducted involving the participants who attended the dairy and swine pilot workshops.

Six teaching team training sessions were conducted during August, September, and October. A total of twelve teams were trained, and they are prepared to deliver workshops across the state during the 1997-1998 program year.

Thirty Strategic Advantage workshops have been scheduled across the state for the 1997-1998 winter meeting season. A state-wide marketing and awareness campaign is expected to begin in October.

Strategic Advantage: Comprehensive Summary

Paul Brown, ISU extension field specialist, Farm Management

DSL-115

In 1994, ISUE field staff in north central and northeast Iowa began to respond to a growing concern among farm families regarding the long-term profitability and competitiveness of their businesses. During the winter meeting seasons of 1995 and 1996, several strategic management workshops were held for swine and dairy producers and their families respectively.

During this same time period, ISUE informal and formal needs assessment activities confirm growing farm family concern over the future direction of their businesses. Two questions were raised frequently by farm families: (1) How do we take what is happening in agriculture and make profitable and competitive changes in our business for long-term success? (2) How can we continue in agriculture and maintain an acceptable quality of life? These are strategic or long-term issues that are best addressed using strategic management concepts.

In March 1996, a cross section of state staff, field specialists, and CEEDs; program directors; and area directors met to establish ISUE long-term program initiatives. For agriculture, strategic management was one of those initiatives. Statewide, ISUE will focus on helping farm managers and their families: (1) learn and apply the principles of strategic management to their farm businesses, (2) identify and evaluate several long-term strategies to increase the profitability and competitiveness of their farm businesses and the well-being of their families, and (3) identify and acquire skills to develop and implement ongoing strategic management in their farm businesses and families.

ISUE is in a very strong position to take leadership and use our unique strengths in developing and delivering this program. ISUE has the following critical elements to offer: (1) an inter-disciplinary staff to consider both the farm business and the farm family; (2) the research, teaching, and outreach expertise to meld together management and strategy with the realities of agriculture technology and production practices; and (3) the field staff and resources to provide farm families with follow-up and support to implement new strategies.

In May and June 1996, the strategic management initiative co-chairs and committee were named.

Getting Started:

During July and August 1996, the strategic management committee began the process to formalize the concept of strategic management for farm families. A review of existing literature and teaching materials was conducted. Progressive farm families were interviewed and asked to provide input on program delivery methods, learning styles, curriculum development, and strategic management concepts. The program name "Strategic Advantage" was selected by the committee.

A proposal was prepared asking state commodity groups, farm organizations, agricultural lending organizations and the farm media to join ISUE in developing the Strategic Advantage program. Committee co-chairs conducted personal visits with the leadership in each organization. Each committing organization was asked to name two members and their spouses to serve on a Design Team. Industry support for this effort has been strong.

Design Team members included two producer members and their spouses from the following organizations: Iowa Pork Producers Association, Iowa Dairy Products Association, Iowa Cattlemen's Association, Iowa Corn Growers Association, Iowa Soybean Association, and Iowa Farm Bureau Federation. Management personnel from these organizations as well as from the Iowa Banker's Association and Farm Credit System also participated. The Design Team was formed in early September and held an introductory meeting on September 27, 1996.

A subcommittee, called the Program Team, was appointed to develop a program and provide leadership. The Program Team consisted of ISUE staff in economics, farm management, family life, livestock, and communications. A professional editor and designer eventually joined the team.

Developing the Curriculum and Program:

During October and November, a first draft of the workshop teaching and activity materials were developed by the Program Team. An earlier search for resources and materials proved to be unfruitful. The strategic management model developed by ISUE for production agriculture served as an outline.

A proposed teaching outline was prepared by the Program Team and sent to Design Team members for their input and review.

Three Strategic Advantage dairy workshops were scheduled in January and February at Dubuque, Decorah, and Sioux Center. In late November, a formal brochure was designed and sent to the printer. The Iowa Dairy Products Association would serve as a cosponsor.

Design Team members meet in Ames on December 3, 10, and 17 with a two-fold purpose. First, to provide input in the development of teaching materials, teaching outline, and workshop format. Their input was critical and would play a key role in marketing the workshop series as "designed by producers for producers." Second, the producer members of the Design Team would actually work through or test the

first draft of the teaching materials using the proposed workshop format. Actually testing the materials in this manner provided the Program Team valuable insight before attempting to take the materials and program to the field.

Anecdotal Evidence: Even though producer members of the Design Team had to switch hats often, the discussion and experience was highly beneficial to those attending. Based on their Design Team experience, several members were able to make some major life-changing decisions. Two months after the Design Team activity one individual decided to discontinue farrowing pigs on his farm. He joined a neighborhood sow coop and will finish a higher volume of SEW pigs in the future. Another decided not to move their large dairy operation to a less concentrated area. Instead, they decided to expand their existing land base.

On December 18, 1996, the Program Team incorporated Design Team suggestions and recommendations into the second draft of materials and modified the workshop format accordingly. Other lessons learned from the experience of the Program Team were also incorporated.

Three Strategic Advantage swine workshops were scheduled in February and March in Iowa City, Iowa Falls, and Griswold. In late December, a formal brochure was designed and sent to the printer. The Iowa Pork Producers Association would serve as a cosponsor.

State Strategic Advantage co-chairs worked with the editor and designer to finalize the second draft materials in late December. Printing occurred in early January. Design Team recommendations were to give ISUE materials a new look. Teaching materials arrived at the Chickasaw County Extension Office the day before the first day of the dairy workshop

Piloting the Program:

1. A marketing and recruitment packet was prepared for field specialists and county staff involved in recruiting. Materials included recruitment and marketing instructions, sample client letter, and two news releases. This packet was sent to the appropriate staff at least one month in advance. Brochures for the dairy workshops were provided four weeks before the first event. Due to a printing snafu, the swine workshop brochures were not available until two and a half weeks before the first event.

In late December, an awareness campaign was begun with Wallaces Farmer. A case study project also was begun using actual case histories of families making strategic changes in their businesses. Side-bar commentary is being provided by Program Team and Design Team members. A total of six cases were published January through March.

Recruited and trained 42 dairy producers and their families at three dairy workshops. Relationships were forged with these participants that will last a life-time. An interactive teaching approach was used with numerous hands-on activities. Discussions and work activities were very engaging. The farm families came wanting a better sense of business direction and left having taken specific action to develop their own strategies for long-term success. Whether farm families were an FSA borrower with 50 cows or had recently expanded their business to 500 cows, everyone gained from this educational opportunity. A follow-up telephone call to each of the dairy participants was made 7 to 10 days after the workshop series ended.

Examples of action taken or changes proposed:

Several families decided to expand their dairy operation.

After session one of the workshop series, one two-generation family had their first formal family meeting which lasted 3 hours.

Several other two-generation families had their first formal family meeting and said the workshop materials raised important questions.

One dairy family decided to have someone else custom raise their dairy replacements, so they could concentrate on milking more cows.

Another dairy family decided to custom raise dairy heifers for others. This strategy would require new facilities which would later be used to expand the dairy operation.

One couple, approaching retirement, who had just experienced a failed two-generation farming arrangement decided to work with Extension staff to do it right next time.

Several families were planning to begin a business succession strategy.

One young couple was planning to grow their dairy operation on a small acreage and purchase most of their feedstuffs.

Several young couples planned to purchase a land base for their operations.

One couple currently renting their farm wanted to enroll in the Farm On program as a beginning farmer applicant.

Due to urban encroachment, two brothers were going to move their dairy to another part of the state.

Another family plans to meet once a month to discuss farm succession and future business direction.

During a family meeting a 10 year old said they wanted to make a difference in their community, so this family decided to make family time a higher priority in their lives.

One dairy manager wanted to improve personnel management skills.

One couple who were 15 years from retirement and had no children interested in coming back to the farm wanted to see their business continue. They plan to enroll as a landowner in the Farm On program.

One young couple plans to purchase the building site from his parents.

Management intensive grazing and seasonal calving would provide another family with more time away from the farm.

One dairy manager wanted to develop and utilize a farm accounting system to make better financial decisions.

One young couple decided to maintain their 50-60 cow operation, but expand and promote their commercial breeding stock enterprise.

One young couple decided that dairying was not in their long-term future.

5. Recruited and trained 29 pork producers and their families at three swine workshops.
6. For the dairy and swine workshops, different end-of-meeting evaluations were utilized because the group dynamics were quite different. The dairy families were recruited by ISUE staff and chose to attend freely. The participants for the swine workshops were primarily recruited by the Iowa Pork Producers Association. Most were part of an elite evaluation group who were involved in critiquing various level of the Pork College. Therefore, these individuals came primarily to evaluate rather than fully participate. With this in mind, the individual impact and end-of-meeting evaluations were quite different than the dairy workshops. Program Team members participated in a teleconference with the IPPA on March 10 to evaluate the workshops and suggest future changes.
7. The Program Team and first year teaching teams met on April 3 for a debriefing session. Suggestions were made to perfect teaching materials and teaching methodology. Assignments were made to add materials for beef and cash grain producers.

Field Specialists teams began individual follow-up consultations with participants beginning after the workshops ended and continued through the summer. The purpose of these visits was to assist families analyze their strategic alternatives and begin implementation of the chosen alternative.

Preparing for Year One:

Farm Management and Marketing Staff reviewed and discussed the Strategic Advantage project at spring in-service training. Field Specialists made the commitment to conduct at least two workshop series in each farm management area with 15 to 17 operations per workshop. A state-wide target of reaching 450 to 600 farm businesses was established for the 1997-1998 program year. Field Specialists agreed to provide leadership in their respective counties for the Strategic Advantage project.
Responsibilities included: (1) establish local teaching teams consisting of farm management, family life, and appropriate livestock/crop specialists; (2) decide upon the enterprise focus of each workshop; (3) schedule one day for a local or regional training session for all teaching team members; (4) schedule at least two Strategic Advantage workshops within local area; and (5) develop a local marketing and recruitment plan with CEEDs to supplement state-wide effort.

A two-hour Strategic Advantage training session for all ISUE staff was broadcast over the ICN on June 9. Fifteen receiving sites were connected with over 125 staff participating. The objectives of the training were to review pilot year accomplishments, future direction of the project, elements of strategy, workshop format, and marketing and recruitment plans.

In late June, the co-chairs met with the editor and designer to finalize teaching materials. Top priority in design was to develop materials that would exhibit a high degree of professionalism and shed a positive light on ISUE. New and innovative packaging and printing techniques were incorporated. Materials are expected to be printed in October.

Through the summer months several promotional items were prepared and distributed. First, an oak-frame display was designed, printed, and made available to each farm management specialist. Second, a promotional brochure and acrylic-frame display poster were printed and distributed throughout the system.

In August, Strategic Advantage is designated as a formal ISUE project under the new programming system. It is the first program to be designated as formal project coming from the field.

A six month follow-up survey was conducted involving the participants who attended the dairy and swine pilot workshops.

A "for Staff Only" web site was constructed on the ISUE web page for the Strategic Advantage Project in August. It is designed to provide staff and teaching teams with background information on the project, supporting materials, teaching materials, workshop dates, and bulletin board.

During September, the co-chairs conducted personal visits with the organizations and commodity groups that participated in the design process. The purpose of these visits was to formalize networking, marketing, and recruiting commitments for the project. Cooperating commitments have been reached with the following organizations and groups: Iowa Farm Bureau Federation, Iowa Soybean Association, Iowa Dairy Products Association, Iowa Pork Producers Association, Iowa Corn Grower's Association, Iowa Cattlemen's Association, Iowa Banker's Association, Farm Credit Services of the Midlands.

Six teaching team training sessions were conducted during August, September, and October. A total of twelve teams were trained, and they are prepared to deliver workshops across the state during the 1997-1998 program year. Again, team members consist of the farm management specialist serving as team leader along with the appropriate family life and crop/livestock production specialists

Year One Delivery

Thirty Strategic Advantage workshops have been scheduled across the state for the 1997-1998 winter meeting season. A target has been set to reach at least 500 farm families who want to improve the competitiveness and profitability of their family farm businesses.

A state-wide marketing and awareness campaign is expected to begin in October.

What Iowa's Manure Law Means to the Dairy Industry

W. J. Powers, assistant professor
of animal science

DSL-116

Introduction

In 1995, Iowa Act House File 519 went into effect. This piece of legislation, intended to regulate confined feeding operations, will continue to be a key factor in determining future growth of Iowa's livestock industry, including the dairy industry. As a result of an increasing global population consuming a greater percentage of meat and dairy products in the daily diet combined with less land available for livestock production, the trend in American agriculture is to have fewer, larger livestock operations. Although House File 519 is considered a law that pertains to confined animal feeding operations (CAFO) dairy operations do not have to be very large in size to feel the impact.

Construction Permits

Operations planning to construct, install, or modify a manure storage facility need to determine if a construction permit must be obtained prior to beginning any construction. Construction permit requirements apply to both open feedlot and confinement operations as follows.

Open feedlot permit thresholds. An open feedlot is defined as an unroofed or partially roofed livestock operation where no vegetation, crop, forage growth, or cover is maintained while animals are confined. An open feedlot must obtain a construction permit if the feedlot has a capacity of 1,000 animal units (one animal unit=1,000 lb liveweight) or 300 animal units and manure is discharged directly to a water of the state or discharged through a man-made drainage system to a water of the state.

Confinement operation permit thresholds. Determination of permit requirement for a confinement operation is based on animal weight capacity of the operation and planned or existing form of manure storage. Table 1 illustrates the maximum animal weight capacity for operations with varying manure storage facilities. Above the indicated animal weight capacity for a given storage system, a Department of Natural Resources (DNR) construction permit is required.

Table 1. Confinement construction permit guidelines.

Animal Weight Capacity (lb)	Storage type
<400,000	No permit needed
>400,000	Anaerobic lagoon Earthen pit Aerobic lagoon
>1,600,000	Formed storage
>4,000,000	Dry storage

400,000 lb=approx. 286 mature cows
1,600,000 lb=approx. 1142 mature cows
4,000,000 lb=approx. 2857 mature cows

Manure Management Plans

Confinement operations are prohibited from discharging manure directly into any accumulation of water, surface or ground. Manure must be contained between periods of application and adequate storage capacity for manure must be provided. Additional storage capacity must be provided if precipitation or wastes from other places has access to the storage structure. Prevention of manure overflow must be provided by maintaining adequate freeboard (earthen basin, 2 ft; unroofed formed structure, 1 f.). Open feedlots must remove all settleable solids prior to discharging to a water of the state. Rainfall resulting from the largest average rain in 24 hours in 25 years must be contained. All operations, including open feedlots, are prohibited from discharging to a sinkhole, public lake, or agricultural drainage well.

All operations that are required to obtain a construction permit, must also file a manure management plan. Additionally, operations not required to obtain a DNR construction permit but that store manure in other than an exclusively dry form and that house livestock other than cattle with an animal weight capacity of greater than 200,000 lb are required to file a manure management plan. So, if a dairy operation also raises a few chickens or

hogs, even if the numbers are few enough to constitute personal-use animals, and all manure is not stored exclusively in dry form, a manure management plan must be filed if the total animal weight capacity exceeds 200,000 lb. A manure management plan must include:

1. Sources and quantities of manure/wastewater generated and nitrogen content.
2. Optimum crops yields and crop usage rates, nitrogen credits, supplemental inorganic fertilizer applied.
3. A recordkeeping system for maintaining information on manure application (location, method, timing, application correction factors).
4. Land area requirement calculations.
5. Identified methods to reduce soil loss and potential water pollution and odor nuisance.

Manure sales. A confinement operation required to submit a manure management plan may submit a "sales of manure" plan if the operation has a history of selling manure or it is considered common practice to sell manure. In this case, a manure management plan must consist of:

1. An estimate of annual animal production and manure volume or weight produced.
2. The total nitrogen produced.
3. A manure sales form stating the name and address of the purchaser, amount purchased, crop yield and usage rate, application methods and timing, location and number of acres where manure will be applied, and manure application rate.
4. A statement of intent from the purchaser of the manure, including name and address of the purchase; statement indicating the intent to purchase manure; the location of the farm and total acres available; and the purchaser's signature.
5. Recordkeeping for a minimum of 3 years.

Separation Distances

Confinement operations, constructing or expanding, are subject to minimum separation distance regardless of whether or not they are required to obtain a DNR permit. Open feedlots are exempt from separation distances. Minimum separation distances are based on animal weight capacity and manure storage system. Distances are measured from the edge of the manure storage structure to the nearest edge of the neighboring structure (house, school, park, river, drainage well, etc.). Table 2 summarizes distance requirements.

Table 2. Separation distances for bovine operations under Iowa's manure law.

Storage Type	Body Weight (lb)	Buildings within incorporated areas and public use areas (ft)	Houses, schools, churches and businesses, unincorporated areas (ft)	Surface intakes, ag drainage wells and sinkholes (ft)	Navigable lakes, rivers, streams (ft)
Anaerobic lagoon, Uncovered earthen pit	<1,600,000	1,250	1,250	500	200
	>1,600,000	1,875	1,875	500	200
	>4,000,000	2,500	2,500	500	200
Uncovered formed pit	<400,000	0	0	500	200
	<1,600,000	1,250	1,000	500	200
	>1,600,000	1,875	1,500	500	200
	>4,000,000	2,500	2,000	500	200
Confined building, Covered earthen pit, Covered formed pit, Washwater storage	<400,000	0	0	500	200
	<1,600,000	1,250	750	500	200
	>1,600,000	1,875	1,000	500	200
	>4,000,000	2,500	1,500	500	200

Water Withdrawal Permits

Operations withdrawing more than 25,000 gallons of water per day (gpd) must apply for a water withdrawal permit form the DNR. Considering a lactating cow typically drinks 50 gpd, only 500 cows, excluding youngstock and washwater, are needed to meet this requirement. Dairies using flushed manure handling systems will typically use 120 gallons of water per cow per day equating to a maximum of 208 lactating cows without having to obtain the water use permit. Permits are granted for up to 10 years. As part of maintaining permit rights, a producer must submit water usage reports to the DNR. Water conservation conditions and a water conservation plan are included in a permit. Additionally, a producer may be required to conduct a controlled aquifer test to determine the effects on other water uses.

The DNR does reserve the right to restrict water use in the event of a drought or local crisis affecting water supplies. Permit modifications may be made if the permitted well causes a water level decline in an unregulated well that existed prior to the permitted well. However, a water usage permit does protect the producer's right to water in the event of future subdivision development. If an unpermitted well is constructed after a dairy operation permits their well, then the dairy producer is not liable for interference of water use.

Animal Disposal

Animal carcasses must be disposed of within 24 hours of death by either a commercial rendering service, burial, or incineration. The DNR serves as the regulatory agency overseeing animal disposal.

Burial. A permit for animal burial is not required if carcasses are buried on land classified by the NRCS as "moderately well drained," or drier, or on tile-drained land. Additionally, carcasses must be buried within 6 feet deep and covered with a minimum of 30 inches of soil. Minimum burial distances are as follows: 50 ft from adjacent property; 100 ft from a private well; 200 ft from a public well; and 500 ft from a neighboring residence. A maximum of two carcasses per acre of animals over 2 months of age are allowed with an unlimited burial of animals less than 2 months of age. Burial in a floodplain area is prohibited.

Incineration. Although a DNR permit is not required for on-farm incineration, emission standards must be met. Incinerators with a burning capacity greater than 1,000 lbs. per hour must not discharge particulate matter in excess of 0.2 grain per standard cubic foot, adjusted to 12 percent carbon dioxide. Incinerators burning less than 1,000 lb per hour must ensure particulate matter discharge less than 0.35 grain per standard cubic foot, adjusted to 12% carbon dioxide. All incinerators must emit visible air contaminants below 40% opacity (smoke heaviness).

Nuisances

Operations of all sizes are vulnerable to nuisance suits. Nuisance conduct is considered as unreasonable or unlawful and causes annoyance, inconvenience, discomfort, or damage to others in the use and enjoyment of their property. Even though a producer complies with all environmental and zoning laws, the producer may still be liable for nuisance. A nuisance defense is provided to the producer under Iowa's right to farm laws. If an operation is deemed to be within an agricultural area established by the county then the operation has a statutory defense against nuisance lawsuits provided the dairy is operating within state and federal regulations and is not found to be negligent. Likewise, the animal feeding operations nuisance defense presumes that ordinary agricultural activities do not bring about loss or detriment to a second party. Again, the defense can not be used if the producer is found to be negligent in complying with state and federal regulations. Despite the existence of nuisance defenses, producers should not consider themselves 'above the law'. The defenses are only valid if the producer meets qualifying criteria. All nuisance suits should be taken seriously and precautionary measures taken to prevent the occurrence of nuisance complaints.

Odor Management: Principles and Practices

W. J. Powers, assistant professor of animal science

DSL-117

Chemistry of Odor Generation

Odors associated with livestock operations may stem from feed storage areas, housing facilities, the animals, and perhaps, primarily, manure storage areas.

Over 75 odorous compounds, in varying proportions, have been identified in livestock manures. The number is much greater if all intermediary degradation forms of primary chemicals are counted. Groups of primary odorous compounds identified include: volatile organic acids, aldehydes, ketones, amines, sulfides, thiols, indoles, and phenols. All of these groups can result from the partial decomposition of manure. The break-down is accomplished by a mixed population of anaerobic bacteria, commonly grouped into either acid-forming or methane-producing classes. The acid formers are responsible for the initial breakdown of complex (odorous) molecules into short-chain compounds, including organic acids. The methane bacteria further reduce the organic acids to methane and carbon dioxide (nonodorous endproducts),if conditions permit action by methane producers.

An accumulation of these intermediate metabolites results in an offensive-smelling product whereas containment of the intermediate compounds for sufficient time to permit methane producers to act completely permits metabolism of the most of the odorous compounds to nonodorous methane. Background levels of sulfur in water also may be a source of odor.

Minimizing Odor Potential

To minimize odor potential, odors must be controlled by limiting or altering the formation of odorous compounds or by controlling the emission of odors. Source control can be approached through dietary manipulation or storage and treatment. Emission control is achieved through storage or treatment.

In a study with lactating Holstein cows, a commercial product, marketed as an odor- control product, was fed and the impacts on manure odor and animal performance measured (7). No performance differences were found between the treatment and control groups, i.e., milk yield, milk fat percentage, milk protein percentage, and solids-corrected milk. Likewise, no differences in manure (urine + feces), feces, or urine odor intensity were found. However, ammonia concentration in urine, feces, and manure was reduced by feeding the additive. Others have found that feeding specific ingredients has been beneficial to odor. Watts and Tucker (8) found that barley-based diets resulted in manure that was less intense than manure generated from feedlot cattle fed sorghum-based diets. Kellems et al. (3) determined that although addition of sagebrush to cattle diets was ineffective in changing manure odor, addition of peppermint oil did reduce manure odor offensiveness. More work is needed in this area; however, feeding diets which more nearly meet the needs of an animal without overfeeding nutrients, particularly protein, do demonstrate a tendency to reduce manure odor (2,4).

Malodor also can be controlled if the accumulation of odorous intermediate degradation metabolites can be effectively controlled through inhibition of formation or by providing an environment that promotes complete degradation to odorless or low-odor endproducts.

Anaerobic digestion is a complete digestion process that provides the additional advantage of containing manure thereby preventing odors from being emitted during the digestion process. The result is a low-odor effluent with the nutritive value of the manure retained. Powers et al. (6) observed odor reductions of over 50% after conventionally digesting flushed dairy manure at 75°F for 20 days. A fixed-film digester uses a porous media for bacterial attachment thereby providing a greater population of bacteria in the digester. The result is a reduced digestion time for successful digestion, and hence, lower capital investment by requiring a smaller digester volume. Powers et al. (6) observed similar odor and performance between a fixed-film digester with a 2.3-day digestion time and a conventional digester with a 10-day digestion time. Although anaerobic digestion is often viewed as not economically viable, this treatment technology is a good option in situations where odor control is a primary objective.

Another approach to odor control may be to limit the nutrient loading of a manure storage/treatment system. Solids separation via gravity settling, chemical flocculation, or mechanical separation may be used to try to improve odor characteristics of stored manure. Although little impact of solids removal on odor characteristics has been conducted, research studying these practices by using dilute dairy manures has demonstrated great promise in removing nutrients, primarily nitrogen and phosphorous (1, 5). Additionally, odor intensity from anaerobically digested dairy manure that was first sieved to simulate mechanical separation was less than unsieved digested manure (6).

Although commercial pit additives have received a great deal of interest, little data is available that demonstrates their effectiveness in the field. Powers et al. (7) added five commercial additives to anaerobically digested dairy manure and undigested manure and found no effect on odor intensity. Concentrations of

various measured chemical analytes commonly associated with odor were altered by product addition: however, odor intensity was not improved. In fact, one product was found to exacerbate odor. Others have found that some products are effective under a given set of conditions. Work conducted by Dwaine Bundy at Iowa State University indicates that some products show potential for odor reduction. A website (http://www.ae.iastate.edu) has been constructed to provide findings from his laboratory and illustrate the test conditions. Caution should be executed in choosing a commercial pit additive. Producers need to be sure that the product has been proven effective under conditions similar to the producer's own.

Other methods that might be employed to reduce odors include covering manure storage areas and providing adequate landscaping to help trap odorous particles from moving downwind.

Conclusions

Odor control measures are an increasingly important part of livestock management plans. Effective means of reducing odors from dairy manure include anaerobic digestion and pre-screening of fibrous solids. Although commercial additives, added to manure and feed, altered concentrations of odor constituents, overall odor intensity was unaffected. More work is needed in the area of odor control to demonstrate a cost-effective means of controlling odors generated from livestock operations.

References

1. Barrow, J. T., H. H. Van Horn, D. L. Anderson, and R. A. Nordstedt. 1997. Effects of Fe and Ca additions to dairy wastewaters on solids and nutrient removal by sedimentation. Appl. Engineering Agric. 13(2): 259 - 267.

2. Hobbs, P. and B. F. Pain. 1995. Odor reduction in fresh pig slurry by dietary manipulation of protein, pp. 5 - 10, *In* International livestock odor conference '95: New knowledge in livestock odor. Ames, IA, October 16 - 18.

3. Kellems, R. O., J. R. Miner, and D. C. Church. 1979. Effect of ration, waste composition and length of storage on the volatilization of ammonia, hydrogen sulfide and odors from cattle waste. J. of Animal Sci. 48(3): 436 - 445.

4. Obrock H., C. E., P. S. Miller, A. J. Lewis, and R. K. Shoemaker. 1997. The effect of reducing dietary protein concentration on odor emission from pig buildings. Presented at: American Society of Animal Science Midwestern Section, March 17-19, Des Moines, IA. Abstract number 38.

5. Powers, W. J., R. E. Montoya, H. H. Van Horn, R. A. Nordstedt, and R. A. Bucklin. 1995. Separation of manure solids from simulated flushed manures by screening or sedimentation. Appl. Engineering Agric. 11(3): 431 - 436.

6. Powers, W. J., A. C.Wilkie, H. H. Van Horn, and R. A. Nordstedt. 1997. Effects of hydraulic retention time on performance and effluent odor of conventional and fixed-film anaerobic digesters fed dairy manure wastewaters. Trans. of the ASAE 40(5): 1449 - 1455.

7. Powers, W. J., H. H. Van Horn, A. C. Wilkie, C. J. Wilcox, and R. A. Nordstedt. 1998. Effects of anaerobic digestion and additives to effluent or cattle feed on odor and odorant concentrations. J Animal Sci., submitted.

8. Watts, P. J. and R. W. Tucker. 1993. The effects of ration on waste management and odour control in feedlots, pp. 18 - 21, *In* Recent advances in animal nutrition in Australia 1993. University of Armidale, April 18 - 21, 1993.

Challenges and Opportunities in Dairy Manure Systems

Dan Meyer, extension field specialist,
Agricultural Engineering

DSL-118

As dairies get larger there is a trend to try to minimize scraping manure in four and six row drive-through freestall barns. Some producers are flushing the alleys in two percent sloped barns to one or two earthen storages. The earthen storage water is recycled to large vertical storage tanks at the top end of the building since it takes a lot of flush water. The problem that has been developing is that the solids build up in the storages. Also, in the spring when the manure storage overturns like a lagoon, the recycled liquid becomes too thick to flush the alleys with. This problem would even be worse but dairy farmers have moved away from using a lot of bedding in freestalls to mattresses which require very little bedding.

Two dairies in Northeast Iowa are attempting to solve this problem and reduce their bedding cost at the same time. They have installed liquid/solid mechanical separators. The separators can take out up to 16% of the suspended solids (normally 62 to 83% are suspended solids in dairy manure, the rest are dissolved solids). The percentage solids' removal can reach 26% where manure is scraped to the liquid/solid mechanical separator. They are hoping to compost the solids and reuse them for bedding. In the Midwest this concept is new. The winter conditions and high summer humidity make this task more difficult. Iowa State University has given legislative odor-grant money to these operations to assist them in solving their manure handling and odor problems. ISU also will be involved in helping them compost their separated solids. The freestall bedding option will be evaluated if the compost gets dry enough (35% is the lowest compost moisture level possible). Presently ISU is evaluating both forced aeration trials and turning the compost windrows with skid steer loaders on one composting site.

Iowa 4-H Dairy Report

L. H. Kilmer, professor,
Department of Animal Science

DSL-119

The purpose of the Iowa 4-H youth development program is to help youth become productive citizens by developing skills that will benefit them throughout life. The 4-H program emphasizes seven skills: development of a positive self concept; communication; decision-making; learning how to learn; ability to cope with change; citizenship; and leadership.

The Iowa 4-H dairy project utilizes dairy animals as tools to generate interest and enthusiasm to help youth develop the skills listed above. Various dairy related activities and programs have been developed to help accomplish the goals of the 4-H program. Many of these programs are organized and conducted by parents, volunteer leaders, and Iowa State University Extension field staff.

Iowa 4-H enrollment has continued about the same number (2,200) even though number of dairy farms in Iowa has decreased. The Iowa 4-H Dairy Endowment continues to grow with over $98,000 invested and proceeds being used to fund several on-going and new activities each year, including a one-half time summer intern in 1997.

4-H Dairy Conference

The National 4-H Dairy Conference continues to be a popular and well received activity for older youth with 9 Iowa youth participants in 1997. Iowa has been represented by at least 1 and as many as 12 youth each year since this conference was initiated in 1955.

Share-A-Heifer

The Share-A-Heifer program at the ISU Dairy Farm continues to be popular with youth who are not able to have a dairy calf of their own. This year 16 project members worked as a team to care for the project animals daily and then take them to the Story County Fair. Members learn about breeds, animal selection, animal care and management, and develop fitting and showing expertise. Other counties have requested information about the details of the project.

State 4-H Dairy Judging Contest

A junior division continued to be a welcome addition to the State 4-H Dairy Judging Contest as 12 county teams consisting of 40 individuals participated. The senior contest had 18 teams and 51 individuals competing for the opportunity to represent Iowa at an out-of-state contest. The team from Dubuque County finished first and later participated in the National 4-H Dairy Cattle Judging Contest that is held in conjunction with the World Dairy Expo in Madison, WI. The top senior judges who were not on the winning county team participated in the North American International Livestock Exposition Youth Dairy Cattle Judging Contest in Louisville, KY. These four youth were from Black Hawk, Delaware, Scott, and Winneshiek Counties.

4-H Dairy Quiz Bowl

The 4-H Dairy Quiz Bowl continues to gain interest, especially now that there are three age divisions to ensure more equitable competition. Six teams competed in the senior, 5 in the intermediate, and 2 in the junior division. The winning senior team from Clayton County represented Iowa in the national dairy quiz bowl contest which was held in Louisville, KY.

Dairy Youth Pentathlon

A "Dairy Youth Pentathlon" was conducted at the National Cattle Congress after a very successful initial offering in 1996. Eleven teams with 32 contestants participated in the 1997 event.

Other programs and activities such as State Fair Youth Dairy Show, 4-H Dairy Production Contest, and Dairy Youth Classic continued without major changes. Two other successful programs, 4-H Dairy Roundup and 4-H Dairy Camp were not offered in 1997 due to other commitments of staff who would conduct the activities. Both events will be offered in 1998.

DAIRY RECORDS MANAGEMENT SYSTEMS

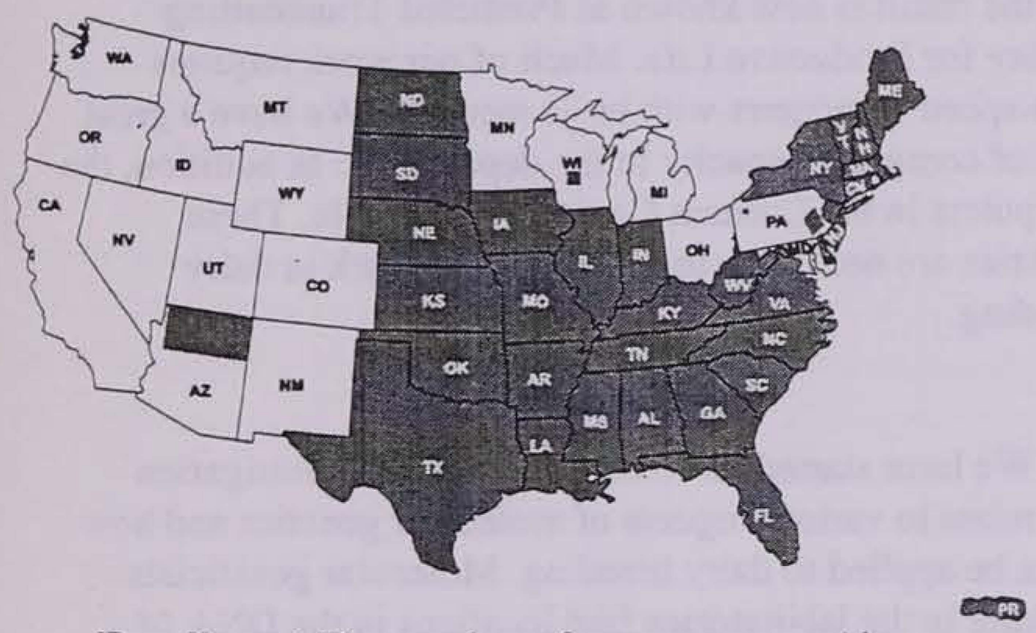
Greg E. Palas, dairy records management systems manager

DSL-120

Dairy Records Management Systems was formed by a merger of MidStates DRPC at Iowa State University and DRPC Raleigh at North Carolina State University in November 1996 and completed in June 1997. DRMS currently processes Dairy Herd Improvement Association records on over 1.7 million cows each month making it the largest volume processing center in the United States.

Mission Statement DRMS will provide member affiliates and other clients with high quality products and management services in a cost effective manner for the ultimate benefit of Dairy Herd Improvement dairy farmers.

Service Region Twenty-five DHIA service affiliates with herds in 41 states participate in the merged operation of DRMS. Nineteen DHIA affiliates are members of DRMS and are involved in the decision making and financial control of the organization.



Enrollment The number of cows processed by MidStates DRPC and DRPC Raleigh has steadily increased since 1970. The enrollment figures for the merged organization shows a steady growth. A higher volume of cows and herds for processing means a more economical cost for producers. It also ensures that staff is available to quickly respond to programming needs for DHIA reports or the PCDART system used by producers, DHI technicians, consultants, and extension staff.

Support and Service Staff in Raleigh and Ames provide telephone support to DHIA staff so they can provide support in their area. Annual training meetings for affiliate staff are provided, and DRMS staff attends local meeting throughout the service region to gain local exposure to customer needs or concerns. All herds are processed in Raleigh, but printing and mailing of reports from other sites ensures prompt turnaround of reports. Currently, reports are mailed and printed in Raleigh, NC, Ames, IA, and Ithaca, NY. Maintaining excellent support and prompt service as well as staying in touch with customer needs is a top priority and a major part of how the DRMS mission statement is achieved.

Mailed DHI reports include over 40 predesigned lists and summaries as well as custom-designed reports that are color laser printed. Producers can choose between four monthly reports with individual cow data, three cow pages, two somatic cell count summaries as well as a variety of herd data summaries.

PCDART Dairy Management System is a computer software package that provides a complete system to input, monitor, and manage reproductive/health changes, bST, prostaglandin, body condition scores, heifer growth, and many other aspects of the modern dairy. Herd trend graphs and individual cow scatter plots allow comprehensive analysis. Enrollment in PCDART for producers currently totals over 1,060 herds with 390,000 cows. More than 50% of these herds (538 herds) milk less than 200 cows, showing that PCDART is an essential management tool regardless of herd size. PCDART also is used by DHI technicians to input test day data. Over 450 consultants and veterinarians use PCDART to analyze DHI data from client herds. PCDART interfaces with most automatic milk recording systems for easy data transfer. PCDART data also can be transferred to various analysis programs including CTAP and DairyComp 305.

DRMS Advisory Board Six DHIA member producers and three DHI or extension staff members, serve as an Advisory Board to advise DRMS staff and approve major changes in programs or operating procedures.

DRMS Clients:

14,000	Producers with 1.7 million cows receiving full service
2,000	Producers with 200,000 cows receiving limited service from technician software
1,000	DHI technicians using TPE and PCDART for technicians
1,060	Producers using PCDART for 390,000 cows
450	Professional consultants accessing over 1,200 herds monthly
265	DHI managers, extension specialists, and county extension agents
18	Member service affiliates
7	Contract service affiliates

Overview of Dairy Cattle Breeding

A. E. Freeman, distinguished professor of agriculture; and P. J. Berger, professor of animal science

DSL-121

The overall objectives of dairy cattle breeding research are to generate new knowledge and to provide this information to breeders in Iowa and the nation. We are attempting to accomplish this by developing new knowledge that will be of value in the future and also have information that can be provided to the breeding industry to answer present-day problems. Because over 75% of the dairy cattle in the United States are bred artificially, our work will have more impact if it can eventually be applied through the artificial insemination (AI) industry and breed associations. However, there is usually a great deal of ground work that needs to be done before research can be applied on an industry-wide basis.

Education of undergraduate and graduate students is central to our service to the people of Iowa, the United States, and over the world. We participate in undergraduate teaching and our breeding group has total responsibility for graduate education in animal breeding and genetics in this department. We have three specializations under the Animal Breeding Ph.D. degree: quantitative genetics, molecular genetics, and immunogenetics. Within each of these specializations students take about the same courses and they work on one or more research topics on the species of their choice. We have had students that worked in dairy breeding that have been in all three specializations, but by far the most have worked in quantitative genetics. The Animal Breeding group has reorganized the complete graduate curriculum. We considered what we had been teaching and what we thought should be taught, then reordered the material into logical sequences for courses and now we are teaching these courses. Some revision may be necessary over time, but we feel good about the courses and their content. We also are teaching a beginning animal breeding course over the Iowa Communications Network (ICN) to people in the state.

Personnel

People working totally or in part in dairy breeding are:
Dr. A. E. Freeman, Professor
Dr. P. J. Berger, Professor
David Kelley, Agricultural Specialist
Jay Beck, Superintendent Agricultural Research Station
Mary Healey, Systems Specialist I
Gloria Lantz, Clerk III
Becky Stone, Clerk Typist III

The following are graduate students, where they are from, and the topic of their research:
Melvin Kuhn—graduate student, Mendon, IL, Effect of preferential treatment on sire and cow evaluation;
Steve Kelm—graduate student, Waterville, MN, Evaluation sires for health of their daughters;
Gamal Abdel-Azim—graduate student, Cairo, Egypt, Maximum genetic gain using molecular markers as an aid to selection;
Santos Nerilson—graduate student, Vicosa, Brazil, Adjusting records on cows milked 3X to a 2X basis for breeding evaluations;
Seyrani Koncagul—graduate student, Turkey, Genetic prediction of breeding values in population under selection;
Christy Meyer—graduate student, Iowa City, IA, Recent trends and factors affecting stillbirths in Holsteins.

Facilities

We are fortunate to have the herd at Ankeny to use for research and where experience managing the herd has often pointed out problems that lead to research projects. Calving problems experienced in the herd led to the calving ease evaluations that are now computed at Iowa State by Dr. Berger and are distributed around the world. Another example is how to use type scores to predict herd life. Our initial work relating type scores to herd life was augmented by UDSA-AIPL, where the sire evaluations are computed, and the result is now known as Predicted Transmitting Ability for Productive Life. Much of our work requires high-speed computers with large memory. We have a great deal of computer capacity in the department. In addition, the computers in the Durham Center are available. These facilities are necessary to do competent work in dairy breeding.

New investigations

We have started on some new areas of investigation that relate to various aspects of molecular genetics and how it can be applied to dairy breeding. Molecular geneticists working in the laboratories find locations in the DNA of some families or lines that differ from the DNA of other families or lines. These polymorphisms in the DNA may or may not be associated with traits of economic importance. Finding which are markers for economic traits is a developing science. Determining which polymorphisms are markers is a statistical problem. We have simulated markers and genes that code for economic traits, called quantitative trait loci (QTL), on a chromosome in a computer. Then we used various statistical methods to find which was the most efficient in locating the QTL. Usually some family structure is used to locate QTL. We developed methods that use half sister families as they exist in usual AI populations. Some of these methods look good in their power, or ability, to detect QTL and others do not seem to be as efficient. We have applied these methods to data that Dr. Sue Denise,

University of Arizona, collected and detected markers for growth hormone.

In another study, we are attempting to determine the maximum, or near maximum, gain that could be made using markers that account for various parts of the genetic variation for a trait such as protein of milk production. The increase in production for milk, fat, and protein production is not likely to be great over what is now being done in practice. This is because the sires of sires of AI bulls now are selected with an accuracy of about 80%. However, the accuracy of bull mother selection is about 40%. We are now making about 300 lb. of genetic gain for milk per cow per year by using conventional progeny testing. Additional gains for production by using markers may be marginal, but it is important to determine whether money should be spent on marker-assisted selection for production, other methods to improve production, or on improvement of other traits. Another area in which markers could be of great value is in aiding in selection for improved health of daughters of AI bulls by looking at the immune system of sires as a means to predict the health of their daughters. We are currently starting to do this.

Genetic prediction of breeding values

Methods to improve the accuracy of genetic prediction of breeding values by using animal models are under investigation. Unknown ancestors, genetic trends and changes in genetic variance associated with region or other environmental factors complicate the analysis of field data. Data that models national breeding programs by using sires imported from other countries over many generations are being used to answer basic, fundamental questions that can lead to enhanced genetic predictions of breeding value.

Dairy Breeding Research Herd

D. H. Kelley, agricultural specialist II; N. J. Beck, superintendent Agricultural Research Station; and A. E. Freeman, distinguished professor of agriculture

DSL-122

We are privileged to be able to use the herd at Ankeny for genetics research projects of long duration. The herd members are Holsteins, of which more than 90% are registered and carry the prefix I-O-State. We are currently milking approximately 150 cows. Our total inventory of wet cows, dry cows, and replacements of all ages is about 360 head.

The milking herd is housed in free stall barns that are bedded with sand. We have one barn that allows us to measure individual feed consumption on 40 to 45 cows at a time. The remainder of the milking herd is housed in three conventional free stall barns and the cows are fed in fence line bunks. Milking is done twice a day in a double-5 herringbone parlor equipped to electronically record and collect milk weights at each milking.

Baby calves are housed in individual 4 feet by 8 feet pens until after they are weaned. These pens are easily dismantled for cleaning and disinfecting between occupants. If calves are doing well and eating dry food, they are weaned at about 4 weeks of age. After weaning, calves are placed in groups according to their size and are rotated through a series of group pens. Heifers are initially bred at 14 months of age if they have reached a body weight of 750 lb. Calves post-weaning, heifers, and dry cows are kept in loose housing.

Corn silage is produced on the research farm and stored in bunker silos and bags. All other feedstuffs, including concentrate mixtures, whole cottonseed, corn gluten, urea supplement, and hay, are purchased from outside vendors. Hay is purchased on a quality basis by using penalties and bonuses based on relative feed value (RFV) and percent moisture to arrive at a final purchase price. Concentrate mixtures and supplements are formulated to meet our specifications.

All animals are fed one of several different total mixed rations (TMR). The milking herd is fed one of three TMRs. These rations are formulated to sustain high, medium and low levels of production. All cows and heifers receive the medium ration for the first week after they calve. They are then moved into the pen receiving the high ration. They remain there for the next approximately 90 days regardless of their daily production. This allows all cows time to express their production potential and enables us to minimize any bias in the research data. After this time, the cows continue to either receive the high ration or are moved to the pen receiving the medium ration based on their daily production. This now occurs at 85 lb./day for cows and at 70 lb./day for heifers. Likewise, an animal's daily production also dictates when she moves from the medium level ration to the pen receiving the low-level ration. This move occurs when production drops below 50 lb./day for cows and 40 lb./day for heifers. The farm superintendent has the prerogative to move animals from medium to low ration at an earlier stage should these cows begin to exhibit excessive body condition.

Replacement heifers are fed a TMR formulated to allow them to reach a body weight of 1,200 lb. at the time of first calving. It is our intent to have cows dry off with a body condition score of about 4.0. Dry cows are fed hay and a restricted amount of a high fiber-corn silage diet that will maintain this body condition through the dry period.

Foundation cows for this herd were purchased as open heifers from 38 Iowa breeders as beginning in 1968. The primary focus of the research through 1988 was milk production achieved using bulls whose proofs were high versus those whose proofs were breed average. The current selection experiment began with inseminations made in 1986. Cows and heifers from the milk selection project were assigned at random to one of two groups. The selection criteria for the two sire groups was the sum of pounds of fat plus pounds of protein in their proof. One group of sires is the highest available, the other group is selected to be at the level of the average of the breed. The most recent rolling herd average (September 1997) was 20,736 lb. milk, 722 lb. fat, and 678 lb. protein.

Cows in the herd have been and continue to be used for a wide variety of experimental needs in addition to the aforementioned long-term genetic study. This is achieved because we collect complete health, growth, and production data on all animals. These animals are members of two contrasting selection lines that, because of the herd's origin, represent the national Holstein population. Data generated by the herd have been used recently to study: (1) mitochondria function; (2) genetics of immune function, (3) how to genetically alter milk composition; (4) genetic differences in the normally circulating growth hormone; (5) changes in bovine leukosis and any genetic influences on bovine leukosis virus under normal herd management; (6) changes in health and reproductive performance as related to genetic differences; and (7) feed efficiency in two genetic lines and in different maternal families. In addition, the herd allows students from the College of Veterinary Medicine to be trained in reproductive medicine.

The city of Ankeny has begun construction of a north-south access street through the University property. At present, this construction has no effect upon our research, but it may signal future changes.

Genetic Change Using Information about Actual Genes Compared with Genetic Change Using only Production Records

M. T. Kuhn, graduate student in animal breeding;
R. L. Fernando, professor of animal science; and
A. E. Freeman, distinguished professor of agriculture

DSL-123

Summary and Implications

Genetic evaluation currently relies solely on production records of cows. It is now possible, however, to determine some of the actual genes that an animal carries. This study looked at one particular way that this new type of genetic information might be used in a breeding program. The approach considered here was to simply incorporate the genetic information into each animal's genetic value estimate (e.g., PTA) and then base choice of breeding stock on these "new" genetic value estimates. This approach, to using this new genetic information, results in greater genetic gain in early years, compared with just using production records, but less genetic gain in the long run. The primary implication is simply that further consideration needs to be given as to how **best** to use the new genetic information.

Introduction

In the past, it was not possible to directly determine exactly what genes an animal carried, and estimation of genetic values (e.g., PTAs) has relied solely on the use of production records. New laboratory techniques, however, allow for determining some of the actual genes that an animal carries. These genes are called quantitative trait loci, or QTLs.

There are many different ways that this QTL information might be used in a breeding program. The most straightforward approach would be to just simply incorporate that information into each animal's genetic evaluation and then just "proceed as usual." In other words, still base choice of breeding stock (sires, for example) on estimated genetic values (e.g., PTAs) but now the genetic evaluation would also include the new QTL information.

When using this approach, one important point to bear in mind is that there are many genes affecting traits such as milk yield and only a few of these genes will be known, via the new laboratory techniques. Therefore, production records must still be used, in conjunction with the QTLs, to estimate an animal's genetic value, otherwise the other (unknown) genes affecting the trait would not be included in an animal's genetic value estimate.

The main objective of this research was to compare the amount of genetic progress obtained, in both the short and long term, when genetic evaluations do and do not include QTL information. We consider the case where only one gene (QTL) is known.

Materials and Methods

First a note on terminology. In animal breeding, the process of choosing which sires to use and which cows to use to produce new female replacements is called "selection." You select which bulls to use and which cows you'd like to have producing new female replacements.

One approach to addressing the objective of this research would be to select one set of cows, and their mates, based on estimated genetic values which do not include QTL information; then, take another set of cows and do the same thing except base selection decisions on estimated genetic values which have incorporated the QTL information and, at the end, just look and see which one did better and by how much. To use this approach with actual, live animals, however, would be prohibitively costly, both in terms of time and financial costs. Fortunately, it is possible to program a computer to simulate (or imitate) the same thing that would happen if real cows and bulls would have been used and so this simulation approach was used in this study.

To summarize, the method used to compare genetic progress obtained with and without QTL information was as follows:

- Simulate data and base selection on use of records only
- Simulate data and base selection on use of records and QTL information

and then just compare which approach resulted in the greatest genetic gain over time. The "simulate and select" step was carried out for a total of 30 generations and amount of gain, for each approach, was determined at each generation. Note that "use of records" for dairy bulls would mean, for example, use of female progeny records. The key distinction here is just whether records only are used or if records and QTL information are used, when making selection decisions. Selection based on records only will be called phenotypic selection, whereas selection based on a combination of records and QTL information will be called QTL selection.

Results and Discussion

In general, QTL selection resulted in the most genetic gain in early generations but then phenotypic selection caught up and, in the long run, most genetic gain was typically made by phenotypic selection. Recall that there are actually many genes, other than just the known QTL, affecting traits such as milk yield. The reason for the long term advantage of phenotypic selection was because it built up an advantage at genes other than the QTL, and QTL

selection was never able to compensate for its sacrifice at the other genes. Basically, then, QTL selection results in an initial thrust and beats out selection based just on production records, but then selection based only on production comes back and wins in the long-term.

It should not, however, be hastily concluded that QTL information is not worthwhile but rather simply that further consideration needs to be given as to how to best use this information. Some researchers at Iowa State, for example, have looked at optimum weighting of the information from production records and a QTL when estimating genetic value.

Furthermore, the approach to using QTL information considered here (simply incorporate it into each animal's estimate of genetic value, and then "select as usual") is only one possible way to use the information. Other possibilities, such as using it in mating, rather than selection, decisions also exist and perhaps should be explored.

Another possibility might be to use QTL information only to improve secondary traits such as somatic cell count or reproduction. Genetic improvement in primary traits, such as production, could then rely on selection based on genetic evaluations from phenotypic information only, at least until the optimal use of QTL information has been explored further.

Genes affecting production and other economically important traits continue to be identified and almost assuredly will provide new avenues for bringing about even greater genetic gain than is currently occurring. How best to use the QTL information, however, is still under study.

Estimating Quantitative Trait Loci in Half Sib Families Under a Random Model with Missing Parental Genotypes

M.L. Martinez, visiting scientist; N. Vukasinovic, visiting scientist; and A.E. Freeman, distinguished professor of agriculture

DSL-124

Summary and Implications

The results of this study demonstrate that the random model approach performs well when applied to Quantitative Trait Loci (QTL) mapping in livestock populations with half-sib family structures. The QTL are genes that actually code for a trait of economic importance. QTL with large effects, which account for a large proportion of total phenotypic variation, can be detected with high power and accurately located, even with missing parental marker information. The method, however, fails to precisely locate a small QTL, especially when the QTL is close to the end of the chromosomal segment.

Introduction

Dr. Kehrli characterized the immune profiles of 60 sires now being progeny tested by 21st Century Genetics. The objective is to rank sires for health of their daughters in producers' herds. Producers are recording health incidences of daughters to determine how well the immune profiles of their sires actually predict daughter's health in the "Health Traits Project."

This study was started with the anticipated use of molecular markers for health traits to follow up on the Health Traits Project. We have collected blood samples as a source of DNA from as many of the daughters of the test bulls as we could and also collected blood from the dams of the daughters of the test bulls. It is difficult and expensive to collect, record, and use all the health problems that daughters of sires have in commercial herds. Determining the immune profiles of sires would be much easier. Our question in this work was, could we use the normal, or existing, half-sib family structure to locate markers for health traits and with what accuracy could we find markers for the QTL for the health problems.

Materials and Methods

The random model approach is based on the phenotypic similarity between genetically related individuals. The similarity between relatives can be thought of as one where a few genes, or QTL, affect the expression of a trait. The second part can be thought of as many genes each with small effects that also affect the trait, but the affects of the many individual genes are not known. This approach uses the information that the more closely animals are related the more they should produce alike.

The idea that the more similar family members are the more they should be alike was not new, but we developed the theory that was needed to apply this to existing dairy cattle populations and also extended the theory to include use of information on the mothers of the calves, when all mothers had data and when some of the data related to mothers were missing. The mothers referred to here were the cows that were bred to the bulls being used in the Health Traits Project. This theory can be used in general, but it also applies directly to our Health Traits Project.

The method that was used to test the theory that was developed was to simulate genes on a chromosome in the computer. In addition to the genes that we assumed to be coding for effects, or the QTL, where we knew their location on the chromosome, we also generated marker genes at varying distances from the QTL. The QTL and marker genes were located along the length of the chromosome. Thus, we knew the location of the QTL and their effects on production and we also knew where the marker genes were located. We also used different levels of heritability, so that we could make inferences to traits with different levels of heritability. Examples are: low heritability of about .05 to .10 for reproduction, medium levels of about .25 to .35 for milk and protein pounds, and higher levels of .5 to .6 for percentages of fat and protein. After all was programmed to run on the computer, we applied our ideas to determine how well we could find the QTL by using the markers. This is a very computer-intensive application and we used hundreds of hours of time on Iowa State's large work stations.

Results

The results showed that if the QTL was located near the end of the chromosome that the estimate of the location of the QTL was biased, and this was most pronounced when heritability was small. Also the estimated effect of the QTL was underestimated. However, when the QTL was not on the end of the chromosome the estimation of where the QTL was located and the effects of the QTL were unbiased and the method performed very well. In all cases, it was easier to find the QTL and estimate its effect when heritability was at medium or high levels.

Use of information that the dam brought to this approach also was investigated. Information on the dam contributed minimally to locating the QTL or estimating the effect of the QTL on the trait. This is because we only have one sample of DNA from the dam as opposed to a sample of DNA from each daughter of a sire, thus half-sib data are more useful to locate and characterize the effects of QTL. We anticipate these results will be useful to us in future work and to other scientists. Ultimately the producers and consumers will benefit.

Differences in Feed Efficiency in Dairy Cows

Ann Williams, graduate student in animal breeding; and A. E. Freeman, distinguished professor of agriculture

DSL-125

Feed is the largest cost of production for most dairy producers. Yet, because of the difficulty and cost of measuring the feed intake and refusals, few, if any, producers can afford to determine feed efficiency on their cows. There is research that indicates that feed efficiency increases as production increases. Much of this work was done where cows were fed strictly according to production and probably does not now apply when cows are allowed to eat all they can consume. Further, much of the data that indicates this strong positive correlation between feed efficiency and production were from trials with few numbers and the cows did not continue for a complete lactation. Even so, there is little doubt that, in general, this strong correlation does exist between grass-fed efficiency and production.

As we select for higher and higher production, unanswered questions remain: Are we improving feed efficiency, Are we actually not changing feed efficiency, or Could feed efficiency be decreasing? We completed a long-term selection experiment at Ankeny where selection was for high and average milk. This was done by selecting the highest bulls for predicted transmitting ability (PTA) milk and bulls with breed average PTA for milk. The genetic difference in the two selection lines was 2,900 lb. of milk. The cows were fed and managed alike to the best of our ability. An interesting question is what was changed in the cows to account for this large difference. While this experiment was being conducted, we did not have facilities to measure feed efficiency.

Materials and Methods

We started a new selection experiment by randomizing cows from the milk-only selection experiment into two groups. One group is being bred to artificial insemination (AI) bulls that are the highest for PTA for combined pounds of far plus protein and the other group is being bred to breed average AI bulls selected for PTA for pounds of combined fat plus protein. We now have facilities for measuring feed consumed and refusals at the Ankeny dairy. We have measured total lactation feed intake on about 150 cows at the beginning of this fat plus protein selection experiment. As time progresses, we will measure feed efficiency again and determine whether the differences generated by selection are just because the high cows consume more, or because selection produces more subtle metabolic changes.

We have a 48-stall barn that has Calan gates installed. When we are collecting feed intake, each cow has a transponder around her neck and only the one gate is programmed to open for one transponder. Thus, the feed that each cow consumes can be determined by weighing the feed into each feeder and weighing refused feed. We have a cart that allows automation of much of the work of getting individual feed intake. Feed was weighed to the cows for two feedings per day and refusals were weighed three times per week. Feed fed and refused was sampled each week, composited across 2 weeks and determinations of feed ingredients were determined on the composited 2 week samples. Body weights and condition scores were taken at 4 week intervals by Jay Beck and Dave Kelley. Three rations were fed. All cows and heifers are fed the most nutrient-intense ration after they calf. This ensures that all cows have an equal opportunity to produce, and they are assigned to lots with less concentrated rations as they drop in production.

We have now completed the first round of determining feed efficiency. The gates have been taken out of the barn and the barn is now operated as a normal free-stall barn with feed fed in front of the posts that supported the Calan gates.

Results

We have summarized some of the results of the feed efficiency data. We conducted a digestibility trial on 24 cows for the two selection lines. There were no significant differences between selection lines. Lines had not had time to diverge enough to detect differences. These data will give a benchmark for future reference. The average, standard deviations, and ranges for digestibility are given in Table 1.

Table 1. Means and standard deviations for nitrogen digestibility, dry matter digestibility, gross energy digestibility, and digestible energy.

Trait	Average (%)	Standard Deviation	Range
Nitrogen Digestibility	0.6734	0.0582	0.5321—0.7711
Gross Energy Digestibility	0.5651	0.0778	0.3834—0.6863
Dry Matter Digestibility	0.5640	0.0754	0.3904—0.6912
Digestible Energy (kcal/g)	2.456	0.3432	1.655—3.004

These values are in the normal range of data published. Notice that the standard deviations (a measure of variation) are small relative to the averages; thus, we have rather precise measures of digestibility. Analyses of these data showed that differences among rations, parity, and days in milk significantly affected digestibility for nitrogen gross energy, dry matter intake, and digestible energy.

Gross feed efficiency was determined on 141 cows. The average gross feed efficiency was 1.344. This says that for each pound of feed fed, the cows produced 1.344 lb. of milk. Average actual production was 59 lb. and feed intake was 44.97 lb. Most of the cows were first-calf heifers and all data are on a lactation basis. The three rations were converted to a total digestible nutrients (TDN) basis to have a common measure of feed intake across rations. Average lactational body condition scores, TDN intake and year-season of calving had significant effects on lactational feed efficiency.

These data were analyzed by 4 week time periods. In some periods there were significant differences between selection lines for combined fat plus protein and among maternal lineages. These differences were not large. There were, however, large differences between parities, year season of calving, body condition scores, % TDN consumed, and days in milk on gross feed efficiency. Older cows were more efficient, probably because they produced more and were not growing as much as the heifers. Year-season of calving had the largest effect in the early part of lactation. Body condition scores had the largest effect on feed efficiency between weeks four and 16, as expected. When body condition scores were considered the same time as body weights, body condition scores were always more important as an effect on feed efficiency.

We have determined from this work, where cows were offered all the feed they could consume, that it is not necessary to measure feed intake for a complete lactation to get a good estimate of efficiency. Measuring feed intake for any 8 week period between 56 to 140 days in milk is correlated (r=.85) with total lactation feed efficiency. Thus, with the same labor costs, we can measure feed efficiency on many more cows than getting complete lactation feed efficiency.

These data are not completely analyzed, but we now have a first measure of feed efficiency. Thus, when we take feed measurements later in the experiment, we will be able to determine whether differences in intake or digestibility can account for the difference between lines selected for high and average combined pounds of fat and protein.

Marker-Assisted Selection in Dairy Cattle

Gamal A. Abdel-Azim, graduate student in animal breeding; and A. E. Freeman, distinguished professor of agriculture

DSL-126

Introduction

In recent years animal breeders have recognized molecular genetics as a new source of information for genetic evaluation of domestic animals. In general, there are three overlapping phases for finding and using this information:

1. finding and mapping polymorphisms in DNA that may be potential markers,
2. relating the polymorphisms to traits of economic importance and thus establishing markers, and
3. using these markers in breeding programs.

The first phase is rapidly advancing. About 1,500 to 2,000 polymorphisms are known in cattle. Most of these polymorphisms are not currently known to be markers for any trait, but some are genes that are known to code for specific traits. Knowledge is progressing in the second phase in methods of determining which polymorphisms are indeed markers of economic traits. For example, markers are known for growth hormone, the weaver condition, and dumps. Little is known about how to use these markers for selection of young sires in applied breeding programs, and how artificial insemination (AI) bulls should be used in the commercial population of cows to maximize genetic gain by using markers. Simulation studies have generally shown gains by using marker-assisted selection were larger than with conventional selection in the first few generations (5,6). Others have shown that in long term selection gains are about the same (2). However, none of these studies simulated current dairy breeding programs, and used a different population structure than in the dairy industry; thus, the results could not be interpreted relative to today's dairy breeding.

Our goals were to determine maximum genetic gain obtained by incorporating marker data in genetic evaluation models used in current dairy breeding programs, and to compare this with conventional selection. The optimum use of marker information is to be studied to determine how to maximize genetic gain.

Research outline

Few studies are available that have studied marker-assisted selection (MAS). Land and Thompson (4) discussed MAS from a theoretical viewpoint and found that MAS improved the efficiency of selection, simple MAS was studied by (5, 6, 8), but none of these studies was related to practical dairy breeding. In this project MAS is studied in a population similar to the Holstein population. Generations overlap and the best sires are used more than sires of lower merit.

First, the maximum expected gain will be determined by assuming that each marker is the coding gene, then this assumption will be relaxed by allowing markers to be linked at varying distances from the coding genes or quantitative trait loci (QTL) on the same chromosome. The recombination rate, r, between markers and QTL will vary. Part of the genetic variance will be due to QTL and the larger part will be due to many other genes with small effects, i.e. polygenic component of variance.

The population structure will be representative of the Holstein breed in herd size and production level and scaled to 30,000 cows per generation, a number found to be satisfactory by Kuhn et al. (3). The USDA-AIPL animal model (7) will be used to evaluate sires and dams where the effects of QTL and polygenes are combined. All combinations of the following variables will be simulated:

1. number of genes constituting the QTL (1 to 4),
2. magnitude of additive genetic variance due to the QTL (5, 10, and 20%),
3. single and flanking markers,
4. gene frequencies of the QTL (0.2, 0.3, and 0.5),
5. heritability (0.1, 0.3, and 0.5), and
6. recombination rates (r=0.0, 0.1, and 0.3, where r=0.0 means the marker is the coding gene).

Simulation

A base population will be simulated and used for many generations of selection to determine the maximum genetic gain obtained when combining marker information with phenotypic information, to compare this with conventional selection, and to find an optimum way of using marker information in dairy cattle breeding programs. To simulate a dairy record, one has to account for environmental and genetic factors as

$$Y_i = E_i + A_i + e_i, \text{ where}$$

Y_i refers to a dairy record for individual i, which was simulated by adding environmental factors E_i and genetic factors A_i, in addition to a random error, e_i.

For any individual i, the genetic factors were simulated as usual genes. The whole genome of an individual was assumed to be composed of p genes and q QTLs. QTLs are also genes but one QTL could be one gene or a group of many genes tightly linked together. The breeding value of an individual is accounted for by the p genes and the q QTLs, and can be written as

$$A_i = a_{i1} + a_{i2} + \ldots + a_{ip} + qtl_{i1} + qtl_{i2} + \ldots + qtl_{iq}$$

ie, A_i is divided into two parts, the polygenic component, sum of a_is, and the QTL component, sum of qtl_is, i = 1, 2, ..., n, j=1, 2, ..., p, and k=1, 2, ..., q. Therefore, p loci were simulated for the polygenic component, q QTLs were simulated for the QTL component.

In simulating the data, the additive genetic variance was divided into two components, one component due to polygenes and the other due to QTLs. Gene frequencies of the polygenes were sampled from a beta distribution with special parameters ($\alpha = \beta$), Table 1. This distribution has a range of values from 0 to 1 and is concentrated about ½, so that the intermediate values have more probability of being sampled than the extreme values (near 0 and 1). The reason for that was to avoid the extreme values that have equal probability of being sampled to nonextreme values if a uniform (0,1) distribution would have been used. The uniform distribution has a range of values from 0 to 1, all of them have equal probability; therefore, sampling from a uniform implies that all values, intermediate and extreme, have the same chance of being sampled.

Because the data will go through many generations of selection, loci with extreme allele frequencies are likely to be fixed soon and their contribution to the genetic variance is lost as a result. On the other hand using a fixed value of ½ for all loci is unrealistic because it represents a cross of two inbred lines that do not have anything in common with the Holstein population.

Table 1. Allele frequencies, genotypic values, and additive genotypes or breeding values for each of 20 loci, simulated for the polygenic component, a sample data set.

Locus	Allele freq	Genotypic value	Additive genotype		
			A_1A_1	A_1A_2	A_2A_2
1	0.59	372.65	305.57	-67.08	-439.73
2	0.57	370.21	318.38	-51.83	-422.04
3	0.18	477.06	782.39	305.32	-171.74
4	0.32	392.91	534.35	141.45	-251.46
5	0.20	458.21	733.13	274.92	-183.28
6	0.64	381.84	274.92	-106.91	-488.75
7	0.68	392.91	251.46	-141.45	-534.35
8	0.40	374.12	448.95	74.82	-299.30
9	0.16	499.94	839.90	339.96	-159.98
10	0.52	366.86	352.18	-14.67	-381.53
11	0.55	368.41	331.57	-36.84	-405.25
12	0.37	379.62	478.32	98.70	-280.92
13	0.56	369.23	324.92	-44.31	-413.54
14	0.46	367.74	397.16	29.42	-338.32
15	0.70	399.95	239.97	-159.98	-559.94
16	0.57	370.21	318.38	-51.83	-422.04
17	0.74	417.85	217.28	-200.57	-618.41
18	0.42	371.35	430.76	59.42	-311.93
19	0.33	389.79	522.31	132.53	-257.26
20	0.48	366.86	381.53	14.67	-352.18

Having sampled the gene frequencies belonging to each locus and based on the knowledge of the locus proportion of the total additive genetic variance, genotypic value of the j^{th} locus, g_j, was computed as $((\sigma_j^2 / 2p_j(1-p_j))^{1/2}$, where σ_j^2 denotes the additive genetic variance of the j^{th} locus (1). Additive genotypes for each locus were then computed as $2(1-p_j)g_j$, $(1-2p_j)g_j$, or $-2p_jg_j$ and were assigned to individuals after sampling their genotypes from a trinomial distribution with three outcomes, $A_{j1}A_{j1}$ with probability p_j^2, $A_{j1}A_{j2}$ with probability $2p_j(1-p_j)$, and $A_{j2}A_{j2}$ with probability $(1-p_j)^2$. Table 1 lists allele frequencies, genotypic values, and additive genotypes for 20 loci of a sample data set. This approach was repeated for all loci, then their respective additive genotypes were summed to form the aggregate breeding value for each individual. Figure 1 is a graphical representation of the frequency distribution of the aggregate breeding values of 30,000 individuals. The values 1 to 25 on the horizontal scale are coded breeding values. The point 1 represents the smallest breeding value and the point 25 represents the largest breeding value.

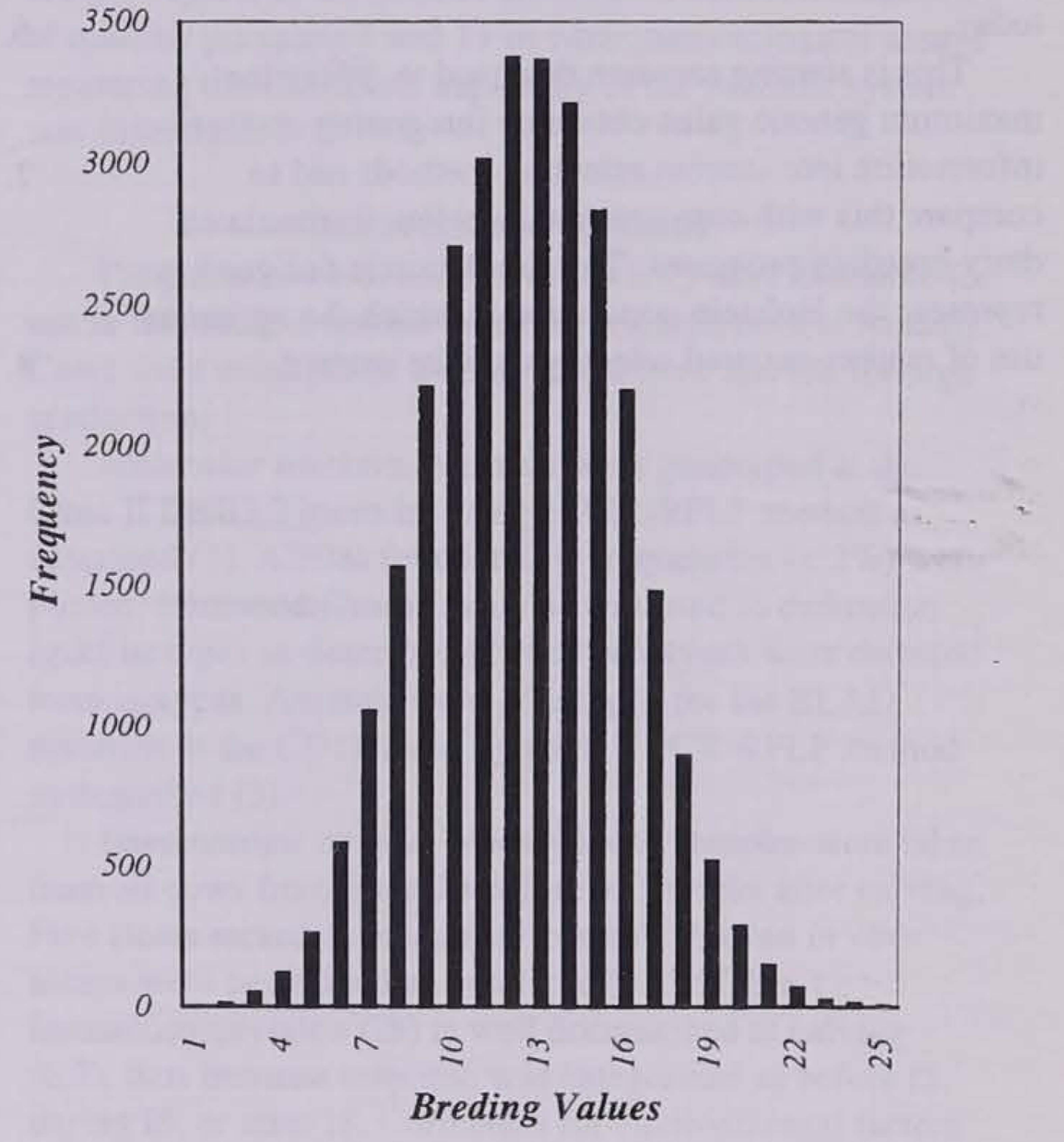


Figure 1. Breeding values were not sampled from a normal distribution, however, gene frequencies were sampled from a symmetric beta centered about ½. The breeding values came out to be normally distributed.

Final thoughts

As mentioned simulation will go on for several generations of selection. Animals will be evaluated with and without marker information. Based on Gibson's results (2), it might be thought that in the long run, conventional selection will catch up and perhaps surpass MAS. However, it is important to understand that Gibson's study was extremely theoretical and his simulation model was unrealistically

hypothetical. Gibson considered a fixable gene affecting the quantitative trait and direct selection based on a QTL genotype. In practice, however, selection is likely to be based on marker genotypes linked to QTLs, where recombinations take place. The linkage between markers and QTLs is expected to fade or weaken generation after generation, which could hinder if not stop the fixation of the QTL. Also, in cases other than assuming only additive relationships between alleles, the QTL might not be fixable. For example, in the case of complete dominance the number of generations required to eliminate the recessive is very large. Finally, detecting more marker-QTL associations in the future may keep selection based on marker information always superior to conventional selection. It will be eccentric to think that knowledge will stop at the few marker-QTL associations discovered today.

This is starting research designed to determine maximum genetic gains obtain by integrating marker information into current selection methods and to compare this with conventional selection in simulated dairy breeding programs. The simulation is designed to represent the Holstein population in which the optimum use of marker-assisted selection is to be studied.

References

1. Falconer, D. S. Introduction to quantitative genetics. Longman, England.
2. Gibson, J. P. 1994. Short-term gain at the expense of long-term response with selection with identified loci. 5th World Congress Genet Appl Livest Prod 21: 201.
3. Kuhn, M. T., P. J. Boettcher, and A. E. Freeman. 1994. Potential biases in predicted transmitting abilities of female from preferential treatment. J. Dairy Sci. 77: 2428.
4. Land, R. and R. Thompson. 1990. Efficiency of marker-assisted selection in the improvement of quantitative traits. Genetics 124: 743.
5. Meuwissen, T. H. E. and J. A. M. van Arendonk. 1992. Potential improvement in rate of genetic gain from marker-assisted selection in dairy cattle breeding schemes. J. Dairy Sci. 75: 1651.
6. Meuwissen, T. H. E. and M. E. Goddard. 1996. The use of marker haplotypes in animal breeding schemes. Genet. Sel. Evol. 28: 161.
7. Wiggans, G. R., I. Misztal, and L. D. Van Vleck. 1988. Improvement of an animal model for genetic evaluation of dairy cattle in the United States. J. Dairy Sci. 71(Suppl. 2): 45.
8. Zang, W. and C. Smith. 1992. Computer simulation of marker-assisted selection utilizing linkage disequilibrium. Theor Appl Genet. 83: 813.

Association of Molecular and Physiological Markers of Immunity with Measures of Mastitis in Periparturient Holstein Cattle

S. C. Kelm,[1] J. C. Detilleux,[1] A. E. Freeman,[1] M. E. Kehrli, Jr.,[2] A. B. Dietz,[2] L. K. Fox,[3] J. E. Butler,[4] I. Kasckovics,[4] and D. H. Kelley[1]

[1]Iowa State University, Ames.
[2]National Animal Disease Center, USDA-ARS, Ames, IA.
[3]Washington State University, Pullman.
[4]University of Iowa Medical School, Iowa City.

DSL-127

Summary and Implications

Relationships between estimated breeding values (EBV) for mastitis measures and molecular markers of immune function were examined. Relationships between EBV for mastitis measures and EBV for 11 *in vitro* immunologic assays also were investigated. Data were available from 137 periparturient Holsteins. Mastitis measures included somatic cell score (SCS), clinical mastitis, and intramammary infection with major or minor pathogens. Molecular markers included alleles of the DRB3 locus of the bovine major histocompatibility complex, IgG2 isotypes, and bovine leukocyte adhesion deficiency (BLAD) genotypes. Immune assays measured neutrophil function, serum immunoglobulins and mononuclear cells, and lymphocyte blastogenesis. A gene substitution model was used to estimate the effects of molecular markers on EBV for mastitis measures. Markers explained up to 40% of the variation in genetic measures of mastitis. Presence of allele *DRB3.2*16* was associated with higher EBV for SCS, thus this allele may indicate greater mastitis susceptibility. Alleles *DRB3.2*8*, $IgG2^b$, and the dominant BLAD allele were associated with increased EBV for clinical mastitis. Alleles *DRB3.2*11*, **23*, $IgG2^a$, and the recessive BLAD allele were associated with decreased clinical mastitis. Significant associations were found between *DRB3.2* alleles and EBV for intramammary infection by major and minor pathogens. Pearson correlation coefficients between EBV for mastitis and EBV for immunologic assays were calculated. Several correlations were significantly different from zero. In general, cows with low EBV for mastitis measures had high EBV for measures of neutrophil function, low serum IgG_1, and low numbers of circulating mononuclear cells.

Introduction

Identification of individuals that are resistant to disease is desirable but difficult in commercial populations. Estimated breeding values (EBV) are based on phenotypic information from an individual and its family members and can be used to identify resistant individuals in lieu of direct challenge. After identification of resistant individuals, the biological causes of disease resistance can be examined. Potential causes include superior immune response capabilities or the possession of single genes with large effects.

The major objective of this study was to measure the association between EBV for mastitis measures and three molecular markers in dairy cattle: the MHC class II DRB3 genotype, the IgG2 allotype, and the BLAD genotype. As a second objective, the genetic association between measures of mastitis prevalence and 11 *in vitro* immunological assays measuring the functional capability of the immune system was investigated.

Materials and Methods

Periparturient Holstein cows (n=137) were available for use in this study from the breeding research herd at Ankeny. Cows were managed as a commercial herd and fed for high production.

Molecular markers. Animals were genotyped at the Class II DRB3.2 locus by using a PCR-RFLP method as described (1). Alleles found at low frequencies (< 2%) were pooled. Immunodiffusion analysis was used to determine IgG2 isotypes as described (2) and genotypes were deduced from isotypes. Animals were genotyped for the BLAD mutation at the CD18 locus by using a PCR-RFLP method as described (3).

Immunologic assays. Weekly blood samples were taken from all cows from 5 weeks before to 5 weeks after calving. Five steers served as laboratory controls. Eleven *in vitro* assays were performed as described (4,5) (Table 1). Immunosuppression (IS) is well documented at calving (6,7), thus immune response was categorized as before IS, during IS, or after IS. Correction for environmental factors was accomplished by calculating EBV for each cow for individual assays, as described (6).

Mastitis measures. Four measures of mastitis were used: somatic cell score (SCS), clinical mastitis (CM), and intramammary infection by major pathogens (IMI Major) or minor pathogens (IMI Minor). Lifetime CM incidence and monthly SCS records were used for each cow. The IMI status of each cow was based on cultural evidence with duplicate quarter milk samples taken at -35, 0, and 35 days relative to calving. The EBV for mastitis measures were computed as described (8).

Statistical analysis. The effect of each molecular marker on the four measures of mastitis was estimated with the following gene substitution model:

$$D_i = MHC_i + IgG2_i + BLAD_i + e_i.$$

In this model, D_i represents the EBV of cow i for SCS, CM, IMI Major, or IMI Minor. Also, MHC_i represents the sum of the following product from m=1 to 12: gene substitution effects for allele m in the DRB3.2 genotype times the number of copies (n) of allele m in the genotype of cow i, where n = 0, 1, or 2. The sum of gene substitution effects is restricted to zero. The variables $IgG2_i$ and $BLAD_i$ are defined similarly. An estimable gene substitution effect was expressed as the difference from the mean of all alleles for each locus (9). Tests of significance were computed using a two-tailed Student's *t* test. Pearson correlations between EBV for SCS, CM, IMI Major, IMI Minor, and EBV for immunological assays before, at, and after IS were calculated. Computations were completed using procedures of SAS software (10).

Results

Table 2 summarizes the frequencies of DRB3.2 alleles, IgG2 allotypes, and BLAD genotypes. Estimates of the gene substitution effects are summarized in Table 3. Note that a positive gene substitution effect is unfavorable, i.e. the allele is associated with undesirable or high EBV for mastitis measures.

The presence of *DRB3.2*16* was associated with a significant increase in EBV for SCS (Table 3). Allele *DRB3.2*8* was associated with an increase in EBV for CM while *DRB3.2*23* and **11* were associated with decreases in EBV for CM. Allele *DRB3.2*24* was shown to have a significant, undesirable effect on IMI Major and *DRB3.2*3* was associated with IMI Minor. The presence of the CD18 mutant allele and $IgG2^a$ was associated with significant decreases in EBV for CM (Table 3).

Tables 4 and 5 summarize the correlations between EBV for immunological assays and EBV for SCS and CM, respectively. Mononuclear cell counts and two neutrophil measures were associated with decreased EBV for SCS (Table 4). Several correlations between EBV for CM and EBV for immune response were significantly different from zero but were not readily interpretable. This was also true for correlations involving EBV for IMI Major and EBV for IMI Minor (data not shown).

Discussion and Conclusions

The results of this study confirm earlier research on potential use of DRB3.2 alleles as markers of disease susceptibility (11,12). The agreement between results of the three reports is noteworthy; *DRB 3.2*8* and *DRB3.2*16* were associated with greater disease susceptibility in all three studies. Furthermore, *DRB3.2*24* was associated with increased EBV for IMI Major in this study and was associated with increase susceptibility to bovine leukosis (12), whereas alleles *DRB3.2*11* and **23* were favorably associated with disease status (12), in agreement with the present study.

Carriers of the mutant CD18 allele had significantly lower EBV for CM; however, sample size was small as only 14 heterozygotes were identified. Further studies involving larger populations are necessary to rule out sampling error. Carriers of $IgG2^a$ also had significantly lower EBV for CM. We believe this is the first report of functional differences in IgG2 allotypes.

Interpretation of the relationship between EBV for immunological assays and EBV for mastitis measures is problematic because a cause-and-effect relationship between the variables cannot be established. It is clear, however, that animals with a favorable EBV for SCS possessed neutrophils with greater functional ability and greater numbers of circulating monocytes (Table 4).

References

1. van Eijk, M. J. 1992. Anim. Genet. 23: 483.
2 Heyermann, H. J. 1992. Mol. Immunol. 29: 1147.
3 Shuster, D. E. 1992. Proc. Natl. Acad. Sci. USA. 89: 9225.
4 Detilleux, J. C. 1995. Vet. Immunol. Immunopathol. 44: 251.
5 Kehrli, M. E., Jr. 1990. Am. J. Vet. Res. 51: 1826.
6 Detilleux, J. C. 1994. J. Dairy Sci. 77: 2640.
7 Cai, T. Q. 1994. Am. J. Vet. Res. 55: 934.
8 Detilleux, J. C. 1995. J. Dairy Sci. 78: 2285.
9 Ostergard, H. 1989. Livest. Prod. Sci. 22: 49.
10 SAS® User's Guide: Statistics, Version 6 Edition. 1989. SAS Inst., Inc., Cary, NC.
11 Dietz, A. B. 1997. J. Dairy Sci. 80: (in press).
12 Lewin, H. A. 1994. Anim. Biotechnol. 5: 183.

Note: For greater detail, the reader is encouraged to see Kelm, S. C., *et al.*, 1997. J. Dairy Sci. 80:1767.
Results can also be seen at the NADC Virtual Conference website http://www.nadc.ars.usda.gov/virtconf/main.htm.

Table 1. Assays of immune function performed in the present study.

Neutrophil assays:
Random migration under agarose
Ingestion of bacteria
Cytochrome C reduction
Myeloperoxidase-catalyzed iodination
Resting chemiluminescence
Stimulated chemiluminescence
Serum concentration:
IgM
IgG_1
IgG_2
Lymphocyte blastogenic response to concanavalin A
Number of blood monocytes

Table 2. Frequencies for DRB3.2 alleles, IgG2 isotypes, and BLAD genotypes.

	Number	Frequencies
DRB3.2 allele		
**8*	52	21.3
**11*	43	17.6
**23*	21	8.6
**24*	21	8.6
**22*	20	8.2
**16*	16	6.6
**27*	13	5.3
**12*	11	4.5
**26*	7	2.9
**3*	7	2.9
**28*	6	2.5
others	27	11.1
IgG2 allotype (genotype)		
AA (*$IgG2^a/IgG2^a$*)	48	39.0
AB (*$IgG2^a/IgG2^b$*)	59	48.0
BB (*$IgG2^b/IgG2^b$*)	16	13.0
BLAD[∂]		
CD18 / CD18	110	88.7
CD18 / D128G	14	11.3
D128G / D128G	0	0.0

[∂] Normal allele: *CD18*. Mutant allele: *D128G*.

Table 3. Gene substitution effects for DRB3.2 alleles of the bovine major histocompatibility complex, for alleles designated by IgG2 isotype, and for alleles at mutation responsible for the bovine leukocyte adhesion deficiency (BLAD) on EBV for mastitis indicators.

	Somatic cell score	Clinical mastitis score	IMI caused by major pathogen	IMI caused by minor pathogen
DRB3.2 alleles				
*8	0.002	0.040†	0.001	-0.031
*11	-0.013	-0.038†	0.006	-0.021
*23	-0.039	-0.070*	0.007	0.041
*24	-0.042	-0.024	0.108**	0.002
*22	0.032	0.028	-0.055	0.028
*16	0.084*	0.001	-0.004	-0.039
*27	0.017	0.042	-0.019	-0.041
*12	0.006	-0.038	-0.001	-0.023
*26	-0.057	0.029	-0.015	0.049
*3	0.016	0.044	-0.033	0.068†
*28	-0.034	-0.005	-0.047	-0.025
others[∂]	0.028	-0.009	0.054†	-0.009
IgG2 alleles				
IgG2A	-0.024	-0.035*	0.002	-0.011
IgG2B	0.024	0.035*	-0.002	0.011
BLAD alleles[b]				
CD18	0.024	0.077*	-0.040	-0.018
D128G	-0.024	-0.077*	0.040	0.018

†P<0.10, *P<0.05, **P<0.01

∂ DRB3.2 alleles designated "others" have frequencies below 2%.

b Normal allele: *CD18*. Mutant allele: *D128G*.

Table 4. Pearson correlation coefficients between EBV for SCS and EBV for immune response measures taken before, during, and after immunosuppression.

	Time relative to immunosuppression		
	Before	During	After
Random migration	-0.158	n.a.[∂]	-0.036
Ingestion	-0.091	n.a.	-0.002
Cytochrome C reduction	-0.080	-0.173[†]	-0.094
Iodination	-0.146	-0.115	n.a.
Chemiluminescence - resting	-0.053	-0.067	n.a.
Chemiluminescence - stimulated	n.a.	-0.259*	0.148
IgG_1	n.a.	0.152	0.263**
IgG_2	-0.108	-0.043	0.138
IgM	0.045	0.127	0.023
Concanavalin A	-0.074	-0.056	-0.056
Mononuclear cells	-0.203[†]	-0.227*	-0.198

[†]P<0.10, *P<0.05, **P<0.01

[∂] Breeding value not available as estimated heritability is less than or equal to zero.

Table 5. Pearson correlation coefficients between EBV for CM and EBV for IR measures taken before, during, and after immunosuppression.

	Time relative to immunosuppression		
	Before	During	After
Random migration	-0.321*	n.a.[∂]	-0.013
Ingestion	0.004	n.a.	0.292*
Cytochrome C reduction	0.202	-0.086	-0.022
Iodination	0.058	-0.035	n.a.
Chemiluminescence - resting	0.204	0.227[†]	n.a.
Chemiluminescence - stimulated	n.a.	-0.171	0.080
IgG_1	n.a.	0.215[†]	0.373**
IgG_2	0.060	-0.062	0.194
IgM	0.167	0.029	0.049
Concanavalin A	-0.165	-0.127	0.143
Mononuclear cells	-0.205	-0.052	-0.062

[†] P<0.10, *P<0.05, **P<0.01

[∂] Breeding value not available as estimated heritability is less than or equal to zero.

Preliminary Report on the Developmental Potential of Oocytes Recovered from Postpartum Dairy Cows

J. N. Caamaño, graduate research assistant;
M. L. Pugh, graduate research assistant;
L. L. Timms, associate professor; and
C. R. Youngs, associate professor of animal science

DSL-128

Summary and Implications

Poor reproductive efficiency of postpartum dairy cows is a major concern in dairy cattle operations. The objective of this experiment was to study the developmental competence of oocytes recovered from postpartum dairy cows. Postpartum dairy cows were subjected to ultrasound-guided follicular aspiration two times per week for 4 weeks beginning approximately 30 days after calving. Harvested oocytes were matured, fertilized, and cultured *in vitro*. We were able to produce blastocyst stage embryos from of all the cows subjected to this procedure. Once refined, this procedure will enable dairy cattle producers to produce live calves from postpartum dairy cows at a time when they ordinarily make no reproductive contribution to the herd. Further, by altering follicular growth patterns via ultrasound-guided oocyte retrieval, it may be possible to enhance reproductive efficiency of postpartum dairy cows by stimulating the ovulation of higher quality oocytes. Although these results are preliminary, we believe great potential exists for application of these technologies under field conditions. Future experiments and statistical analysis of data from this study will help us to understand better the reproductive potential of postpartum dairy cows.

Introduction

One continual quest of any dairy cattle manager is to increase animal productivity. The genetic improvement tools of artificial insemination and embryo transfer have been successfully used to achieve increased productivity. Other reproductive technologies may prove useful to further increase the genetic merit of dairy herds. Researchers elsewhere have demonstrated the ability to obtain offspring from oocytes harvested from pregnant cows, clinically infertile females, and prepuberal calves by using ultrasound as a tool to guide the collection of oocytes directly from a female's ovaries. Oocytes harvested in this manner can be matured, fertilized, and developed under *in vitro* conditions.

One of the challenges faced by managers of high-producing dairy cows is establishing a pregnancy in the postpartum period (the period after calving). The postpartum period has been the subject of numerous studies regarding the initiation of ovarian activity and reestablishment of estrous cycles. However, information concerning the production of embryos from postpartum cows is sparse. Using ultrasound-guided follicular aspiration, coupled with *in vitro* fertilization (IVF), it may possible to produce embryos from oocytes harvested during the postpartum period.

The objective of this experiment was to study the developmental competence of oocytes recovered from postpartum dairy cows.

Materials and Methods

Experimental animals. Ten multiparous Holstein dairy cows, 3 to 7 years of age, were selected on the basis of calving dates (close proximity to one another) and were randomly assigned to one of two experimental groups. One group (n=5) was assigned to ultrasound-guided follicular aspiration twice a week (from approximately postpartum day 30 to day 60). The second group (n=5) served as a non-aspirated control. Body weights (kg) and body condition scores (on a scale of 1 [emaciated] to 5 [obese]) were recorded at the beginning and the end of the experiment.

Ultrasound and oocyte retrieval equipment. An Aloka 500 Micrus ultrasound console was used with a 5 MHz convex array transducer housed in a 60-cm plastic handle containing a stainless steel needle guide. For oocyte retrieval, a 17 gauge, 55-cm length single lumen needle was used to aspirate the ovarian follicles with the aid of a regulated vacuum pump (K-MAR-5000) that produced a flow rate of 22 ml/min.

Procedures. In both groups, ultrasound was performed daily to monitor changes in ovarian structures (e.g., follicles, corpora lutea). Ultrasound monitoring began when cows were at approximately postpartum day 25. Each animal was restrained in a squeeze chute, and the perineal region was washed. The ultrasound transducer (covered with a latex condom) was inserted inside the vagina, and each ovary was visualized by gentle repositioning (via rectal palpation) to the point just across the vaginal wall from the ultrasound transducer. All ultrasonography was recorded on video to enable

subsequent reevaluation and measurement of ovarian structures.

On the day of oocyte retrieval, designated cows were handled in a similar manner except that epidural anesthesia (2% lidocaine hydrochloride) was administered to prevent contractions of the tail and rectal muscles. After the ovary was repositioned to enable visualization, the retrieval needle (previously introduced into the needle guide) was inserted through the vaginal wall and into the ovarian follicles. Vacuum pressure was applied to aspirate the follicles, and all follicles ≥3 mm in diameter were aspirated.

After follicular aspiration was completed, the Em-Com filter containing the harvested oocytes was rinsed twice, and its contents were placed into a gridded petri dish to search for oocytes by using a stereomicroscope. Oocytes were evaluated and handled as described below.

In addition to the collection of oocytes and ultrasound examinations, blood samples were taken from each cow three times per week during the entire experiment. Blood was collected into heparinized tubes (to prevent clot formation), centrifuged, and plasma was collected and frozen for subsequent analysis of hormone concentrations.

Oocyte classification and in vitro *methods.* The cumulus oocyte complexes (COC) harvested from each cow were divided into two groups based on oocyte morphology. Oocytes with even cytoplasmic pigmentation and with greater than two complete layers of cumulus cells were classified as excellent/good. Oocytes with less than three complete layers of cumulus cells (or partially/totally denuded) and/or with an uneven cytoplasmic pigmentation were classified as fair/poor.

Oocytes from the same donor, grouped by quality as indicated above, were matured in Tissue Culture Medium 199 supplemented with 10% fetal bovine serum at 39°C for 22–24 hours in an atmosphere of 5% CO_2 in humidified air. Frozen-thawed semen from a single bull was prepared for *in vitro* fertilization (IVF) by using Brackett-Oliphant (BO) medium, and COC were placed in fertilization drops (5×10^6 sperm/ml) consisting of BO medium supplemented with 10% bovine serum albumin (BSA). Cumulus cells were mechanically removed immediately after the 6-hour insemination period. Presumptive zygotes were cultured for 7 days in CR1AA medium with 0.2% BSA (and without co-culture cells) at 39°C in an atmosphere of 5% CO_2, 5% O_2, and 90% N_2. Blastocyst formation rate was determined on day 7 (day 0=onset of insemination), and blastocyst cell numbers were determined by DNA-specific cell staining. At the same time that oocytes from postpartum cows were being matured, fertilized, and cultured *in vitro*, ovaries from slaughterhouse cows were aspirated and harvested oocytes were handled following the same procedures as described above. The slaughterhouse oocytes served as a quality control of our IVF system.

This experiment is still in progress. Data presented here are preliminary and have not been statistically analyzed. Only simple arithmetic means are given. Reproductive performance of these cows, as well as their levels of milk production, will be monitored and analyzed at the end of the experiment.

Results and Discussion

Cow body weight remained essentially the same from the beginning (648 kg) to the end (643 kg) of the experiment. Similarly, body condition score was similar at the beginning (3.4 score) and the end (3.5 score) of the experiment. These data suggest that nutritional management of the experimental animals was appropriate.

A total of eight aspiration sessions was performed in each treatment cow. The results of the oocyte retrievals for each cow are presented in Table 1. The overall efficiency of oocyte recovery was 53.7%. The number of follicles available for aspiration varied considerably among animals.

The results obtained with oocytes collected from slaughterhouse ovaries are presented in Table 2. The results showed that the IVF system was performing well, so any poor results obtained from the postpartum cow oocytes can not be directly attributed to a potential deficiency in the IVF system.

The quality of oocytes harvested from each cow and the cleavage and blastocyst formation rates after *in vitro* maturation/*in vitro* fertilization/*in vitro* culture (IVM/IVF/IVC) are shown in Table 3. Overall, oocytes classified as excellent/good produced higher cleavage and blastocyst formation rates than oocytes classified as fair/poor. This result was not unexpected, as the same occurs with slaughterhouse oocytes. Although variation among cows in the quality of oocytes was evident (Table 2), we were able to produce blastocysts from of all the cows subjected to ultrasound-guided follicular aspiration during days 30 to 60 postpartum. (Blastocyst stage embryos are those most often used for embryo transfer to a recipient female).

We are encouraged by these preliminary results. The ability to produce a blastocyst from oocytes recovered prior to day 60 postpartum gives producers the option to produce a live calf (via transfer of IVM/IVF/IVC embryos) before this female is mated via artificial insemination to establish the next pregnancy.

Our research on the postpartum cow has two main implications. One is to produce an extra live calf without delaying the normal time of breeding for the postpartum dairy cow. The second implication is to study the effect of altering (via ultrasound-guided oocyte retrieval) follicular growth patterns in postpartum cows to assess if this may

be used to enhance reproductive efficiency in the postpartum period.

By removing oocytes from the cow, it is possible to study them independent of the hormonal and nutritional environment of the cow from which they came. Poor reproductive efficiency of postpartum cows may be due to hormonal imbalances, metabolic problems, or other factors not associated with the oocyte. This study will enable us to ascertain the role of the oocyte in the reproductive process in postpartum cows.

The data obtained from analysis of hormonal profiles and subsequent statistical analysis of all data gathered will help us to better understand the possibilities of how to apply these technologies in the field. Future studies will need to include the transfer of embryos to recipient females and the use of exogenous hormones to increase the number and size of follicles present in the ovaries of postpartum cows.

Acknowledgments

The authors gratefully acknowledge Cindy Achen, Paul Amundson, and John Kent for their assistance with the experimental animals; Howard Tyler for access to laboratory space at the farm; Cari Fleming, Kristin Sieren, and Almudena de Arriba for assistance with ultrasound exams and oocyte retrievals; and Donna Johnston for typing the report.

Table 1. Transvaginal ultrasound-guided oocyte recovery from dairy cows on days 27 to 61 post-partum.

Cow #	No. follicles aspirated	No. (%) oocytes recovered
1304	87	43 (49.4)
1126	61	34 (55.7)
1232	79	54 (68.3)
1112	18	9 (50.0)
1204	68	28 (41.0)
TOTAL	313	168 (53.7)

Table 2. *In vitro* embryo production from oocytes obtained from slaughter house ovaries.

No. of oocytes	No. (%) of oocytes that cleaved	No. (%) of oocytes that formed blastocysts
199	150 (75.4)	50 (25.1)

Table 3. *In vitro* embryo production from oocytes harvested from post-partum dairy cows.

Cow #	Oocyte quality	No. (%) oocytes	No. (%) of oocytes that cleaved	No. (%) of oocytes that formed blastocysts
1304	excellent/good	19 (44)	10 (52.6)	4 (21.1)
	fair/poor	24 (56)	8 (33.3)	1 (4.2)
	TOTAL	43	18 (41.9)	5 (11.6)
1126	excellent/good	9 (26)	1 (11.1)	0 (0.0)
	fair/poor	25 (74)	2 (8.0)	2 (8.0)
	TOTAL	34	3 (8.8)	2 (5.9)
1232	excellent/good	0 (0)	0 (0.0)	0 (0.0)
	fair/poor	54 (100)	2 (3.7)	1 (1.9)
	TOTAL	54	2 (3.7)	1 (1.9)
1112	excellent/good	6 (67)	2 (33.3)	2 (33.3)
	fair/poor	3 (33)	0 (0.0)	0 (0.0)
	TOTAL	9	2 (22.2)	2 (22.2)
1204	excellent/good	5 (18)	2 (40.0)	0 (0.0)
	fair/poor	23 (82)	5 (21.7)	1 (4.3)
	TOTAL	28	7 (25.0)	1 (3.6)
TOTAL	excellent/good	39 (23)	15 (38.5)	6 (15.4)
	fair/poor	129 (77)	17 (13.2)	5 (3.9)
	GRAND TOTAL	168	32 (19.0)	11 (6.5)

Evaluation of Accuracy and Characterization of Estrus Activity as Monitored by an Electronic Pressure Sensing Mounting System for Estrus Detection in Dairy Cattle

Stacia Piggott, Deb Fitkin, Andrea Steffens, undergraduates, and Leo Timms, extension dairy specialist, Animal Science Department

DSL - 129

Summary and Implications

Heat detection efficiency in cows (71 cows, 136 estrous cycles) using an electronic mount or pressure sensing system was 89% and 89% (May to October 1996 and November to April 1997) while heat detection accuracy was 88% and 78%, respectively, with a visual detection efficiency of 68%. Heat detection efficiency and accuracy in heifers (93 heifers and cycles) was 92% and 93% for electronic mount with a visual efficiency of 73%. Overall means for estrus parameters for cows (two time periods) and heifers as measured by the electronic system were 4.1, 3.6, and 9.9 mounts/estrus, 8, 4, and 9.6 hrs estrus duration (first to last mount), and 14, 9.6, and 33 total seconds spent mounting/estrus. Average time required for the electronic system for cows was 13 minutes/day with patch application taking about seven minutes/cow. These results indicate that the electronic pressure sensing mount system was an effective estrus detection tool for both cows and heifers, took minimal time, and may provide a viable alternative to visual detection depending on cost: benefit analysis for individual herds.

Introduction

Successful reproductive management is essential for dairy farm financial maximization. Weaknesses in estrus detection have been shown to be the major deterrent to optimizing reproductive efficiency and is the major reason for excessive days open and lost income, especially in herds using artificial insemination. Research has substantiated that animals standing to be mounted is the cardinal sign of estrus, and breeding based on observing this results in significantly higher conception rates as compared to secondary signs. A problem exists in observing mounting as many animals, especially high producing animals or animals undergoing a stressor (heat, footing, etc.) have decreased estrus durations and intensities. This coupled to the fact that 67% of mountings occur from late evening to early morning make estrus detection a formidable task. Simple pressure sensing mount detectors and tail head chalking have been used as an aid to enhance mounting recognition. However, there are false positives associated with these, and they can't pinpoint the time of mountings. An electronic pressure sensing mounting system was developed to not only detects mounts electronically, but also to supply information on time and duration of mountings through a computer software program. The objectives of this study were to evaluate the efficiency and accuracy of estrus detection using the electronic mount system compared to visual detection, and characterize the estrus activity of dairy cows and heifers using data from the electronic system.

Materials and Methods

Cows

Lactating dairy cows at the Iowa State University Dairy Farm between 30 to 45 days postpartum were selected to be fitted with the electronic mounting transmitter (Heatwatch (HW), DDX, Inc., Boulder, CO). This battery-operated reusable pressure sensing radio frequency transmitter (2" x3") was fitted into a durable tightly woven, nylon envelope that is sewn to a 10" x 8" web patch with a 12" tail strap. This patch was then sprayed with glue and allowed to partially solidify while the tailhead of the animal was being sprayed with the same glue. The mesh patch was placed on the tailhead and smoothed out to ensure that it was completely attached and tail strap was glued to the tail.

After the patch and tail strap were applied each cow number was entered into the HW software in the farm computer with the corresponding transmitter number from the unit on her back. This allowed for easy identifications as to which animal was being mounted. It was also possible to enter in other information for each animal such as a calving date and housing area.

The computer was checked daily for mounting activity, and each cow's total mounts, including duration of each mount, were recorded. Visual heat detection was also recorded although visual detection and artificial insemination were completed by farm personnel. All cows were checked daily to see if any patches had been lost or were loose, and proper maintenance was completed. The total time (checking the computer and patch maintenance) was recorded in a journal to estimate the average time spent with the HW system.

Secondary observations of estrus were also completed. Vaginal mucus electrical conductivity was measured daily using a Estron vaginal probe (Estrogenix, Inc.) on all cows prior to first breeding and 18-24 days post breeding. Cows were palpated twice/week prior to first breeding by a veterinarian. The uterus was scored on involution (1-normal, 3-abnormal) and tone (1-turgid, 3-flaccid), and each ovary was palpated to detect any growing follicles or other structures and their relative size. At >30 days post-breeding the cows were palpated to determine pregnancy status. If the animal was confirmed pregnant the transmitter was removed.

Estrus duration was defined as interval between first and last HW mount. Cow data was split into two time

periods for analysis [May to October(SF) and November to April(WS)] corresponding to seasonal differences in housing or the amount of time confined to a tie stall as compared to outside free roaming.

Heifers

Each month, HW transmitters were applied to heifers that had been synchonized using an injection of prostaglandin (Lutalyse, Upjohn, Inc.) or Norgestomet implant (Synchromate B, CEVA Lab, Inc.) with transmitters applied on the day of prostaglandin injection or implant removal. Visual observation for estrus was performed by farm personnel. Transmitters were kept on heifers until inseminated, 3 weeks post application, or until they became detached. This trial was conducted over a one year period.

Results and Discussion

Cows

A total of 71 cows and 136 heats were monitored (26 cows, 44 estrus cycles for SF; 45 cows, 92 estrus cycles for WS). Efficiency and accuracy of estrus detection for HW and visual are shown in Table 1. HW estrus detection efficiency and accuracy for SF, WS, and combined data were 89 and 88%, 89 and 78%, and 89 and 84%, respectively, while visual detection efficiency was 68% for SF. HW estrus detection efficiency was significantly better than visual. HW efficiency for SF and WS was similar, but accuracy was 10% less for WS compared to SF. During WS, cows are housed inside except for three hours/day or when they were in the holding pen waiting to be milked (3x) as compared to SF where cows are only inside three hours/day. Accuracy decreased or false positives increased during WS due to increased potential to rub the transmitter on posts separating the stalls due to an increased amount of time spent in the tie stalls (18 hrs vs 3 hrs). Characterization of different estrus parameters are shown in Table 2. Overall means for SF and WS were 4.1 and 3.6 mounts/estrus, 8 and 4 hours estrus duration, and 14 and 10 total seconds spent mounting/estrus. All parameters for WS were less than SF, again reflecting the decreased time available for free movement and mounting. The amount of time required/day for checking the computer and performing transmitter maintenance was 13 minutes/day with initial transmitter application or re-application taking about seven minutes per cow.

Heifers

A total of 93 heifers and estrous cycles were monitored. Heat detection efficiency and accuracy using HW were 92 and 93%, with a visual efficiency of 73% (Table 1). Means for estrus parameters (Table 2) as measured by HW were 9.9 mounts/estrus, 9.6 hours estrus duration, and 33 total seconds spent mounting/estrus. HW efficiency in heifers (similar to cows) was significantly better than visual. All estrus parameters for heifers were higher compared to cows. Heifers were housed in three sided loafing barns with unrestricted access to outside lots and this accounts for higher number of mounts, and longer duration of estrus as well as total mounting times.

There were some problems experienced with patch or transmitter adherence. Application of transmitters on very humid or rainy days (heifers) led to decreased attachment times due to the inability of the glue to adhere or bind to a wet surface. Also, during winter, decreased attachment times were seen due to longer hair coats. Clipping the hair area where the patch was applied significantly improved adherence times.

Overall, the electronic pressure sensing mount system for estrus detection was an effective estrus detection tool for both cows and heifers, and took minimal time or labor. This system may provide a viable alternative to visual detection and should be evaluated using partial budgets on an individual farm basis.

Table 1. Efficiency and accuracy of estrus detection as measured by an electronic pressure sensing mounting system (Heat Watch) or visual detection in dairy cows and heifers.

Trial*	Parameter%	Heat Watch	Visual
SF	Efficiency	89	-
	Accuracy	88	68
WS	Efficiency	89	-
	Accuracy	78	-
C	Efficiency	89	-
	Accuracy	84	-
Heifers	Efficiency	92	73
	Accuracy	93	-

*SF = May to November, 26 cows, 44 estrus cycles;
WS = December to April, 45 cows, 92 estrus cycles;
C = combined cow data for both periods;
Heifers = 93 estrus cycles over a one year period.

Table 2. Characterization of different estrus parameters in dairy cows and heifers as measured with an electronic pressure sensing mounting system.

Trial*	Mounts/ Estrus	Estrus duration (hrs) (first-last mount)	Time Spent Mounting/ Estrus (dec).
SF	4.1	8	14
WS	3.6	4	10
Heifers	9.9	9.6	33

* SF = May to November, 26 cows, 44 estrus cycles;
WS = December to April, 45 cows, 92 estrus cycles;
Heifers = 93 estrus cycles over a one year period.

Dairy-Related Research Conducted by the Nutritional Physiology Group

Jerry W. Young, professor of animal science;
Donald C. Beitz, distinguished professor of Agriculture; and Gary L. Lindberg, assistant professor of animal science

DSL-130

Six staff members employed by the Department of Animal Science are primary contributors to dairy-related research in the Nutritional Physiology Group of the Department of Animal Science. They are Donald C. Beitz, Lee H. Kilmer, Gary L. Lindberg, Leo L. Timms, Howard D. Tyler, and Jerry W. Young. In addition, Jesse P. Goff, Ronald L. Horst, Brian J. Nonnecke, and Timothy A. Reinhardt are employed by the USDA and are located at the National Animal Disease Center just outside Ames. These USDA employees have collaborator appointments in the Department of Animal Science and make major contributions to dairy research. An overview of their research appears elsewhere in this Dairy Report.

The broad research mission of the Nutritional Physiology Group is to study the physiology and biochemistry of nutrient use for animal production processes and for the maintenance of animal health and to produce significant research results that can be used by the dairy industry.

Milk Production

Projects relating to applied milk production problems include an evaluation of pedometers for reproductive management in dairy cattle, evaluation of barrier products to prevent dry-period mastitis, and evaluation of alternative therapeutics for mastitis. In the future, potentially useful ideas for improvement of profitability of the modern dairy production enterprise will continue to be tested.

The effectiveness of raising calves for herd replacements has major long-term effects on the profitability of milk production. Therefore, we have projects oriented toward improving the survival of dairy calves to minimize economic losses. There are studies to examine the role of abnormal maternal conditions on neonatal immune functions, to determine the potential for chronic alterations of glucose regulatory mechanisms through acute intervention in the neonatal period (i.e., neonatal imprinting), and to develop strategies for increasing survivability of calves born prematurely. Future studies will include characterization of changes in oxygen status, acid-base balance, and cellular immune status in the neonatal period; determination of the effects of hypoxia or hyperoxia on immune function parameters; and elucidation of the critical oxygen threshold for respiratory burst activity.

Other ongoing research includes determination of milk protein and fatty acid compositions in milk, including quantification of variation of different milk proteins and fatty acids in milk, and relationships of these components to each other. Relationships of mitochondrial DNA genotypes to milk production are being investigated. There is research on understanding how DNA is targeted to cells through receptor-mediated uptake and on the fate of the DNA after uptake. Future research will focus on applications of DNA targeting procedures to use genes to modify animal metabolism and production.

Animal Health

Lactation ketosis is a significant metabolic disease of lactating dairy cows and costs dairy farmers millions of dollars each year. The occurrence of ketosis seems to usually be preceded by fatty liver, which is a hidden gateway disease that remains undiagnosed until some secondary condition becomes apparent. We have expended much effort in development and characterization of a realistic model of lactation ketosis that is being used to determine factors that influence susceptibility to ketosis and to trace the sequence of metabolic events that lead to a diagnosis of clinical ketosis. A major focus is now being placed on development of preventatives and treatments for fatty liver and ketosis. Evaluation of glucagon, the hormone that generally acts opposite of insulin, was the major focus of a USDA project that was completed recently. The expression of rate-limiting genes for glucose synthesis in the liver also is being evaluated. This research emphasis will continue as we seek industrial collaboration to provide a marketable preventative or cure for fatty liver and lactation ketosis.

Parturient paresis, better known as milk fever, also is a major disease that affects early lactation dairy cows. A major amount of research in this area has been coordinated through collaborators at the National Animal Disease Center, but much of the research involved graduate students obtaining M.S. or Ph.D. degrees in nutritional physiology. Such efforts have been very productive in increasing understanding of this disease and will continue to be emphasized.

Other focuses on animal health include effects of nutrition on immune status and the relationships of mitochondrial DNA genotype to susceptibility to diseases. The major nutritional agents tested to date for effects on the immune system have been vitamins A and D and their

derivatives. Future research will focus on elucidating factors that affect the immune system and on learning how to use those factors to greatly decrease the incidence and severity of diseases affecting dairy cattle.

Human Health

A major focus has been to develop techniques to use an enzyme, cholesterol reductase, or acceptable bacteria containing the enzyme to decrease the cholesterol content of milk and dairy products, and also to help decrease concentrations of cholesterol in human blood. The major products of dairy production are food products to meet human needs and desires. It is very important that consumption of those products promote the long-term health and well-being of people. We have conducted studies involving feeding cattle unsaturated lipids to increase the unsaturation of fats in milk and meat and to decrease the amount of fat in milk. Medical recommendations and consumer desires are for more polyunsaturated and monounsaturated fats and less total fat in human diets, and our research will continue to emphasize production of more desirable foods. Research also dealt with nutrients, including fat, protein, calcium, and vitamin D, that can alter dietary regulation of blood cholesterol in humans. A strong emphasis will continue toward modifying milk and meat to produce foods that will be more healthful for people consuming these foods.

Prepartal Energy Intake of Dairy Cows Affects Severity of Fatty Liver and Susceptibility to Ketosis

A. R. Hippen, research assistant,
J. W. Young, professor of animal science;
P. She, research assistant;
G. L. Lindberg, assistant professor;
and D. C. Beitz, distinguished professor of agriculture

DSL-131

Abstract

To observe the effects of prepartal energy intake on fatty liver and ketosis in dairy cows postpartum, 24 multiparous Holstein cows were assigned to one of two treatments. During the last 30 days before calving, cows were fed either a normal dry cow diet (control) or a diet calculated to provide 50% greater energy intake from carbohydrates (overfed). After calving, livers of overfed cows had higher concentrations of triacylglycerol (TAG) and lower concentrations of glycogen than did livers of control cows. From days 6 to 13 of lactation, concentrations of glucose, urea nitrogen, and insulin in blood were lower for overfed cows than for control cows, and concentrations of ß-hydroxybutyrate (BHBA) and nonesterified fatty acids (NEFA) were greater in blood from overfed cows than from control cows. Of the 12 overfed cows, four exhibited ketonemia, ketonuria, acetone-like breath, and depressed appetites. Two of the overfed cows became spontaneously ketotic and required treatment before day 14 of lactation. No ketonemia was observed in any of the cows that had less than 5% TAG in the liver. The incidence and severity of ketonemia increased in cows as concentrations of TAG increased in livers. Body condition scores of cows at calving were correlated poorly with incidence of fatty liver after calving. Increased energy intake during the dry period, however, caused increased severity of fatty liver and susceptibility to ketosis.

Introduction

Metabolic disorders in dairy cows are generally related to nutrition and frequently become apparent shortly after parturition. In particular, fat cow syndrome is a peripartal disease often accompanied by dystocia, retained placenta, milk fever, decreased resistance to infection, and ketosis. Fat cow syndrome in a dairy herd is apparent by the presence of extremely over-conditioned dry cows.

Over-conditioning of dairy cows during the dry period can lead to the development of fatty liver because excess body fat is mobilized to meet the energy demands of parturition and early lactation. Fatty acids mobilized from adipose tissue that are in excess of the liver's capacity to export fat will accumulate in the liver and create fatty liver. This accumulation of fat in the liver can cause liver TAG concentrations to be as high as 30% of liver wet weight compared with normal concentrations of 1 to 4%. Also, the severity of fatty liver is thought to be related to the incidence of ketosis in early-lactation dairy cows.

The objective of this research was to observe the effects of excessive energy intake during the dry period on development of fatty liver and susceptibility of dairy cows to ketosis after calving.

Materials and Methods

During the last 30 days before calving, 24 multiparous Holstein cows were fed either according to NRC recommendations for energy intake (control) or offered excess grain in a quantity that would increase daily energy intake by 50% (overfed, Table 1). Body condition scores (BCS) were recorded 15 days prior to parturition and at 1 and 14 days in milk (DIM). Cows were weighed at 1, 6, and 14 DIM.

Table 1. The diets fed to control and overfed cows during prepartum period.

Ingredient	Control	Overfed
	---------- kg/d ----------	
Corn silage	1.6	1.6
Concentrate[1]	0.9	0.9
Alfalfa hay	0.6	0.6
Corn gluten feed	0.4	0.4
Oat silage	0.4	0.4
Cracked corn	--	5.5
Grass hay	ad libitum	ad libitum

[1]Concentrate consists of rolled corn (72%), soybean meal (24%), Na_2CO_3 (1.1%), Ca_2PO_4 (1.1%), MgO_2 (0.6%), NaCl (0.6%), Ca_2CO_3 (0.3%), and X-Cel Ruminant Trace Mineral Premix ®(0.3%).

Liver samples collected at 6 and 14 DIM by puncture biopsy were analyzed for content of TAG and glycogen. To measure carbohydrate status, plasma samples collected daily from 7 to 14 DIM were analyzed for concentrations of glucose, urea nitrogen, and insulin. Susceptibility to ketosis was indicated by analysis of plasma samples for concentrations of BHBA and NEFA.

A cow was considered to be clinically ketotic if, as measured by Ketostix® (Miles, Inc.), her plasma ketones

were greater than 40 mg/dl, ketones were evident in her urine, and her appetite was depressed without evidence of a complicating factor such as a displaced abomasum or infection.

Data were analyzed as a complete randomized block and Pearson correlation coefficients were calculated by GLM procedures of SAS.

Results

Body weights, though numerically higher for overfed than for control cows, were not statistically different at any time measured (Figure 1A). At 15 days before calving, BCS were greater for overfed than for control cows; however, by 1 day postpartum, BCS were not different between the two groups (Figure 1B).

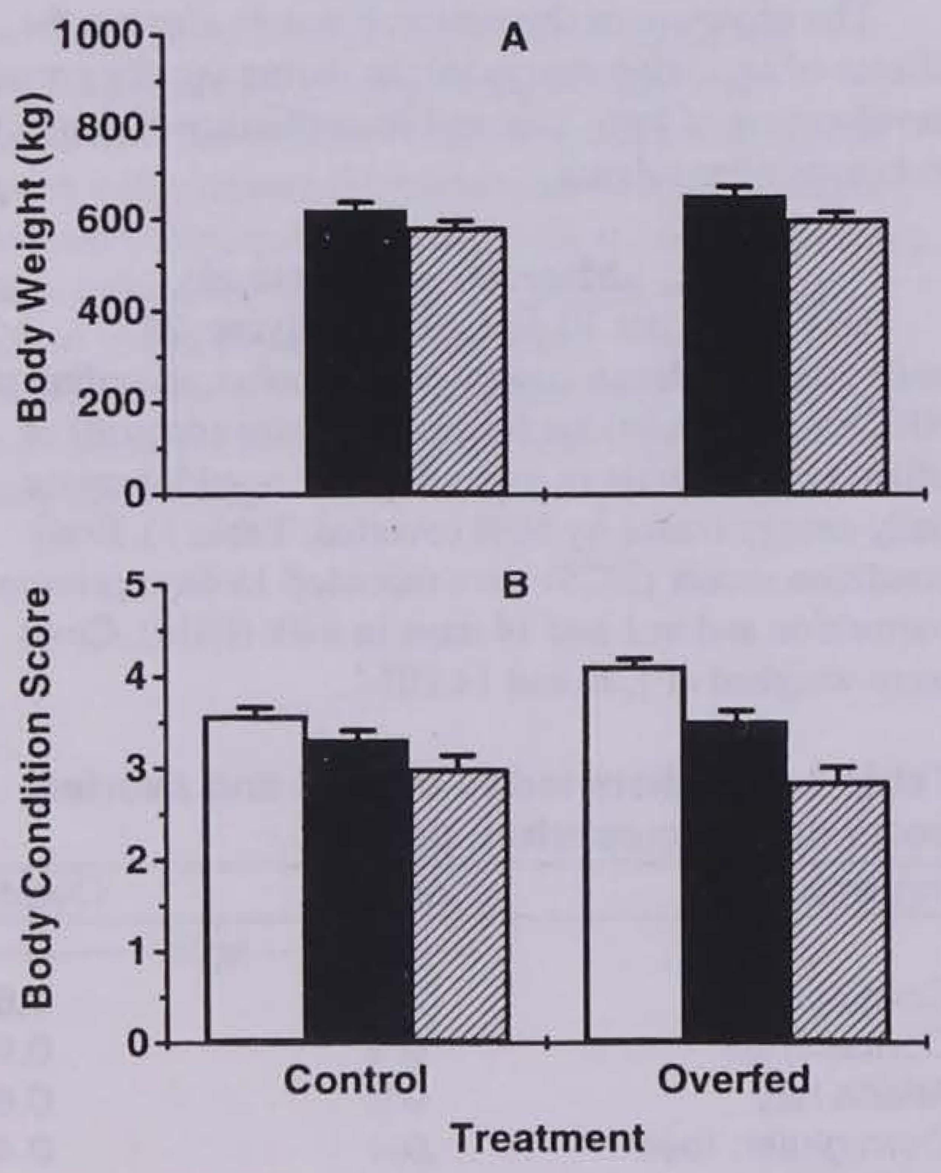


Figure 1. Body weights (A) and body condition scores (B) of control and overfed cows 15 days prepartum ( ) and on days 1(and 14 (postpartum. Effects in model; body condition score, treatment P<0.003.

Liver TAG concentrations were greater in overfed than in control cows at both 6 and 14 DIM (Figure 2A). Six days after calving, livers of overfed cows contained 12.1% TAG compared with 2.7% TAG (P<0.01) in livers of control cows. Concentrations of TAG increased in livers of both groups of cows at 14 DIM to 13.7 and 2.9% for overfed and control cows, respectively. Conversely, liver glycogen concentrations were lower in overfed than in control cows (1 vs. 2.3%, P<0.01) and increased slightly in both groups at 14 DIM (Figure 2B).

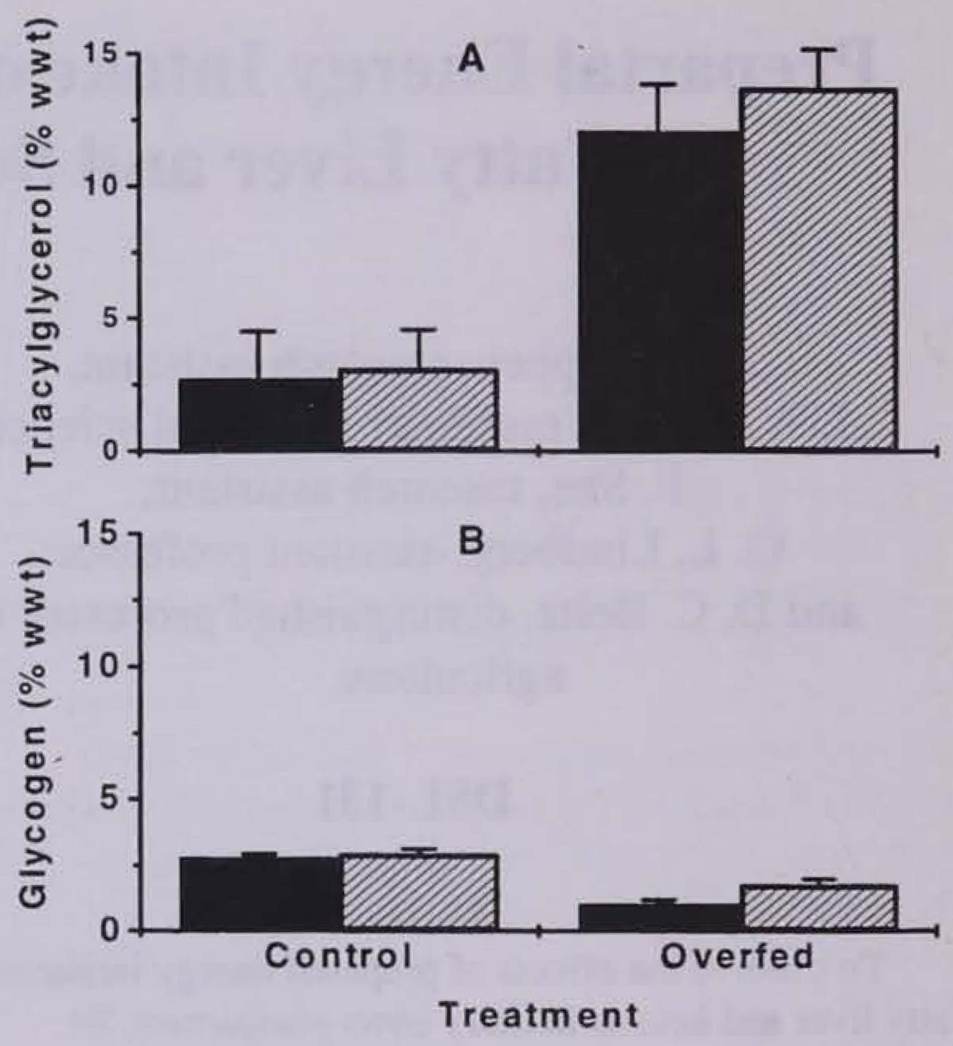


Figure 2. Concentrations of triacylglycerol (A) and glycogen (B) in livers of control and overfed cows on days 6 (and 14 (postpartum. Effects in model; triacylglycerol, treatment P<0.002; glycogen, treatment P<0.0006.

During the second week after calving, concentrations of plasma glucose, urea nitrogen, and insulin were decreased in overfed cows compared with control cows (P<0.05, Figure 3), indicating decreased carbohydrate

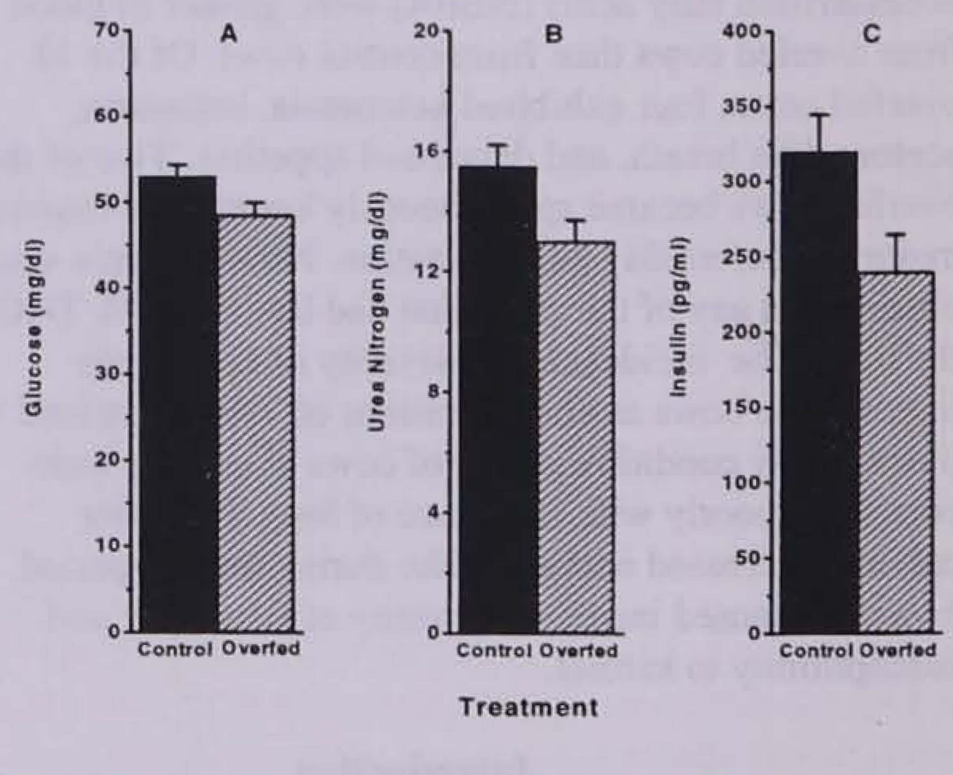


Figure 3. Plasma glucose (A), urea nitrogen (B), and insulin (C) concentrations in control (and overfed (cows. Effects in model; glucose, treatment P<0.04; urea nitrogen, treatment P<0.02; insulin, treatment P<0.03.

status. During the second week postcalving, concentrations of the lipid-based plasma metabolites BHBA and NEFA were greatly increased by overfeeding during the dry period (Figure 4).

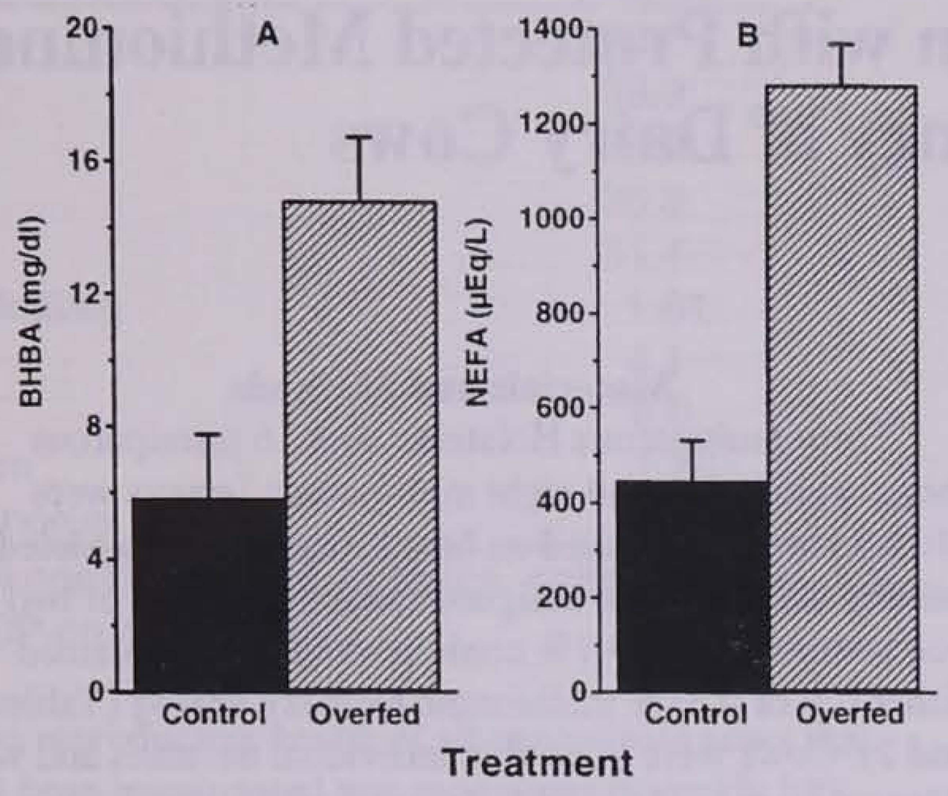


Figure 4. Plasma ß-hydroxybutyrate (BHBA, A) and nonesterified fatty acid (NEFA, B) concentrations in control (▮ and overfed (▨ cows. Effects in model; BHBA, treatment, P<0.004; NEFA, treatment P<0.001.

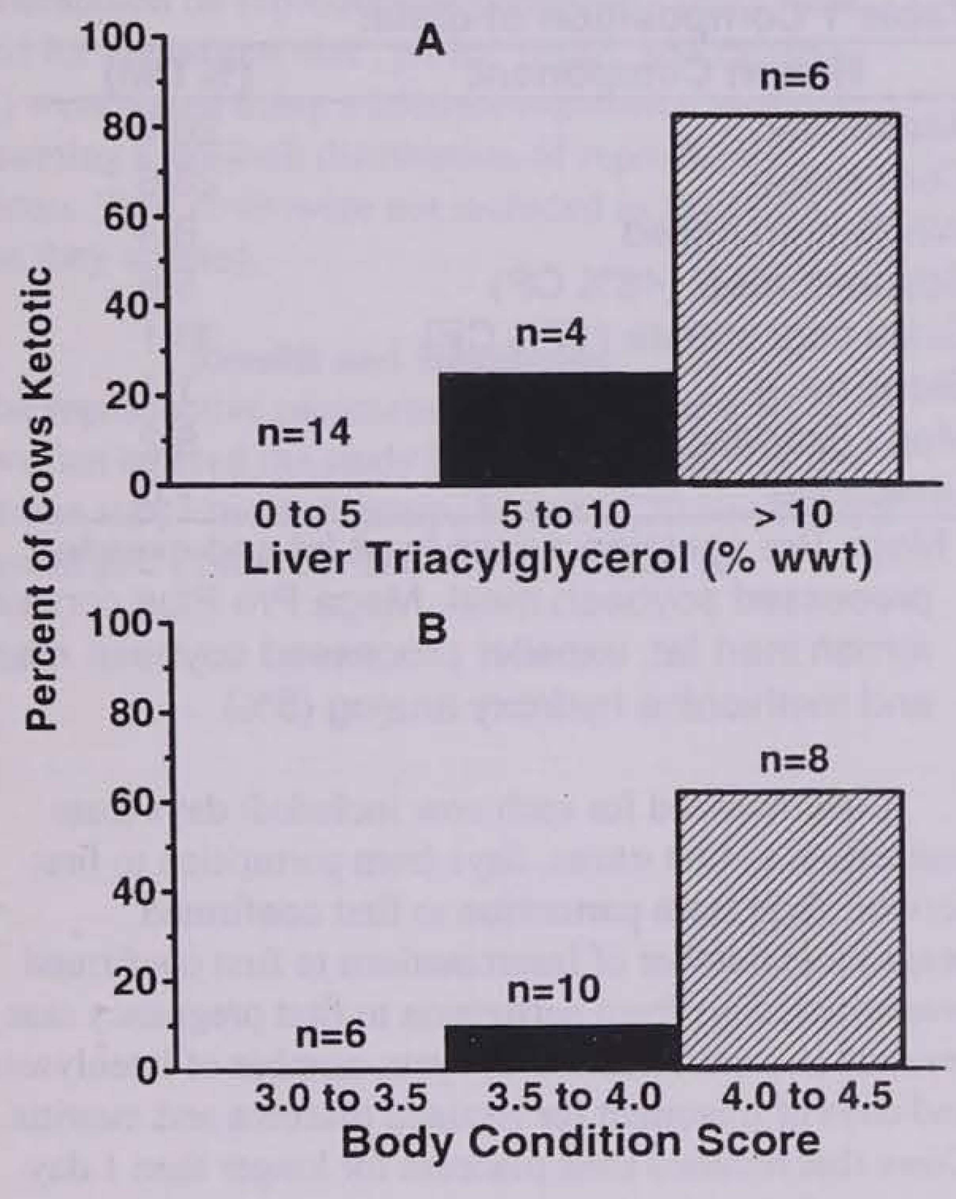


Figure 5. Incidence of ketosis by percent of liver triacylglycerol (A) and body condition score (B) at day -15 prepartum.

None of the control cows became clinically ketotic during the first 2 weeks of lactation, but six of the overfed cows became clinically ketotic during this time. Additionally, one other overfed cow and one control cow exhibited ketonemia as evidenced by plasma BHBA concentrations greater than 12 mg/dl. The number of cows becoming ketotic was greater as both liver TAG and BCS increased (Figure 5).

Because of rapid loss of body condition scores during calving, BCS 1 day after calving were poorly correlated to liver TAG. Body condition scores 15 day before calving, however, were more highly correlated to liver TAG concentrations (Figure 6). Furthermore, correlations of plasma ketone bodies were more highly correlated to BCS 15 days before calving than 1 day after calving (R^2 = 0.28 vs. 0.43 for day -15 and 1, respectively, data not shown).

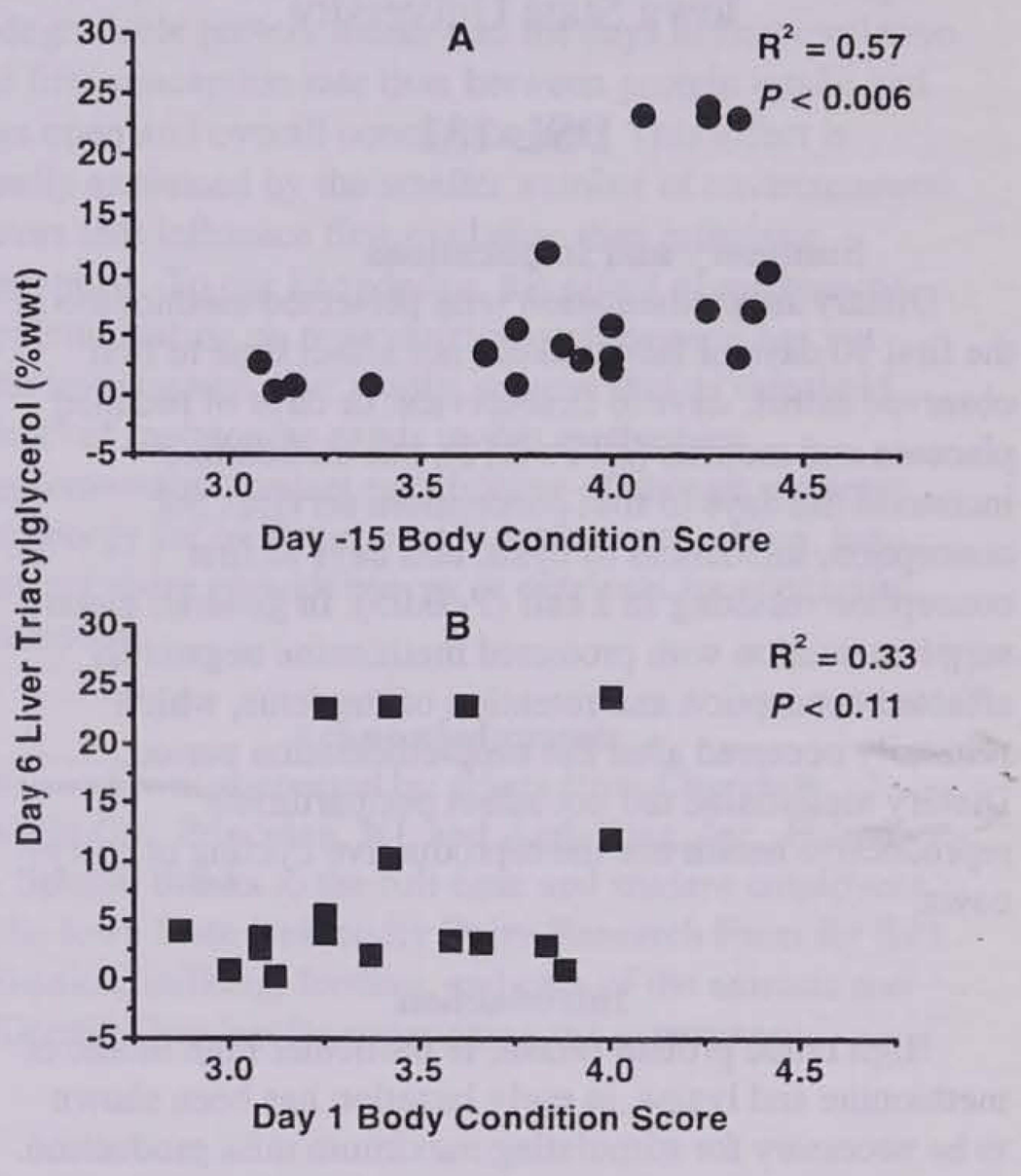


Figure 6. Correlations of liver triacylglycerols at day 6 postpartum to body condition scores 15 days prepartum (A) and 1 day postpartum (B).

Conclusions

1) Overfeeding dairy cows during the last 30 days prepartum leads to an increase in the severity of fatty liver and incidence of ketosis.

2) Evaluation of BCS 15 days prior to calving gives a better prediction of severity of fatty liver and susceptibility to ketosis than does BCS immediately after calving.

3) The incidence of postpartum metabolic disorders may be more closely related to prepartum nutrition than to body condition.

Effects of Dietary Supplementation with Protected Methionine on Reproductive Efficiency of Dairy Cows

G. Bobe, graduate research assistant;
G. L. Lindberg, assistant professor; and
L. H. Kilmer, professor
Department of Animal Science
Iowa State University

DSL-132

Summary and Implications

Dietary supplementation with protected methionine in the first 90 days of lactation did not affect time to first observed estrus, days to first service, or days of retained placenta and metritis (all P>0.10), but methionine increased the days to first conception, services per conception, incidences of cysts, and days to first conception resulting in a calf (P<0.05). In general, dietary supplementation with protected methionine negatively affected conception and retention of the fetus, which primarily occurred after the supplementation period. Dietary methionine did not affect periparturient reproductive health nor the reproductive cycling of dairy cows.

Introduction

High crude protein intake, in particular high intake of methionine and lysine, in early lactation has been shown to be necessary for stimulating maximum milk production. Some, but not all studies have shown a decrease in reproductive performance with increased crude protein intake. Because profits are related to reproductive performance as well as milk production, a negative relationship between dietary crude protein intake and fertility would have a negative impact on the income of the dairy industry. The major causes of unsatisfactory breeding performance are inadequate reproductive management and occurrence of reproductive disorders, such as retained placenta, metritis, and ovarian cysts. Therefore, when investigating the influence of diet on milk production, the incidences of reproductive disorders also need to be investigated.

Our goal was to determine whether dietary methionine supplementation in a high crude protein diet influences the occurrence of reproductive disorders and otherwise affects the reproductive performance of dairy cows.

Materials and Methods

Forty multiparous Holsteins, and 16 primiparous (heifer) Holsteins and eight multiparous Jerseys were blocked in groups based on breed, parity, and expected calving date and then assigned randomly to one of two postpartum diets (19.7% crude protein) that contained either 0% or 0.31% methionine hydroxy analog (Tables 1 and 2). Cows were housed in individual tie-stalls and were allowed to exercise outside for approximately one hour three times each day at milking time throughout the supplementation period (91 days). All diet ingredients were mixed and fed as a total mixed ration to cows ad libitum twice daily in amounts to achieve feed refusals of 5 to 10%.

Table 1 Composition of diets.

Ration Component	(% DM)
Alfalfa hay	20.9
Corn silage	28.6
Whole cottonseed	8.4
Soybean meal (48% CP)	5.6
Grain concentrate (17% CP)	28.1
Blood meal	1.1
Mega Pro/Mega Pro Plus[a]	6.2
Limestone	1.0

[a] Mega Pro contains rumen inert fat and expeller processed soybean meal. Mega Pro Plus contains rumen inert fat, expeller processed soybean meal, and methionine hydroxy analog (5%).

Data recorded for each cow included: days from parturition to first estrus, days from parturition to first service, days from parturition to first confirmed pregnancy, number of inseminations to first confirmed pregnancy, days from parturition to first pregnancy that resulted in a calf, number of cysts, number of luteolyses, and days of treatment for retained placenta and metritis. Cows that retained their placenta for longer than 1 day were treated with a broad spectrum antibiotic i.v. and a 500-ml saline infusion in the uterus every other day until the placenta was expelled. Cows with metritis (fever, yellow discharge) were treated with a broad spectrum antibiotic i.v. and an intrauterine Nolvasan infusion once daily.

Table 2 Nutrient composition of diets.

Chemical Composition	(%DM)
CP	19.7
UIP	39.0
ADF	22.3
NDF	36.8
NFC[a]	31.4
NE_L, Mcal/kg	1.61
Fat	4.1
Ash	8.0
Calcium	1.26
Phosphorus	0.39

[a] NFC=non-fiber carbohydrates calculated as 100-NDF-CP-crude fat-ash.

The reproductive health of all postpartum cows that had not been inseminated was monitored biweekly by palpation of the reproductive tract per rectum. Ovarian cysts were either luteolyzed or treated with GnRH. Visual observation of estrus was used to establish first estrus date. Reproductively healthy cows were bred at the first observed estrus after 50-d postpartum.

For statistical analysis, the effects of methionine supplementation on reproductive parameters (data were adjusted for prepartum diet , parity, breed, and calving season) were tested using a lifetime regression analysis and assuming a Weibull distribution of reproductive parameters. Two cows were not included in the study because they aborted.

Results and Discussion

The reproductive parameters show that only 36 of the 64 cows that entered the study had another calf. For the methionine supplemented group, 14 out of 32 cows had a subsequent calf (Table 3). The low reproductive performance of the methionine-supplemented group was not caused by periparturient disorders; to the contrary, the occurrences of retained placenta and metritis was lower in the methionine group. Neither was the low reproductive performance of the methionine-supplemented group caused by a delayed onset of estrus or signs of heat. The problem was that the cows in the methionine-supplemented group did not conceive and had a higher occurrence of ovarian cysts than the non-supplemented group. Both problems occurred primarily after the supplementation period, which ended at 91 days after calving.

In the published research literature, a negative relationship has been observed more often between high or degradable protein intake and for days to first ovulation and first conception rate than between protein intake and days open and overall conception rate. This effect is usually explained by the smaller number of environmental factors that influence first ovulation than influence conception. To our knowledge, the effect of methionine supplementation on reproductive performance has not been investigated. Our results suggest that a "threshold effect" of methionine exists in that methionine supplementation causes partitioning of enough nutrients and energy for ovulation and reproductive cycling, but does not spare enough energy or nutrients for successful conception.

Acknowledgments

This work was supported by grants from Church & Dwight Co., Princeton, NJ, and Agri-King, Inc., Fulton, IL. Special thanks to the full-time and student employees at the Iowa State University Dairy Research Farm for data collection, milking, feeding, and care of the animals and to Dennis Crawley for supervising the experiment.

Table 3 Averages of reproductive parameters and occurrences of disorders in cows fed different amounts of protected methionine.[a]

Parameter	Control	Methionine	P>F[b]
Reproductive performances:			
Days to activity left ovary	53 (n=28)	57 (n=22)	0.05
Days to activity right ovary	58 (n=30)	60 (n=27)	0.38
Days to first observed estrus	63 (n=31)	59 (n=27)	0.76
Days to first insemination	91 (n=30)	89 (n=26)	0.58
Days to first conception	107 (n=26)	125 (n=17)	<0.01
Services per conception	1.54 (n=26)	2.06 (n=17)	<0.01
Days to first conception resulting in a calf	112 (n=22)	125 (n=14)	<0.01
Occurrences of reproductive disorders:			
Days of retained placenta and treated metritis	3.56 (n=32)	2.00 (n=30)	0.07
Ovarian cysts	0.67 (n=30)	0.93 (n=27)	0.04
GnRH treated cysts	0.50 (n=30)	0.52 (n=27)	0.17
Luteolyses	0.97 (n=30)	1.59 (n=27)	<0.01

[a] Analysis included prepartum diet, postpartum diet, parity, breed, and season of calving in the statistical model.
[b] Probabilities of 0.05 and less are considered to be statistically significant.

Effective Fiber in Iowa Dairy Cow Rations

Dale Thoreson, extension field specialist,
Dairy/Beef/Forages

DSL-133

Dairy farmers, nutritionists, veterinarians, and extension specialists have become concerned with the increase in laminitis, acidosis, and fluctuations in milk production of high-producing dairy herds.

The Penn State Forage Separator was used to examine volunteered samples of total mixed rations (TMR), corn silage, and haylage for their separation into three categories. These results were compared with a large study conducted in the northeastern United States.

Iowa dairy cow rations appear to use corn silage that has less effective fiber than eastern rations; however, the TMR and haylage components appear very similar to eastern states.

Introduction

Effective fiber has become an issue in milking dairy cow rations. This issue relates to the increase in acidosis symptoms and the increase in feet and leg problems of dairy cows.

Several means have been described to measure the effective fiber in a ration. Early measures included (1) no less than 40% dry matter from roughage in the ration; (2) Wisconsin researchers suggested 21% neutral detergent fiber (NDF); (3) Mertens (3) described a guideline as 0.9% of the cow's body weight as forage NDF; (4) Shaver (4) suggested that 5 pounds of long forage particles (>1 1/2 inches) is needed for normal rumen function; and (5) Sniffen (5) suggested calculating effective NDF based on the fraction of a feed that remains on a 1.18 mm-screen.

Penn State University agricultural engineers and dairy scientists developed a three-box unit to measure forage particle size in the field (1).The Penn State Forage Separator separated forages or total mixed rations (TMR) into three groups: (1) those not passing through a screen of 19-mm holes and a thickness of 12.2 mm; (2) particles passing through the larger screen but not through a screen consisting of 8-mm holes and a thickness of 6.4 mm; and (3) a pan for all particles passing through both screens.

Penn State researchers and a commercial forage laboratory (2) conducted an extensive survey (12,920 samples) on forage and TMR samples from farmers in the northeast and mid-Atlantic states.

The purpose of our survey was to determine the characteristics of forage and TMR rations being fed to dairy cattle in northeast Iowa.

Materials and Methods

Three Penn State Forage Separators were used to measure ration's fiber characteristics. Samples were acquired at a series of veterinarians' cosponsored producer meetings in northeast Iowa during the fall and winter of 1996 and 1997, by making individual farmer trouble-shooting calls by Iowa State University Extension (ISUE) field and state dairy specialists, and by agri-industry, who borrowed a separator to conduct individual farm calls. Results from 52 samples have been analyzed and recorded.

Results and Discussion

The largest number of samples were TMR (39). Eight corn silage samples were separated, and five mostly legume haylage samples were separated. Results are described in Table 1.

Table 1 Particle size distribution of total mixed rations, corn silage, and haylage (values expressed as %).

	Top Screen	Middle Screen	Pan
TMR Means	7.7	32.2	60.1
Range of Means	1 to 32.3	10.5 to 49	37 to 77
Corn Silage Means	5.4	42.7	51.8
Range of Means	1 to 8.0	27 to 78	17 to 92
Haylage Means	18.8	39.0	42.2
Range of Means	4 to 47	24 to 58	29 to 65

Lammers (2) reported particle distribution on 831 TMR samples of 7.1%, 35.2%, and 57.7% for the top screen, middle screen, and pan, respectively.

Lammers (2) described 5,395 samples with means of 8.1%, 50.8%, and 41.1% of the samples on the top screen, middle screen, and pan, respectively.

Lammers (2) found their 2,815 legume samples resulted in 16.0, 40.5, and 43.5% of particles on the top screen, middle screen, and pan, respectively.

This small number of forage and TMR samples appears to separate out in roughly the same percentage as samples from the northeastern United States. Notable exceptions may be the corn silage samples, which had 2.7% less particles on the top screen, 8.1% less particles on the middle screen, and 10.7% more particles in the pan. Northeast Iowa dairy farmers may be setting their corn choppers to achieve a shorter theoretical length of cut than dairy farmers in the northeastern United States.

No data were given for corn silage samples processed through kernel processors in the Lammers data. One corn silage sample from our data was chopped by the Klauss Kernel Processor. Its results were 5, 78, and 17% for the top, middle, and pan, respectively. This technology may provide some promise of getting a larger percent of particles to stop

in the middle screen; thus, reducing the possibility of a large amount of highly fermentable particles reaching the cow's rumen shortly after ingestion.

The Penn State Particle Size Separator has been an effective teaching tool. It has helped dairy managers see the effect of today's forage processing and the addition of by-product feeds on the relative distribution of fiber particles in their milking rations.

Acknowledgments

The author thanks Tony Harvey, Lee Kilmer, and the cooperating veterinarians and nutritionists in northeast Iowa for their assistance in collecting these data.

Selected References

1. Lammers, B.P., D.R. Buckmaster, and A.J. Heinrichs, 1996a. A simple method for analysis of particle sizes of forages and total mixed rations. J. of Dairy Science. 79: 922.
2. Lammers, B.P., A.J. Heinrichs, and D.R. Buckmaster. 1996b. Feedstuffs, Vol. 68, No. 41, p. 14.
3. Mertens, D.R., 1988. Nonstructural and structural carbohydrates in large herd management. ADSA, Savoy, IL, p. 219.
4. Shaver, R.D., 1990. Forage particle length in dairy rations. Proc. Dairy Feed Systems Symp. NE ASAE, p. 58.
5. Sniffen, D.J., 1988. Balancing rations for carbohydrates for dairy cattle. Proc. Applications of Nutrition in Dairy Practice. American Cyanamid. Wayne, NJ, p. 25.

Iowa Producers Learn to Put More Pasture in Their Feed Inventory

Tony Harvey, extension dairy/beef specialist, and
Brian Lang, extension crops specialist

DSL-134

Hay is a major crop for northeast Iowa cattle producers. The top five alfalfa producing counties in Iowa are located there – Clayton, Dubuque, Winneshiek, Jackson, and Allamakee. These counties are also major cattle production areas as producers add value to the hay harvested by converting it to milk and meat. Dubuque is the leading Iowa county in dairy cow numbers and Jackson is the top beef cow county.

A significant challenge for northeast Iowa cattle producers is to grow, harvest and store high-quality forages. Obstacles include winter kill of alfalfa plants requiring premature reestablishment as well as rainfall and humidity that make drying hay difficult. In addition, many of the hay harvesting systems are machinery and energy intensive. Compared to when many of these systems were put in place the relationship of energy costs and product value have narrowed.

The difficulty and cost of growing and harvesting quality hay and the continued pressure to reduce costs has stimulated producer interest in management intensive grazing systems. While interest has increased, there have been few places Iowa producers can go for information and education on intensive grazing.

Intensive rotational grazing is an alternative management approach for producing milk and meat. It involves dividing pastures into smaller subsections and grazing animals for short periods of time. This allows for plants to recover before grazing again, thus improving the productivity of the pasture and the nutritional quality of the grasses for livestock.

Benefits of intensive rotational grazing noted by farmers include reduced feed costs, increased pasture productivity, healthier livestock and improved lifestyles. Other positive aspects identified are protection of the soil from erosion, reduced use of petroleum-based fertilizers and pesticides as well as prevention of ground and surface water contamination.

For producers adopting intensive rotational grazing it means learning new management skills. Grazing management requires balancing the relationship between livestock and plants in new ways. Intensive rotational grazing is a technology that takes the form of knowledge, skill and management. It is not something you can buy and put on a field or feed a cow, it is a system that has to be learned.

A way to increase the grazing experiences of producers is to involve them in pasture walks. These walks use the pasture as a field classroom. Participants walk around a pasture discussing what's working and what's not with the grazing system. They allow for producers to help each other learn grazing management strategies.

Iowa State University Extension and the Natural Resources Conservation Service in cooperation with area producers submitted a grazing education and demonstration proposal to the Leopold Center for Sustainable Agriculture. The proposal was accepted and resulted in an increased number of opportunities for northeast Iowa dairy and beef producers to learn about intensive rotational grazing. It supported communication of pasture walks in the region, evaluating the learning of walk participants, in-depth descriptions of several producers using management intensive grazing and forage quality measurements.

Pasture walks were planned with the following objectives:

To assist dairy and beef producers in learning about intensive rotational grazing management practices through farmer to farmer communication and demonstration.

To demonstrate use of fencing materials, paddock layout and water systems.

To teach principles of managing cattle and pasture to improve.

To assist in increasing the number of acres of intensive rotationally grazed pasture.

What was observed and learned from this project?

People came to pasture walks. In northeast Iowa there were 27 pasture walks in 1995 and 26 in1996 attended by approximately 575 and 485 respectively.

Pasture walk participants learned more about management intensive grazing. Ninety-eight percent of the pasture walk participants indicated they increased their ability to assess livestock production problems. They also rated the usefulness of the information presented at pasture walks on a 1 to 5 scale with 1 being not very useful and 5 being very useful at an average of 4.3 for 1995 and 3.95 for 1996.

Walk participants applied what was learned. The 1996 survey respondents (70) indicated actions taken as a result of attending a pasture walk. The percentage of respondents indicating taking a specific action follows:

tried frost seeding (50%)
started to stockpile pasture for late fall or winter grazing (29%)
tried a new forage variety (50)
adopted a new fencing or watering strategy (50%)
increased recording of pasture movements (26%)
adjusted supplemental feed program (21%)
reduced investment in machinery and equipment (24%)
reduced the use of chemicals and fuel (36%)
changed breeding program (14%)

Many utilized intensive rotational grazing as part of a business expansion strategy. In the past three years 67% of

the 1996 respondents increased the number of acres grazed and 63% increased cow numbers. One half of the respondents indicated that the changes resulted in an increase of farm income while 45 percent reported income stayed about the same.

Livestock have long added to the economic viability of rural Iowa farms and communities. The challenge for the future is to discover and adopt production systems that can be sustainable under current economic conditions. Intensive grazing appears to be a production system that can beneficial in helping maintain dairy and beef cow businesses as significant contributors to the economies of northeast Iowa.

The authors would like to especially thank the northeast ISU County Extension Education Directors, John Rodecap and staff at the Northeast Iowa Demonstration Project, Jim Ranum, NCRS Grazing Specialist, the Leopold Center For Sustainable Agriculture, Practical Farmers of Iowa members, Dr. Stephen Barnhart, Extension Forages Specialist, Dr. Jim Russell, ISU Animal Scientist, and Mike Frieburger, watershed project coordinator for their cooperation and support of the management intensive grazing education programs.

Executive Summary of Research in the Metabolic Diseases and Immunology Research Group (USDA/ARS/MWA/NADC)

Ronald Horst, PhD, research physiologist and research leader;
Timothy Reinhardt, PhD, research animal scientist;
Marcus Kehrli, DVM/PhD, veterinary medical officer;
Jesse Goff, DVM/PhD, veterinary medical officer;
Brian Nonnecke, PhD, microbiologist;
Jim Harp, PhD, microbiologist;
and Mark Rasmussen, PhD, microbiologist

DSL-135

The mission of our group is to improve the neonatal and periparturient health and production efficiency of dairy animals by conducting basic and applied research on the pathogenesis and prevention of metabolic and infectious disease of major economic importance. Our group's research is focused on three major diseases: mastitis, cryptospoidiosis, and milk fever. Annual loses to U.S. farmers due directly to these diseases are estimated to be about $2 billion, $100 million, and $150 million, respectively. The following represents a summary of the groups major accomplishments in these and related areas during the past 3 years.

Mastitis--We cloned and sequenced a receptor for bovine tumor necorsis factor (TNF) that will be used to evaluate the role of TNF in coliform mastitis. We have identified genetic variation in the ability of the immune system of cattle to resist and recover from simulated stress conditions. We have shown that coliform bacteria are not always detectable in feces of cattle. This has implications on the use of coliform counts on carcasses in slaughter houses as a monitor of fecal contamination. We have demonstrated the efficacy of an immunomodulator that reduces the incidence of coliform mastitis by 50% in periparturient cows. We have isolated and cloned the genes for bovine granulocyte-colony stimulating factor and interleukin-7, both of which will be evaluated for potential use in naked DNA experiments. We have designed two cDNAs of bovine ICAM(intracellular adhesion molecule)-1 that will be used to express a recombinant soluble form of the protein for use as an anti-inflammatory drug in cattle.

Nutritional immunology--We established a link between corticosteroid-induced changes in the composition of blood mononuclear populations and their functional capacity, and extended observations by showing that mammary function during immediate postpartum period may contribute to observed reduction in the capacities of lymphocytes to secrete immunoglobulin and interferon-γ during this period. Other research indicated that feeding high levels of vitamin A to young calves profoundly increased circulating concentrations of biologically active metabolites of vitamin A and suppressed vitamin E concentrations. These changes were associated with markedly reduced daily weight gain, suggesting a negative impact of excess dietary vitamin A in calves. In related research, 9,13-*di-cis*-retinoic acid, present at unusually high concentrations in maternal and neonatal plasma after parturition, had minimal impact on the the differentiation and functional capacities of polyclonally-stimulated blood mononuclear leukocytes from neonatal calves and adult dairy cows.

Cryptosporidisosis--We determined that administration of sucrose to infant mice prior to challenge with Cryptosporidium parvum drastically reduces infection with the parasite. This effect is specific to sucrose as other carbohydrates tested did not have this effect. We are exploring the mechanism of this protection. We also determined that scrapings from the intestinal wall of adult rats contain a substance (apparently heat-labile) that can transfer protection from C. parvum infection to susceptible infant rats. We are undertaking the characterization of this substance and its ability to induce cytokine production in the recipients. Both of the above projects are model systems for C. parvum infection in calves and may lead to improved treatments for cryptosporidiosis. We successfully introduced a gene coding for a 15/60 kD protein of Cryptosporidium parvum into an enteroadherent, nontoxigenic strain of E. coli. We are now beginning to examine the ability of this construct to colonize the intestines of infant mice and to provoke an immune response to C. parvum. The ultimate goal is to improve this construct, through the addition of further immunostimulatory genes, allow for colonization of calves, and protect against C. parvum infection.

Milk fever--We have recently demonstrated the utility of hydrochloric acid as a source of anions for prevention of milk fever in dairy cattle. We also have established that sulfate is a very poor anion source and is

about 20% as effective as chloride at creating a mild metabolic acidosis and, therefore, milk fever prevention. Factors controlling calcium homeostasis, transport and signaling are largely unknown in the mammary gland. Using PCR and direct PCR sequences, we have identified six calcium ATPases in the mammary gland that belong to three different classes of P-type pumps. Isotypes of these pumps have been identified and they are all highly regulated during lactation. The vitamin D-dependent P-type pump of the intestine was found to be negatively regulated by calcitonin. New competitive PCR assays have been developed to study transcriptional regulation of specific genes in cows. Anti-peptide antibodies to the vitamin D receptor were developed. A role for vitamin A in vitamin D metabolism was confirmed in vivo.

Rumen microbiology--A collaboration with French researchers at the INRA, Lyon, was established to investigate rumen metabolism of fescue alkaloids. This collaboration allows this project to obtain rumen microbes from cattle in Europe that may be useful in fescue detoxification. Rumen bacteria were isolated from goats that reduce selenium in combination with hydrogen. Nucleic acid sequence analysis was used to identify these bacteria and to develop probes useful in studying the ecology of these bacteria in the rumen. A bacteria that degrades nitropropanol also was found to effectively divert rumen fermentation away from its wasteful production of methane. The application of this microbe for controlling methane production in cattle is currently on-going.

Effects of Native Fat in Colostrum and Milk on Concentrations of Fat-Soluble Vitamins in Serum of Neonatal Calves

V. Rajaraman, MSc; B. J. Nonnecke, PhD;
and R. L. Horst, PhD
U. S. Dept. of Agriculture,
Agricultural Res. Service,
National Animal Disease Center,
Metabolic Dis. and Immunol. Res. Unit,
P. O. Box 70, Ames, IA

DSL-136

Summary and Implications

Fat-soluble vitamins and their metabolites are essential to normal growth and development of the neonate. These compounds also have been shown to be important in the ontogeny and responsiveness of the immune system. The objective of the present study was to determine the contribution of fat in colostrum and milk on concentration of fat-soluble vitamins in peripheral blood of neonatal calves during the first week postpartum. During this period, control calves (n=6) were fed normal colostrum and milk, and calves in the treatment group (n=6) were fed skimmed colostrum and milk supplemented with coconut oil. In control calves, serum concentrations of retinol (vitamin A), β-carotene, α-tocopherol (vitamin E), and 1,25-dihydroxyvitamin D increased significantly, relative to values on day 0, during the seven day period. In contrast, plasma concentrations of the vitamins in calves fed skimmed colostrum without native fat remained unchanged from birth through 7 days postpartum. Although serum retinoic acid concentrations in control calves increased during the experimental period, most notably the concentrations of 9,13-*di-cis*-retinoic acid, similar changes were not observed in serum from treated calves. Interestingly, the amount of serum IgG_1 in serum of control and treated was not different, suggesting retention of native fat was not necessary for assuring passive immunity. Thus, removal of native and replacement with coconut oil resulted in marked alterations in fat-soluble vitamins in serum without affecting passive immunity. This treatment method may provide a model system for the study of the specific roles of fat soluble vitamins on the physiology of the neonatal calf.

Introduction

Newborn calves have very low concentrations of vitamin A and α-tocopherol in their sera. With age, these concentrations increase, and adult concentrations are attained by about 2 years of age. Colostrum, milk, and formulated diets are major sources of these vitamins for the bovine neonate. Vitamin A promotes differentiation and maturation of a cells in all tissues, primarily through its metabolites, the retinoic acids (RA) which are derived from retinol and retinyl esters. β-Carotene not only functions as a biological antioxidant but also as a precursor of vitamin A. Vitamin E, a lipid-phase antioxidant, functions in conjunction with selenium. Vitamin D, acting via its metabolite 1,25-$(OH)_2$D plays a critical role in calcium homeostasis. Recent evidence indicates these vitamins also affect broad aspects of immune function and disease susceptibility.

Horst and coworkers have identified a unique RA isomer 9,13-*di-cis*-RA as the most abundant vitamin A metabolite in plasma of the dam and newborn calf. Elevation of 9,13-*di-cis*-RA in neonatal calf serum is dependent on consumption of colostrum by the calf. Although this isomer has been shown to have little biologic activity, studies with rats indicate that 9-*cis*-RA (an isomer with biological activity) may be a precursor to 9,13-*di-cis*-RA. These observations suggest that metabolic events at the time of parturition may influence vitamin A metabolism resulting in increased production of 9-13-*di-cis*-RA and have the potential to influence immune responsiveness in the dam and calf.

The objective of this study was develop an in vivo model in the which elevation in fat-soluble vitamins could be controlled/prevented allowing the effects of these vitamins on general physiology and immune function in the calf to be evaluated. To achieve this objective, newborn calves were fed for the first seven days postpartum, colostrum and milk in which the native fat was replaced with coconut oil, a fat source providing necessary energy but lacking detectable levels of these vitamins.

Materials and Methods

Twelve newborn Jersey calves of cows housed at USDA, ARS, National Animal Disease Center were caught at birth before suckling the dam. During the first week postpartum, 6 calves were fed complete colostrum and milk, and 6 calves were fed skimmed colostrum and skimmed milk that were supplemented with coconut oil [12 and 3.5% (vol/vol)], respectively.

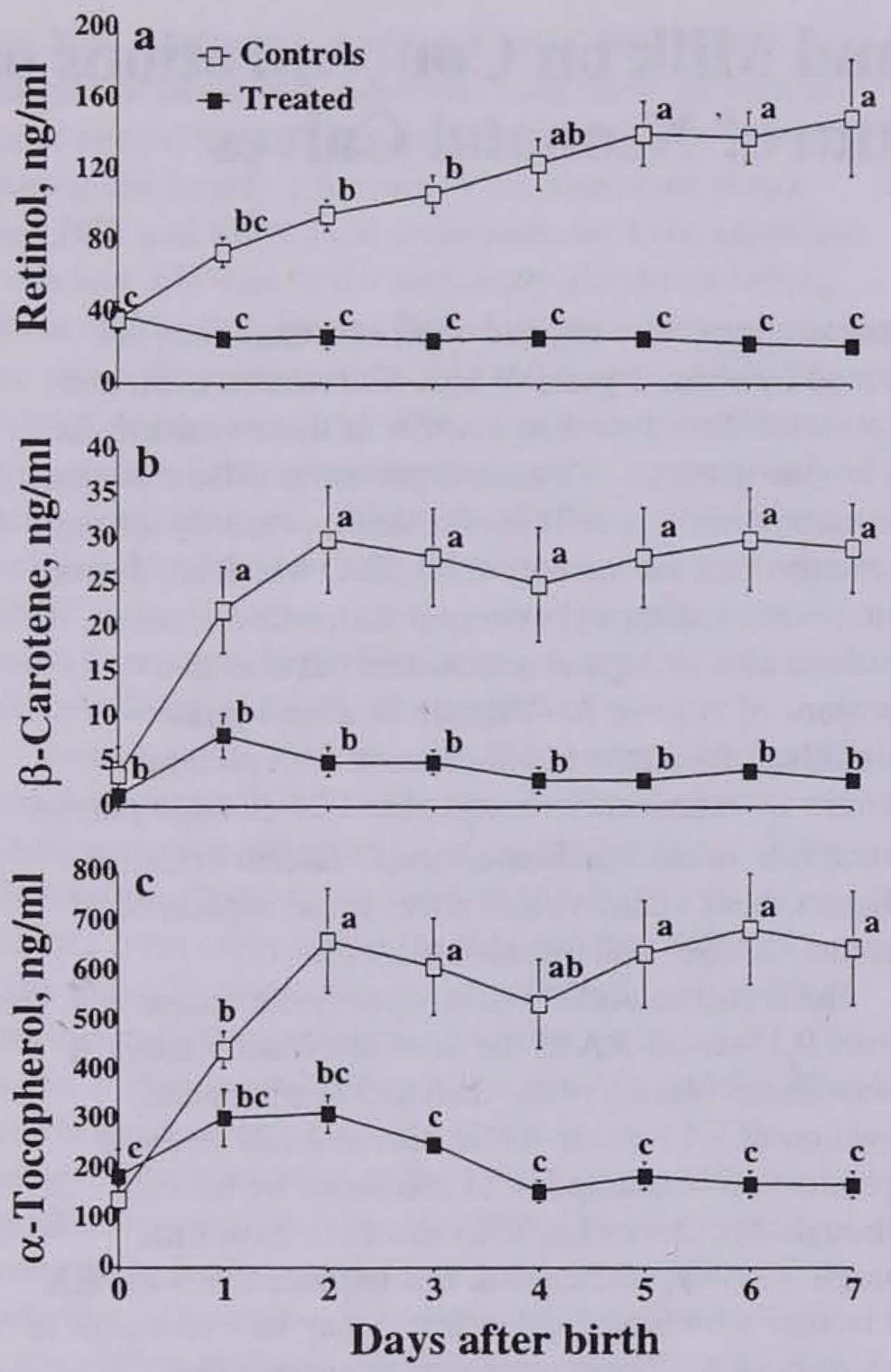


Figure 1. Concentrations of retinol (a), b-carotene (b), and a-tocopherol (c) in serum of neonatal calves treated with normal or skimmed colostrum and milk for 7 days. Means with different superscripts differ (P≤.01).

Colostrum was collected from 10 cows and pooled. Approximately one-half of the volume of pooled colostrum was skimmed. Intact and skimmed colostrum was divided into 4 L aliquots and frozen at -20°C. Each aliquot was thawed immediately before use. Each calf was fed 2 L of pooled colostrum within 3 hours of birth and another 2 L within 12 hours of birth. Calves were subsequently fed 2 L of milk in the morning and afternoon until 7 days postpartum. Blood samples were obtained from calves within 3 hours after birth and once every 24 h for 7 days thereafter. Blood was also collected from two adult Jersey heifers on eight separate occasions during the study. All calves remained clinically normal during the experimental period.

Concentrations of fat-soluble vitamins (retinol, β-carotene, α-tocopherol) in colostrum and milk and serum were determined by HPLC. Concentrations of 25-dihydroxyvitamin D in serum were determined by radioimmunoassay. A nonequilibrium receptor-based assay was used to determine serum 1,25-$(OH)_2D$ concentrations. Serum IgG_1 concentrations were determined by an ELISA.

Data were analyzed by a split-plot repeated measures ANOVA. Significant differences between treatments or days were calculated by Student's two sample *t* -test using least square means. Statistical differences were declared at P≤.05.

Results and Conclusions

Analysis of fat-soluble vitamin concentrations in colostrum and milk-fed calves indicated that normal milk had 3- to 4-fold higher retinol than did colostrum. Skimmed colostrum had very low retinol relative intact colostrum. Normal colostrum and milk had about 8- to 13-fold higher a-tocopherol and 1.4- to 1.6-fold more 1,25-$(OH)_2D$ than did skimmed colostrum and skimmed milk. Normal colostrum had 4.5 fold more β-carotene than did skimmed colostrum. No β-carotene was detected in skimmed milk. 25(OH)Vitamin D was not detectable in normal or skimmed colostrum or skimmed milk. These vitamins were not detectable in coconut oil.

At birth (day 0), serum concentrations of retinol, β-carotene, α-tocopherol, RA isomers, 25(OH)D, and 1,25-$(OH)_2D$ were the same in control and treated individuals (Figs. 1-3). Thereafter, there were progressive increases in the concentrations of retinol, β-carotene, α-tocopherol 1,25-$(OH)_2D$ in the serum of control calves. In treated calves the concentrations of these compounds remained essentially unchanged from birth (day 0) to 7 days postpartum. Serum concentrations of 1,25$(OH)_2D$ were unaffected by treatment.

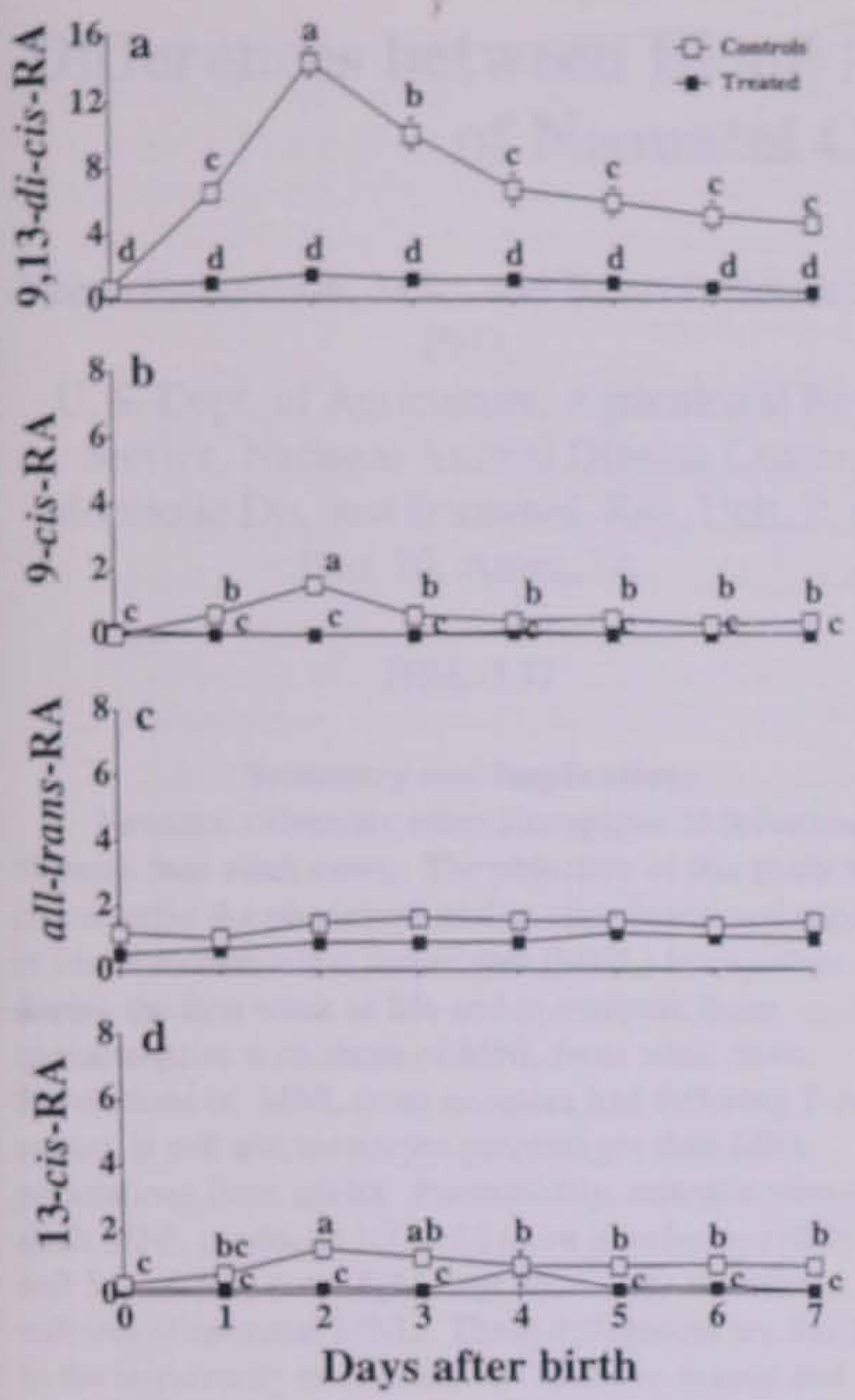


Figure 2. Concentrations (ng/ml) of 9,13-*di-cis*-retinoic acid (RA) (a), 9-*cis*-RA (b), *all-trans*-RA (C) in serum from neonatal calves treated with normal or skimmed colostrum and milk for 7 days. Means with different superscripts differ (P < .01).

Although concentrations of 9-*cis*-RA, *all-trans*-RA, and 13-*cis*-RA concentrations were similar in both groups during the experimental period, the concentration of 9,13-*di-cis*-RA in control calves was significantly elevated relative to concentrations in treated calves from day 1 through day 7.

Immunoglobulin G_1 concentrations in serum were unaffected by treatment. Serum IgG_1, undetectable at birth (day 0), increased to 1500 mg/dl by day 2, and remained unchanged for the duration of the experimental period in all calves (Fig. 4).

In this study, we have shown that it is possible to abrogate the normal and progressive elevation in the concentration of RA and other retinoids that is observed in calves during the first week postpartum by depriving newborn calves of the natural fat fraction of colostrum and milk. Although this treatment did not effect serum IgG_1 or functional capacities of immune cells (data not shown), it did prevent the natural increase in the concentrations of fat-soluble vitamins and their metabolites in serum.

The treatment did not affect serum 25(OH)D levels in serum but did result in lower concentrations of $1,25(OH)_2D$ from days 3-7. The lack of a treatment effect on serum 25(OH)D levels (Fig. 3) might have been due to diet, which was not a significant source of this vitamin. The elevation in $1,25\text{-}(OH)_2D$ observed in 1- and 2 day-old calves is in agreement with earlier reports, which indicate that low concentrations of calcium in plasma at birth activate the 1-α-hydroxylase enzyme, promoting the generation of $1,25\text{-}(OH)_2D$ from 25(OH)D.

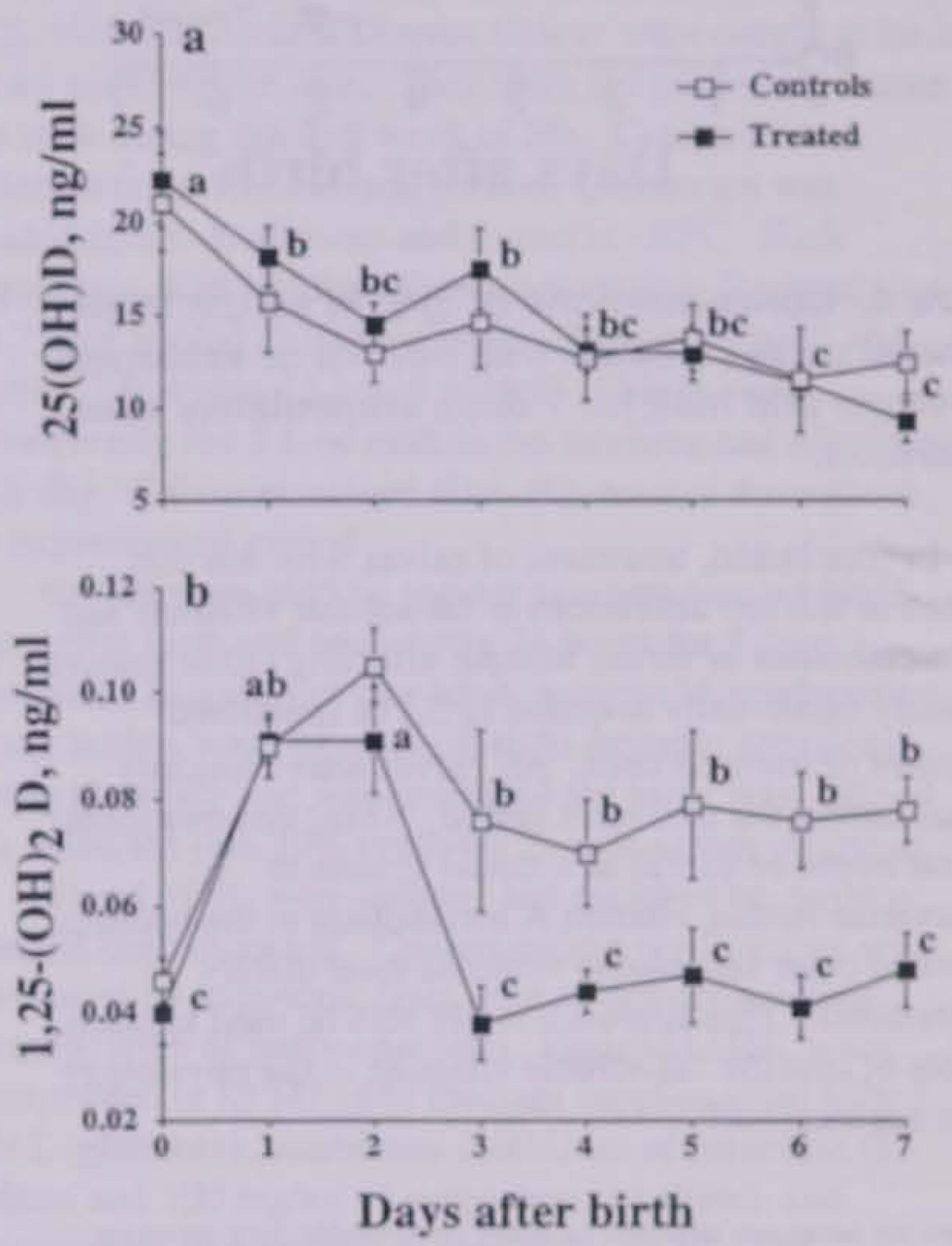


Figure 3. Concentrations of 25-hydroxyvitamin D [25(OH)D] (a) and 1,25-dihydroxyvitamin D [1,25-$(OH)_2D$] in serum from neonatal calves treated with normal or skimmed colostrum and milk 7 days. Means with different superscripts differ (P ≤ .01).

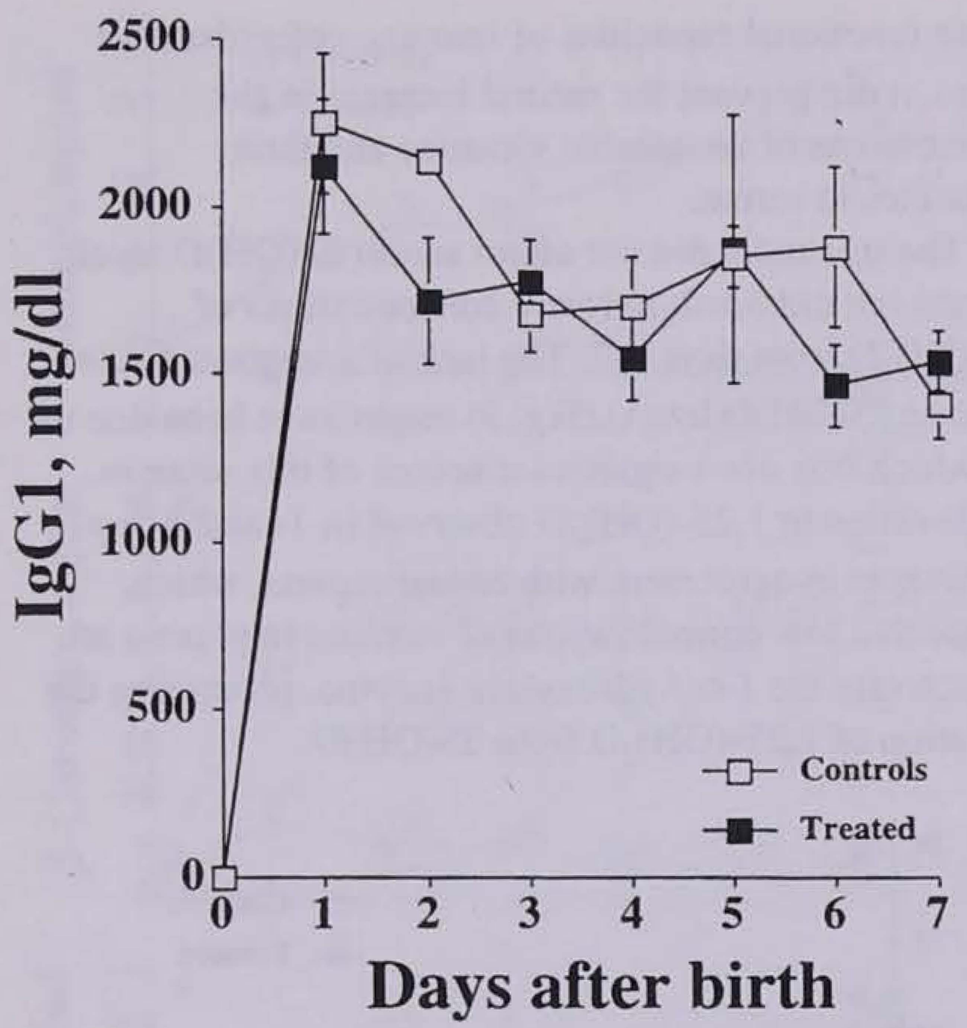


Figure 4. Concentrations of IgG_1 in serum from neonatal calves treated with normal or skimmed colostrum and milk for 7 days immediately after parturition.

In conclusion, treatment of calves with this diet resulted in marked alterations in fat-soluble vitamins and their metabolites in serum without affecting either passive immunity (maternally acquired IgG_1) or functional capacities of immune cells. All calves were clinically normal during the treatment period. Thus, this treatment method might be useful as a model system to characterize further vitamin A metabolism in the neonatal calf when other fat-soluble vitamins meet dietary requirements. This approach might also be used to study the role of specific fat-soluble vitamins in the physiology of the neonatal calf.

[1]Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that may also be suitable.

Differences between Blood Mononuclear Leukocyte Populations of Neonatal Calves and Adult Cows

Veena Rajaraman, MSc; and Brian J. Nonnecke, PhD,
U. S. Dept. of Agriculture, Agricultural Res. Service, National Animal Disease Center, Metabolic Dis. and Immunol. Res. Unit, P. O. Box 70, Ames, IA

DSL-137

Summary and Implications

Neonatal calves are more susceptible to infectious diseases than adult cows. The objective of this study was to characterize the phenotype and in vitro functional capacities of blood mononuclear leukocytes (MNL) from calves during the first week of life and to compare these characteristics with those of MNL from adult cows. Populations of MNL from neonates had differing T-cell subset, B cell and monocyte percentages than MNL populations from adults. Functionally, mitogen-stimulated adult MNL produced 100-fold more interferon-γ (IFN-γ) and 5- to 6-fold more IgM than identically stimulated cultures of neonatal MNL. These differences are likely due to the immaturity of the neonatal immune system and suggest that neonatal MNL are less functionally responsive than adult MNL. Mitogen-induced IFN-γ production and DNA synthesis, and percentages of MHC class II positive cells were lowest in MNL from calves 1 to 4 days of age. These results indicate that there is a period of reduced responsiveness of the immune system in the immediate postpartum period that may contribute to the elevated susceptibility of the calf to infectious diseases.

Introduction

Physiologic immaturity of the neonatal immune system is believed to render the newborn more susceptible to infectious diseases than the adult. Studies with humans and mice indicate that this immaturity is characterized by the presence of T-cell populations that have higher proportions of naive T cells that have the ability to suppress Ig production. They also have higher proportions of antigen-presenting cells with defective costimulatory activity, and decreased abilities to produce cytokines. Calves are known to have higher proportions of γδ T cells and lower proportions of circulating B cells than adult cows. The number of B cells and the ability to produce Ig gradually increase to adult levels by about 20 weeks of age. Serum from 1 day-old calves contains factors other than cortisol that suppress lymphoblastogenesis of calf lymphocytes from the spleen, thymus, and lymph nodes.

The objective of this study was to characterize the phenotype and in vitro functional capacities of blood mononuclear leukocytes (MNL) from calves during the first week of life and to compare these characteristics with those of MNL from adult cows.

Materials and Methods

Twelve newborn Jersey calves housed at the USDA, ARS, National Animal Disease Center were caught at birth before suckling the dam. They were fed pooled colostrum and milk during the first week of life. Colostrum was collected from 10 cows and pooled. Colostrum was divided into 4 -L aliquots and frozen at -20°C. Each aliquot was thawed immediately before use. Each calf was fed 2 L of pooled colostrum within 3 hours of birth and another 2 L within 12 hours of birth. Calves were subsequently fed 2 L of milk in the morning and afternoon each day. Calves remained clinically normal throughout the experimental period.

Calves were bled by jugular venipuncture within 3 hours after birth and once every 24 hours for 7 days thereafter. Blood from two adult, nongravid, nonlactating Jersey heifers was collected on eight separate occasions during the study and was processed the same way. Blood was collected into 10% (vol/vol) 2x acid-citrate-dextrose.

Blood MNL were isolated and enriched by density gradient centrifugation. Contaminating erythrocytes were eliminated by hypotonic lysis prior to density gradient centrifugation of buffy coat cells. Enriched MNL were resuspended in RPMI 1640 medium supplemented with 2 m*M* L–glutamine, antibiotics (100 U/ml of penicillin G sodium and 100 mg/ml of streptomycin sulfate), and antimycotics (0.25 µg/ml of amphotericin B). The medium used for IFN–γ assays was additionally supplemented with nonessential amino acids and 2–mercaptoethanol (55 µ*M*).

Leukocytes in MNL populations were phenotyped using one-color flow cytometry. Density gradient-enriched MNL were washed with and resuspended in cold Hanks' balanced salt solution (HBSS) with 1% heat-inactivated fetal calf serum (FCS) and 0.1% NaN_3 at a density of 10×10^6/ml. Approximately 5×10^5 MNL from each suspension were added to individual wells of a 96 well microtiter plate. Monoclonal antibodies diluted in HBSS with 1 % FCS and 0.1% NaN_3 were added (50 µl aliquots) individually to wells containing the MNL. Plates were incubated at 4°C for 15 min and washed twice by

centrifugation (1,171 X *g* at 4°C for 2 min). Supernatants were removed using a plate washer. Cells were resuspended in HBSS with 0.1% NaN_3 and incubated (4°C for 15 min) with secondary antibody [fluorescein isothiocyanate-conjugated goat $F(ab')_2$ fragments against mouse IgG or IgM, 50 µl]. Plates were washed again, and cell pellets were resuspended in HBSS with 0.1% NaN_3. Nonspecific binding of antibody was assessed by incubating each test sample with secondary antibody alone. Specificities of all primary antibodies were ensured by testing with isotype controls.

A Becton Dickinson FACScan was used for flow cytometric analysis of 5,000 cells that exhibited light scatter properties that were consistent with bovine MNL. An argon laser with an excitation wavelength of 488 nm was used to detect cells associated with fluorescein isothiocyanate-conjugated second antibody. Emission fluorescence was detected with a 530-n*M* bandpass filter and converted to log fluorescence. Markers were positioned for negative control samples to provide a background of 2% and were maintained at this position for all samples taken from the calf. Cells with fluorescence intensity greater than the marker position were considered positive. Fluorescence data associated with each parameter were expressed as a percentage of the gated MNL population.

Secretion of IgM by unstimulated MNL cultures and MNL cultures stimulated with pokeweed mitogen (PWM) was quantified by an ELISA. Cultures were established in flat-bottomed, 24-well polystyrene tissue culture plates seeded with 1.0 X 10^6 cells/ml in a final volume of 1.5 ml containing 5% (vol/vol) FCS in RPMI 1640 medium with antibiotics, antimycotics and glutamine. Resting MNL cultures and MNL cultures stimulated with PWM (0.08 or 0.32 µg/ml, respectively) were incubated at 39°C in a humidified atmosphere containing 5% CO_2 for 14 days. The concentration (ng/ml) of IgM in supernatants was determined by comparison of absorbance of supernatants with absorbance of standards within a linear curve fit.

Secretion of IFN-γ was evaluated in MNL cultures established in flat-bottomed, 96-well polystyrene tissue culture plates seeded with 5 X 10^6 cells/ml in a total volume of 200 µl of RPMI 1640 medium with 5% FCS, 55 µ*M* 2–mercaptoethanol, and nonessential amino acids. Resting MNL cultures and MNL cultures stimulated with PWM (10 µg/ml) were incubated for 48 h at 39°C in a humidified atmosphere with 5% CO_2. Culture supernatants from centrifuged plates (800 X *g* at 4°C for 2 min) were harvested and stored at -80°C until analyzed.

Interferon-γ was measured using an IFN-γ capture ELISA. Absorbance of standards, controls, and test samples was determined at 410 nm using an automated ELISA plate washer and reader. Interferon-γ in test samples was determined from a standard curve of absorbance regressed on $\log_{10}$ values of the IFN-γ concentration for the dilution of the test sample that gave absorbance readings falling in the linear portion of the curve. The IFN-γ concentration in culture supernatants was calculated by multiplying the value from the standard curve by the dilution factor and was expressed as picograms per milliliter.

To evaluate synthesis of DNA by MNL, cultures of MNL were established in flat-bottomed, 96-well tissue culture plates seeded with 5.0 X 10^5 cells/ml in a final volume of 200 µl containing 5% FCS. Lymphocyte proliferation was assessed in unstimulated MNL cultures and in cultures stimulated with PWM (1 µg/ml) or concanavalin A (1 µg/ml). Cultures were incubated for 72 hours at 39°C in a humidified atmosphere containing CO_2 and then pulsed with 18.5 kBq of [methyl-^{3}H]thymidine. Cells were harvested 18 hours later. Retained radioactivity, expressed as counts per minute, was determined by liquid scintillation spectrophotometry.

Data were analysed by the a split-plot type of repeated measures ANOVA.

Results

There were significant (P=0.0001) day-related changes in the percentage of positive MNL that expressed $CD3^+$, γδ T cell, B cell, and major histocompatibility complex (MHC) class II^+ antigens. The other antigens did not change with time. The MNL populations from 1-day-old calves had higher percentages of $CD3^+$ cells than did 3- or 4-d-old calves (P<0.0002; Fig. 1). Percentages of γδ T cells in the MNL population from 1-day-old calves were higher when compared with MNL from 2, 3, 4, and 7 day-old calves and in newborn calves (P=0.0005; Fig. 1).

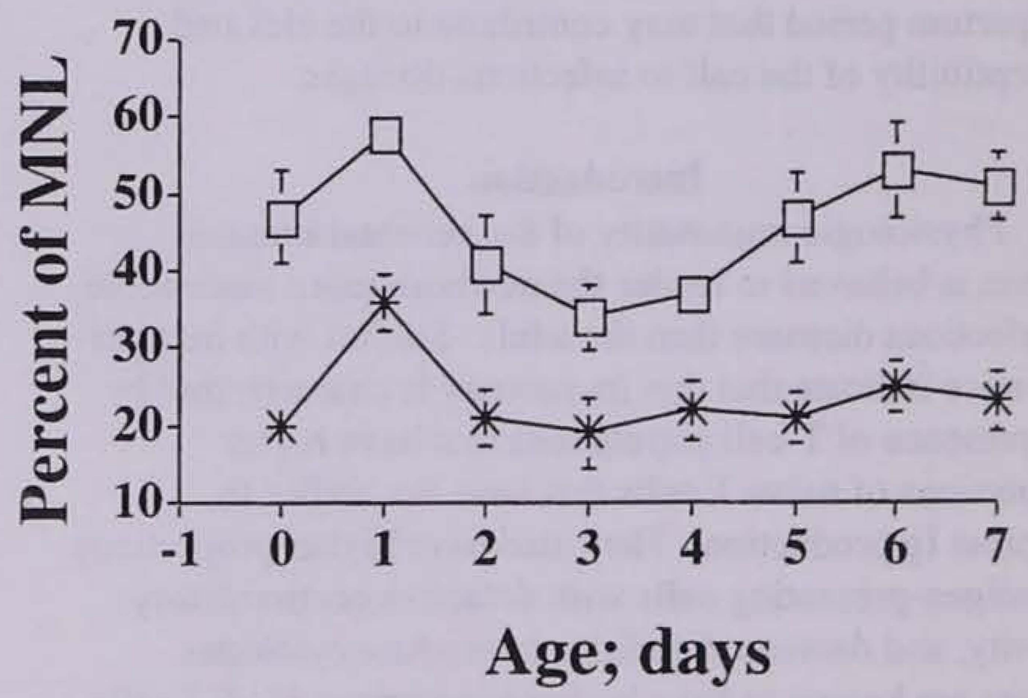


Figure 1: Mean percent of mononuclear leukocytes (MNL) positive for CD3 (open square) or γδ-T cell (x) markers.

Mononuclear leukocyte populations from 1-day-old calves had higher percentages of B cells when compared

with MNL from calves at birth and with MNL from calves at 6 and 7 days of age (P=0.0007; Fig. 2). Conversely, the percentage MHC class II antigen$^+$ cells in MNL populations from 1-day-old calves was lower than that in MNL populations from 2-, 3-, and 4-d-old calves (P=0.0001; Fig. 2).

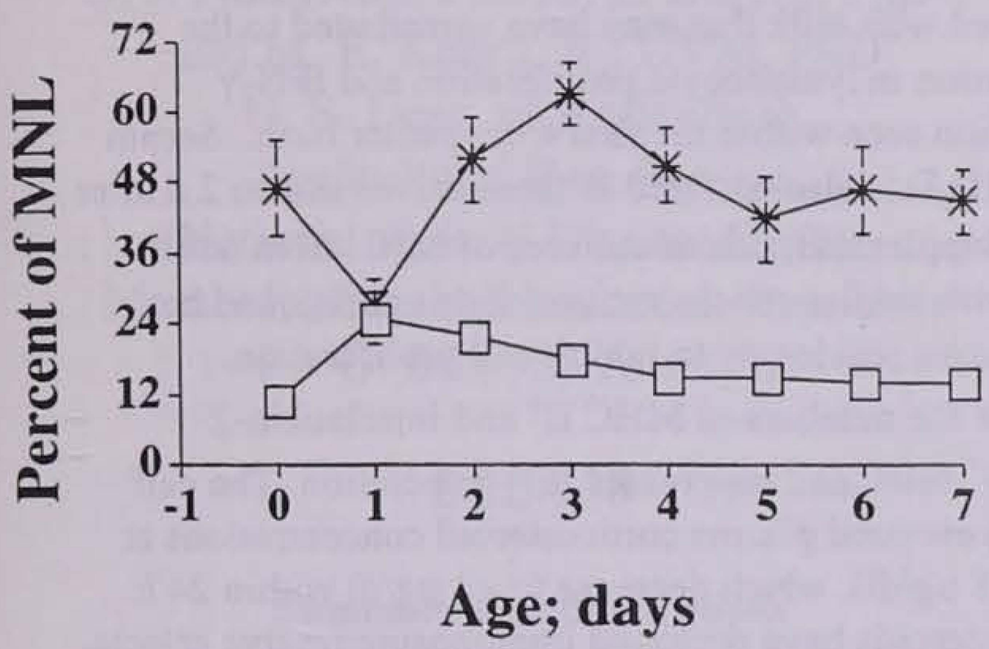


Figure 2: Mean percentage mononuclear luekocytes (MNL) positive for B cell (½) or MHC class II (x) cell surface markers.

A strong positive correlation between percentages of monocytes and MHC class II antigen$^+$ cells was detected (r=0.81; P=0.0001), and a negative correlation between MHC class II antigen$^+$ cells and CD3$^+$ (r=-0.47; P=0.0001) and CD4$^+$ cells (r=-0.46; P=0.0001) was detected.

Mononuclear populations from neonatal calves had significantly higher percentages of CD4$^+$ and CD8$^+$ T cells, γδ T cells, and monocytes, and lower percentages of B cells when compared with MNL populations isolated from adult cows (P≤0.05; Fig. 3).

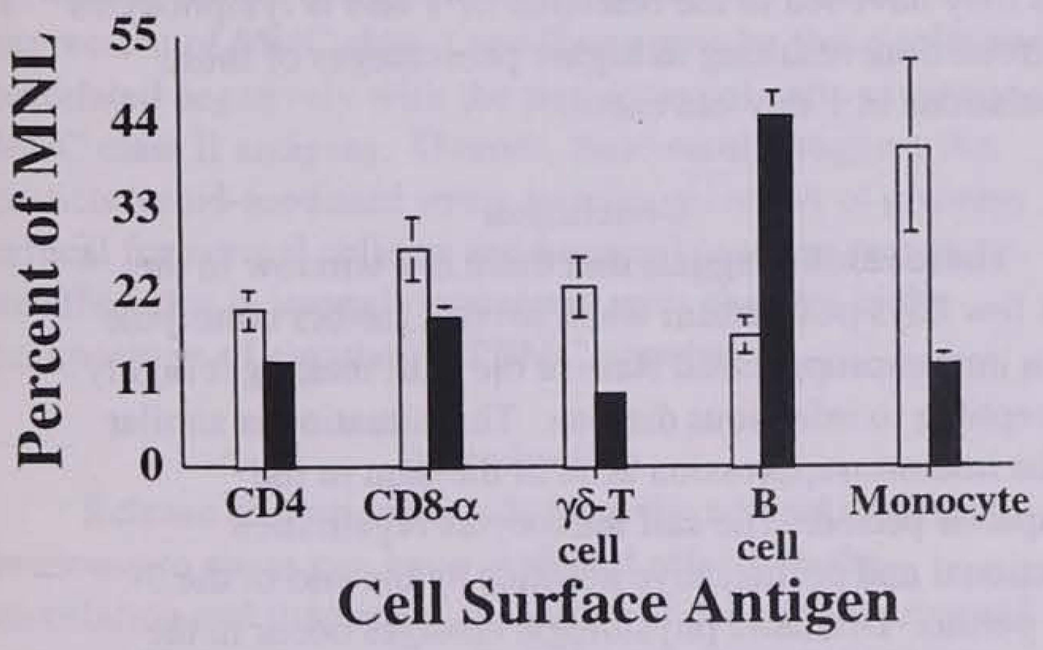


Figure 3: Mean percent of mononuclear leukocytes (MNL) from calves (open bars) or cows (solid bars) that were positive for cell surface markers.

Cultures of MNL from neonatal calves were unresponsive to mitogenic stimulation in terms of secretion of IgM. These cultures secreted a mean of 2.7 ± 0.5 ng/ml of IgM, which remained relatively unchanged over the 7-day period. Although IgM secretion by unstimulated cells from adult cows was similar to secretion by unstimulated cells from calves, adult cells stimulated with PWM secreted five to six times more IgM than did PWM stimulated calf cells (Fig. 4).

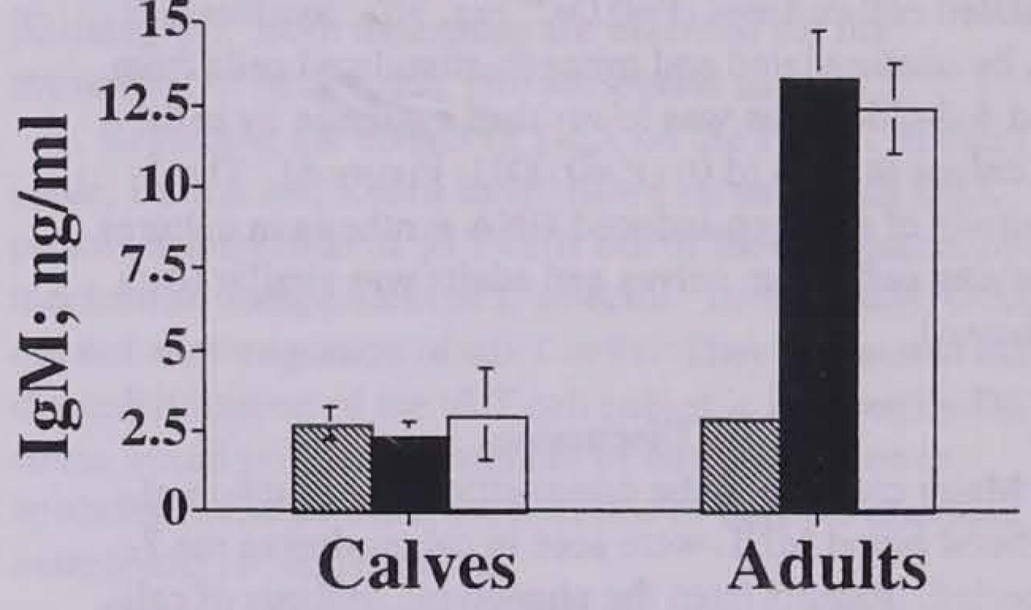


Figure 4. Mean polyclonal IgM production by mononuclear leukocytes (MNL) from calves or cows. The MNL were either unstimulated (hatched bar), or stimulated with 0.08 (solid bar) or 0.32 mg/ml of PWM (open bar).

Interferon-γ secretion by unstimulated calf cells remained low and unchanged (<55 pg/ml) during the 7-day period. Cells from 1- to 4 day-old calves that were stimulated with PWM secreted less IFN-γ than did cells from precolostral (d 0) and 7-day-old calves (P=0.003; Fig. 5).

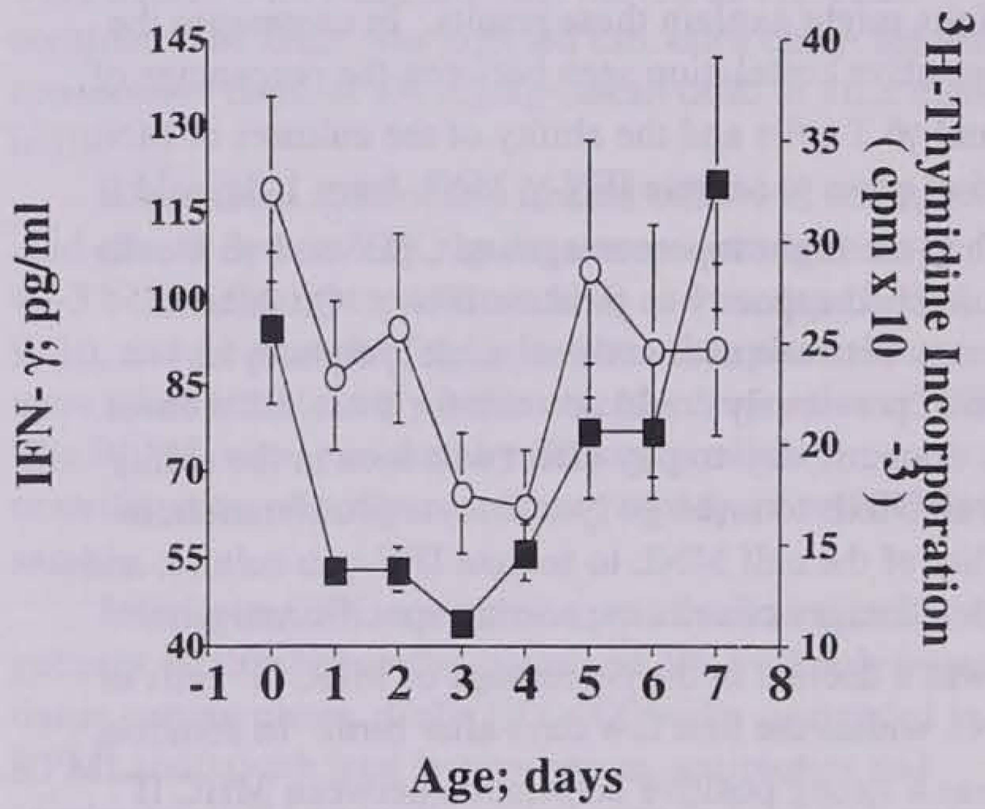


Figure 5: Mean IFN-γ (solid square) production and proliferation (open circle) by peripheral mononuclear leukocytes from calves.

Although IFN-γ secretion by unstimulated cells from adult cows was similar to secretion by unstimulated cells from neonatal calves, cells from adult cows secreted 100-

fold more IFN-γ when stimulated with PWM (8,688 ± 1,616 pg/ml vs. 73 ± 4 pg/ml produced by PWM stimulated calf cells).

Calf leukocytes stimulated with PWM or concanavalin A incorporated significantly greater amounts of [^{3}H]-thymidine than did unstimulated cells (P=0.0001). Synthesis of DNA in cell cultures stimulated with concanavalin A was greater than in parallel PWM stimulated cell cultures (P=0.002; Fig. 7b). Synthesis of DNA by unstimulated and mitogen-stimulated cells from 3- and 4-d-old calves was lower than synthesis by cells from calves at birth (d 0) (P =0.0001; Figure 5). The magnitude of mitogen-induced DNA synthesis in cultures employing cells from calves and adults was similar (data not shown).

Discussion

Major changes in the composition and function of peripheral blood MNL were seen in calves during the 7-day period. Results from the phenotypic analysis of calf MNL populations in this study support earlier reports indicating that neonatal ruminants have higher percentages of circulating γδ T cells and lower percentages of B cells than adult cows. Although mitogen-induced lymphoproliferative responses of calf MNL were similar to the responses of MNL from adults, functional capabilities (i.e., IFN-γ and IgM secretion) were much lower. Observations in humans and mice indicating that neonates have a greater proportion of naive T cells, T cells capable of suppressing Ig production by B cells, and also antigen-presenting cells with ineffective costimulatory function and lower cytokine-producing capabilities when compared with adults might explain these results. In contrast to the strong positive correlation seen between the percentage of $CD3^+$ and γδ T cells and the ability of the cultures of MNL from adult cows to secrete IFN-γ, MNL from 1-day-old calves had the highest percentages of $CD3^+$ and γδ T cells but diminished capacity to produce IFN-γ. Qualitative differences between neonatal and adult lymphocytes mentioned previously could account for these differences.

A dramatic day-to-day effect was seen in the ability of the calf MNL to undergo lymphocyte proliferation, in the ability of the calf MNL to secrete IFN-γ in culture, and in the percentages of cells expressing specific antigens. There was a decline in the percentage of MHC II^+ cells in the MNL within the first few days after birth. In addition, there was a strong positive correlation between MHC II^+ cells and the percentages of monocytes in these MNL populations and a strong negative correlation between MHC II^+ cells and $CD3^+$ and $CD4^+$ lymphocyte subsets. Stimulation of bovine MNL by PWM preferentially enhanced proliferation of $CD4^+$ T lymphocytes. Thus, in this system, the $CD4^+$ T cells were likely the major secretors of IFN-γ. A less than effective accessory cell function, i.e., lesser numbers of MHC II^+ antigen-presenting cells or lower antigen-presenting cells to T cell ratios might have contributed to the reduced proliferation and secretion of IFN-γ by MNL from these calves.

Colostrum contains higher concentrations of immunosuppressive factors such as cortisol, prostaglandins, and transforming growth factor-β when compared with milk that may have contributed to the suppression in lymphocyte proliferation and IFN-γ production seen within the first 4 days after birth. Serum $1,25(OH)_2D$ is also elevated in these calves at 1 to 2 d after birth. Supplementation of cultures of MNL from adult cattle with similar concentrations of this compound has been shown previously to inhibit cell proliferation, decrease the numbers of MHC II^+ and interleukin-2 $receptor^+$ cells, and suppresses IFN-γ secretion. The calf also has elevated plasma corticosteroid concentrations at birth (≥8 μg/dl), which decrease to ≤4 μg/dl within 24 h. Corticosteroids have profound immunosuppressive effects, presumably because they inhibit the activity of transcription factor nuclear factor κB, which is needed for the expression of numerous molecules of the immune system. Dexamethasone suppresses IFN-γ and IgM secretion by mitogen-stimulated peripheral blood leukocytes in adult dairy cows. Thus, these different factors could have contributed to the observed decrease in IFN-γ secretion, or MHC II expression, or both, as well as to the diminished lymphoproliferative responses of the MNL isolated from 1- to 4 day-old calves. The synthetic corticosteroid, dexamethasone, has also been shown to affect adhesion molecule expression in lymphocytes from 15-month-old dairy bulls. Expression of adhesion molecules on peripheral blood MNL could have been influenced by circulating glucocorticoids in 1-d-old calves. This may have led to the retention of T and B lymphocytes in circulation, resulting in higher percentages of these populations in 1-day-old calves.

Conclusion

These results suggest that there is a window in the first few days postpartum when several factors contribute to an immunosuppressed state in the calf, making it highly susceptible to infectious disease. This situation is similar to the immunosuppression seen in the dam in the postpartal period. The calf leukocytes regain their functional and proliferative abilities by the end of the 7-day period. Dramatic physiologic changes occur in the neonatal calf in the first few days after birth. A clearer understanding of these events would increase our knowledge of the health and productivity of cattle.

Function and Composition of Blood Mononuclear Leukocyte Populations from Holstein Bulls Treated with Dexamethasone

B. J. Nonnecke, PhD; J. L. Burton, PhD[1];
and M. E. Kehrli, Jr., DVM, PhD
U. S. Dept. of Agriculture,
Agricultural Res. Service,
National Animal Disease Center,
Metabolic Dis. and Immunol. Res. Unit,
P. O. Box 70, Ames, IA

DSL-138

Summary and Implications

To characterize further the effects of corticosteroid induced stress on the immune system of dairy cattle, functional and phenotypic characteristics of blood mononuclear leukocytes (PBMC) from control and treated [.04 mg dexamethasone (DEX)/kg per d for 3 consecutive d] Holstein bulls were evaluated concurrently. In vivo administration of DEX caused a profound and significant reduction in the capacity of lectin-stimulated PBMC to secrete the cytokine, interferon-γ (IFN-γ) and polyclonal immunoglobulin M (IgM). Changes in the secretion of these proteins were associated with changes in the expression (per cell) of MHC class I and II antigens and the WC1 antigen, and in the proportion of B cells, CD3 T cells, γδ T cells and MHC class II^+ cells. Secretion of both proteins was correlated positively with the percentage of $CD3^+$ T cells (primarily the γδ T cell subset) in PBMC populations and expression of MHC class I and II antigens by these cells and correlated negatively with the proportion of cells expressing MHC class II antigens. Overall, these results suggest that corticosteroid-mediated stress impairs secretion of proteins critical for normal cellular and humoral immune responses, an effect that is strongly correlated with changes in the composition of circulating PBMC population.

Introduction

Release of corticosteroids from the adrenal cortex in response to stress can have profound effects on the circulation and functional capacities of cells of the immune system The peripartum period of the dairy cow has been associated with elevations in the concentrations of plasma corticosteroids, reduction in the functional capacities of blood neutrophils and lymphocytes, neutrophilia, and an increased susceptibility to infectious diseases (i.e. mastitis). Recent research at NADC suggests that in dairy cattle the anti-inflammatory action of corticosteroids is to prevent migration of neutrophils from blood by down-regulating expression of leukocyte adhesion molecules (L-selectin and β2-integrin). Both molecules are essential for the movement of neutrophils into peripheral tissues.

Regarding the effects of DEX on the PBMC of dairy cattle, Burton and Kehrli have shown recently that DEX promotes migration of γδ T cells out of the circulation by a mechanism independent of L-selectin. In this study, DEX did not alter migration of αβ T cells. They postulated that, if a redistribution of the γδ T cell subset is induced by DEX, stress would promote movement of this population to epithelial surfaces when the first line of defense provided by neutrophils is compromised.

Conceivably, compositional changes in PBMC population induced by DEX would influence the functional capacity of this population. The objectives of this study were to evaluate in young dairy bulls the effects of DEX administration on the in vitro capacity of PBMC population to secrete IFN-γ and polyclonal IgM, and to determine whether associations existed between functional capacity and composition of the PBMC population.

Materials and Methods

Young Holstein bulls were used that had reached sexual maturity before studies were initiated. Four bulls were treated with DEX and four served as untreated controls. The DEX was injected i.m. once daily, for three consecutive days, at .04 mg/kg (mean dose of 16.8 mg/d per bull).

Blood from control and treated bulls was collected into acid-citrate-dextrose by jugular venapuncture on days -5, -4, -3 before the first treatment (d 0) and on days 2, 3, 4, 5, 9, 10, and 11 after the first injection. Samples taken on d 2 were taken immediately before the third injection of DEX. The PBMC were enriched by density gradient centrifugation of buffy coats recovered from each blood sample.

Interferon (IFN)-γ secretion was evaluated in PBMC cultures established in flat-bottomed, 96-well polystyrene tissue culture plates. Cells (200,000/well) suspended in RPMI 1640 (with fetal bovine serum, antibiotics and amphotericin B) were stimulated with 0, 5, and 10 μg/ml of pokeweed mitogen (PWM). Cultures, prepared in triplicate, were incubated at 39°C in a humidified atmosphere with 5% CO_2 for 48 hours. Culture supernatants were harvested from centrifuged plates and stored at -80°C.

Interferon-γ (ng/ml) in culture supernatants was quantified by an IFN-γ capture ELISA. The IFN-γ in test

samples was determined from a standard absorbance curve regressed on IFN-γ concentration for the dilution of test samples that gave absorbance readings falling inside the linear portion of the curve. The concentration of IFN-γ in supernatants was determined by multiplying the value from the standard curve by the dilution factor.

Secretion of polyclonal IgM by PBMC was assayed in flat-bottom, 24-well tissue culture plates inoculated with 1.5 X 10^6 cells in 1.5 ml cultures. The PBMC were stimulated with PWM (0 and .08 µg/ml), and incubated 14 d at 39°C. Supernatant (100 µl) from each well was removed after centrifugation (400 x g, 5 min at 18° C) and stored at -80°C. The IgM in supernatants from unstimulated and PWM-stimulated cultures was assayed by an ELISA. The concentration of IgM (µg/ml) was calculated by comparison of the absorbance of unknowns with absorbance of standards (serially diluted bovine IgM) fit within a linear curve.

Indirect fluorescent antibody immunostaining was used to identify T cell subsets, B cells, and MHC class I and II positive cells in PBMC suspensions. Sources, specificities, isotypes, and working dilutions of monoclonal antibodies and secondary antibody are given in a previously published manuscript (J. Dairy Sci., 80[10]:2403-2410, 1997). Antibody-labeled suspensions of PBMC were examined using a FACScan flow cytometer (Becton Dickinson). Acquired data were analyzed using Cell Quest software (Becton Dickinson). Variables recorded for each marker were percentages of cells staining positive and the mean fluorescence intensities of those cells.

Data were analysed by a split-plot type of repeated measures ANOVA. When the interaction of day and treatment was significant, differences were analysed using Student's two sample *t*-test. Pearson's product-moment correlations were computed between PBMC phenotypes and protein (IFN-γ and IgM) secretion. Statistical significance was determined at $P \leq .05$.

Results and Discussion

In vivo administration of DEX to bulls resulted in the simultaneous inhibition of IFN-γ and IgM secretion by their PBMC (Figs. 1 and 2). Changes in the secretion of these proteins were associated with significant changes in the composition of and antigen expression (indicated by MFI of antigen) by the PBMC populations (Figs. 3 and 4). These results indicate corticosteroids affected changes in the PBML population that alter the capacity of mononuclear leukocytes to secrete proteins essential for normal humoral and cellular immune responses. Effects of DEX on PBMC in cattle can be explained by recent studies indicating that corticosteroids can inhibit translocation of the transcription factor, nuclear factor IκB, to the nucleus where it normally associates with response elements essential for induction of genes for a variety of immunoreceptors, cell adhesion molecules, cytokines, hematopoetic growth factors and acute phase proteins. Previously reported changes in L-selectin and CD18 expression on bovine neutrophils and compositional changes in circulating T cell populations in cattle may be consequences of the effects of DEX on multiple genes encoding key immunoregulatory proteins.

Positive correlations between IFN-γ secretion by PBML in vitro and the percentage of $WC1^+$ T cells (consisting mainly of γδ T cells) in the PBMC population (Table 1) are interesting because published data indicate that γδ T cells secrete IFN-γ and that peak production of this cytokine during infection coincides with peak activation and recruitment of macrophages. Conceivably effects of DEX on the distribution and function of the γδ T cell would compromise the first line of defense and protective role provided by this T cell subset.

Because PWM is a T cell-dependent, B cell mitogen for bovine PBML in vitro, the positive correlations between IgM secretion by PWM-stimulated PBML and percentages of T cells ($CD3^+$, $CD4^+$, and $WC1^+$ T cells) were not surprising (Table 1). These results suggest that reduced secretion of IgM in cultures of PBMC was not only due to DEX-induced reduction of B cells in the circulation but also likely due to direct effect of DEX on the distribution T cell populations supporting by B cell function. Positive correlations between the percentage of γδ T cell in the PBML population and the capacity of this population to secrete IgM and IFN-γ suggests that the γδ T cell subset contributes to both humoral and cellular immune response in dairy cattle.

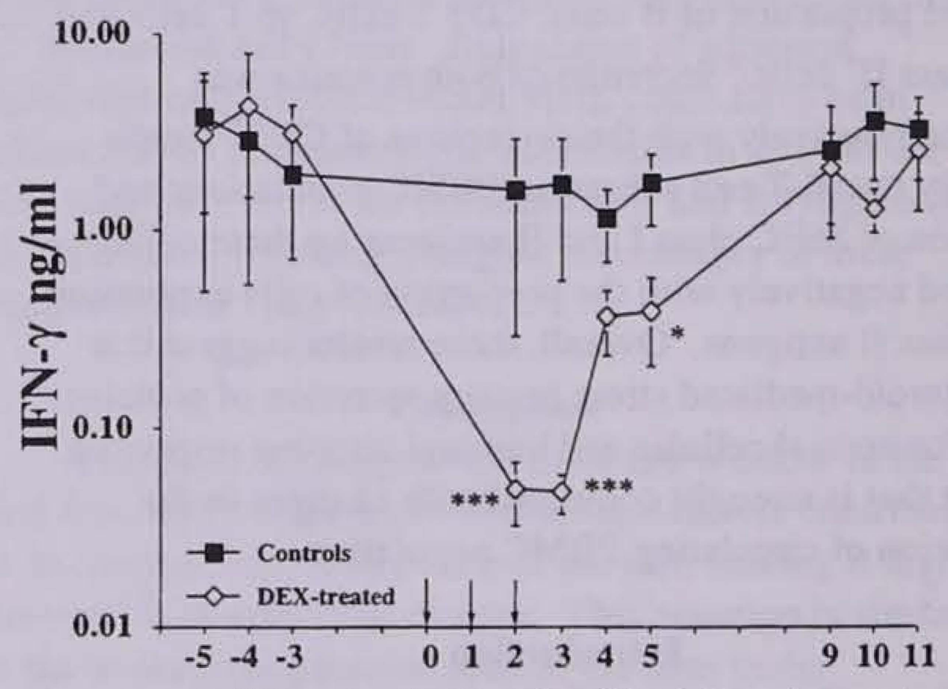


Figure 1: Mean IFN-γ secretion by cultures of pokeweed mitogen-stimulated blood mononuclear leukocytes. Leukocytes were from untreated bulls and bulls injected intramuscularly with dexamethasone. The 3 consecutive days of treatment are indicated by arrows on the abscissa. Secretion by control versus treated bulls on a specific day (*P<.05, ***P<.001)

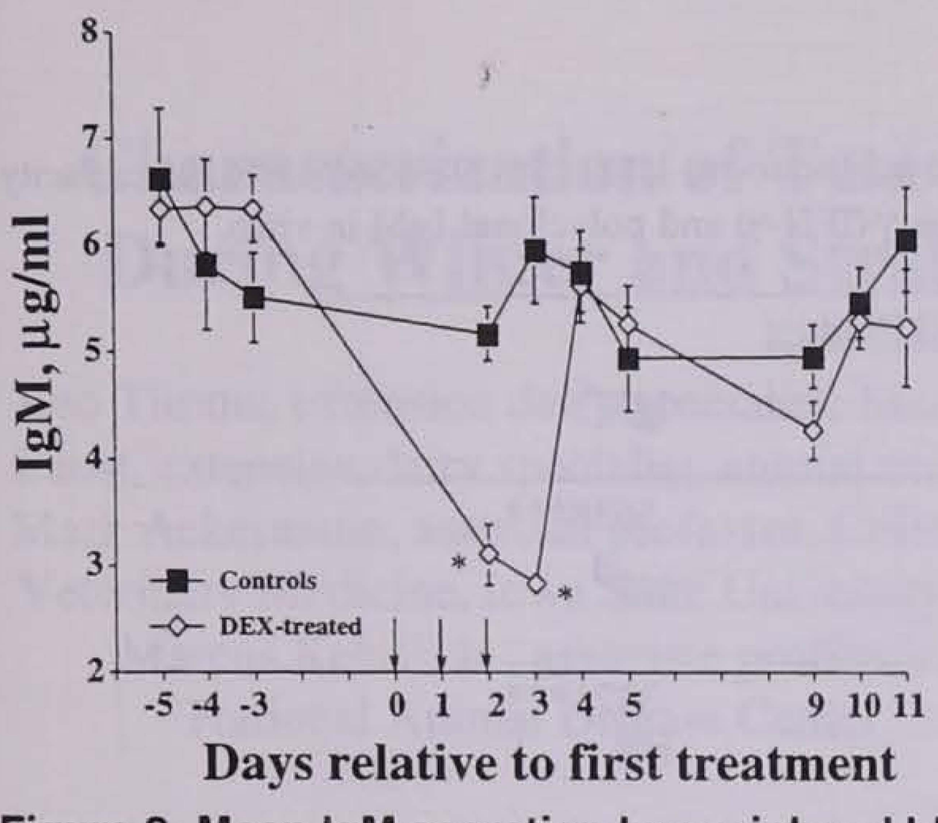


Figure 2: Mean IgM secretion by peripheral blood mononuclear leukocytes stimulated with pokeweed mitogen. Leukocytes were from untreated bulls and bulls injected intramuscularly with dexamethasone. The 3 consecutive days of treatment are indicated by arrows on the abscissa. Secretion by control versus treated bulls on specific day (*P<.05).

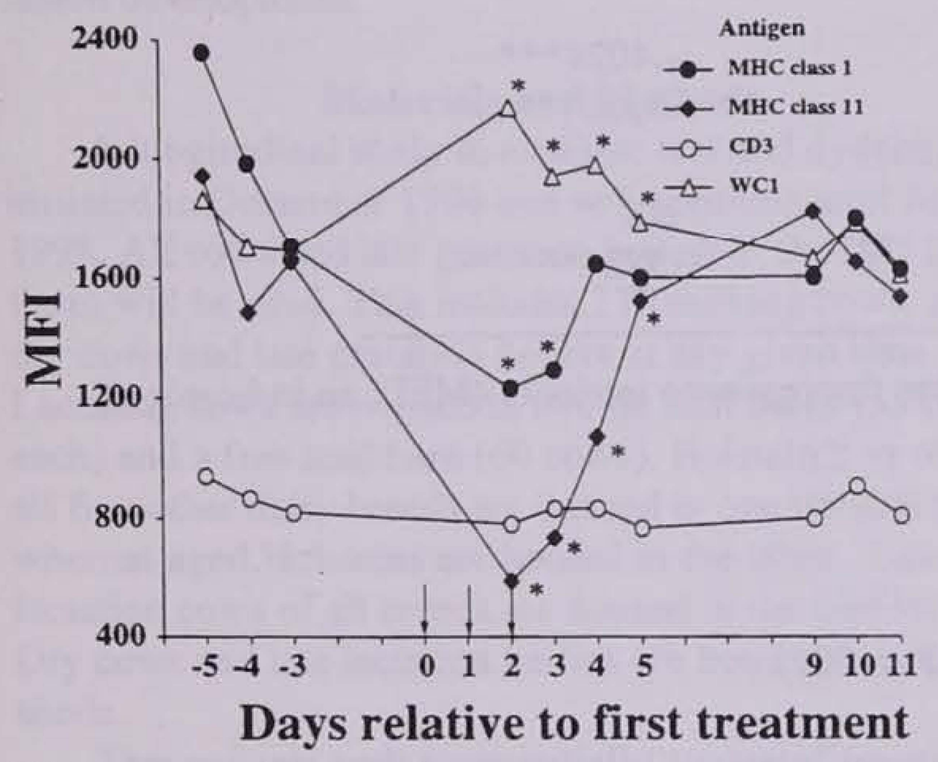


Figure 3: Mean fluorescence intensity (MFI) of various antigens on the surface of blood mononuclear leukocytes from bulls injected intramuscularly with dexamethasone. These leukocyte populations were used in cultures evaluating their capacities to secrete inferferon-g and IgM. Pretreatment versus treatment values (*P< .001).

Administration of DEX increased the percentages of MHC class II^+ cells and simultaneously reduced mean fluorescence intensities of MHC class I and II antigens (Figures 3 and 4), confirming results of studies in dairy cattle and other species. Because of the essential role of these molecules in antigen presentation and recognition, changes induced by DEX would be expected to affect adversely the adaptive arm of the immune system. Positive correlations between MHC antigen expression (MFI of MHC class I and II antigens) and specific functional capacities of the PBMC support this supposition.

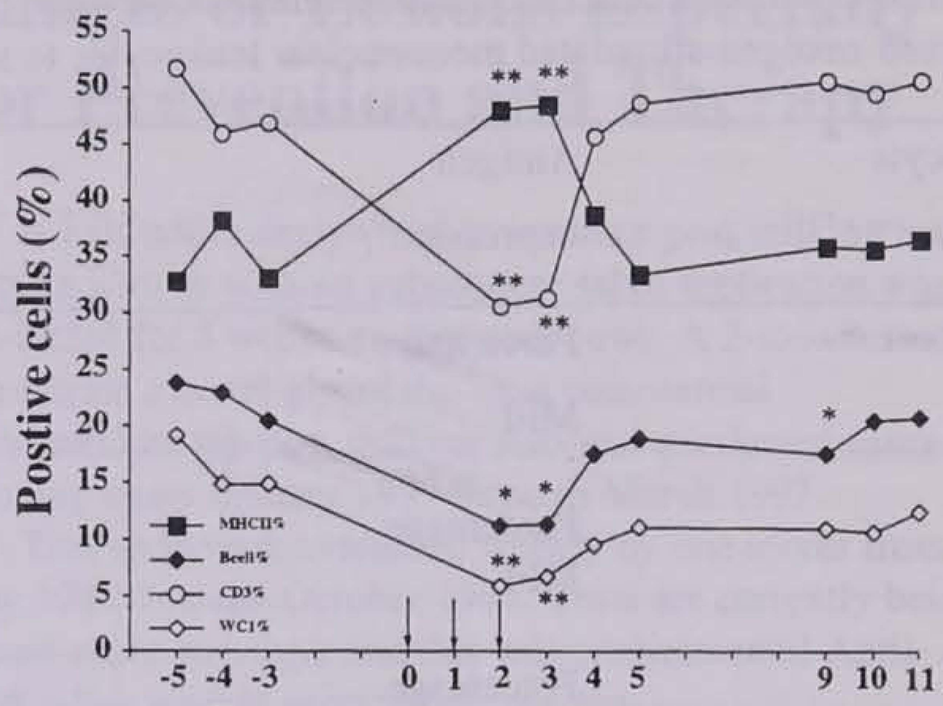


Figure 4. Mean percentages of cells expressing specific antigens within the mononuclear leukocyte population recovered from blood of bulls injected intramuscularly with dexamethasone. This leukocyte population was used to inoculate cultures evaluating their capacity to secrete interferon-g and IgM. Pretreatment versus treatment values (*P < .01, **P < .001).

In conclusion, administration of DEX to dairy cattle caused compositional changes in the PBMC population that were associated with a reduction in functional capacities of PBMC. These results support the recommendation that diagnostic tests measuring cell-mediated immunity (i.e. IFN-γ production in evaluation of *Mycobacterium bovis* infection status) be interpreted with caution in stressed cattle and cattle given corticosteroids because the chance of false negative in such tests would be greater. Similarly, the potential of natural or therapeutically induced elevations in corticosteroids to reduce responsiveness of cattle to vaccines that rely on antibody production for a protective effect should be considered.

Results from the present study linking functional and compositional characteristics of the PBMC population, suggest that the best overall predictor of the capacity of a young bull to mount a normal immune response (as measured by in vitro IFN-γ and IgM secretion) may be expression of MHC class II molecules and the percentage of γδ T cells in the PBMC population. These phenotypic variables might serve as sensitive and easily measured indicators of stress in dairy cattle if associations between composition and functional capacity of PBML are confirmed by subsequent studies.

Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that may also be suitable.

[1]Current address: Department of Animal Science, Anthony Hall, Michigan State University, East Lansing, MI, 48824-1225.

Table 1. Pearson's-product moment correlations between peripheral blood mononuclear leukocyte phenotype and the capacity of pokeweed-mitogen-stimulated mononuclear leukocytes to secrete interferon-γ (IFN-γ) and polyclonal IgM in vitro.

Leukocyte antigen	Antigen expression[a]	Cell Product IFN-γ[b]	Cell Product IgM[c]
CD3	Percentage	.5518***	.5638***
	MFI	.2351*	NS[d]
CD4	Percentage	.3201**	.3321***
	MFI	.2970**	NS
CD8	Percentage	.3984***	NS
	MFI	NS	NS
WC1	Percentage	.4716***	.5695***
	MFI	-.3024**	NS
B cell	Percentage	NS	.3096**
	MFI	NS	NS
MHC class II antigen	Percentage	-.5263***	-.4024***
	MFI	.5572***	.5222***
MHC class I antigen	Percentage	-.4176***	NS
	MFI	.5572***	.3824***

[a]Percentage of mononuclear leukocytes expressing specific antigen and mean fluorescence intensity (MFI), an indicator of antigen density per cell.

[b]IFN-γ secretion induced with PWM at 10 μg/ml

[c]IgM secretion induced with PWM at .08 μg/ml

[d]NS = correlation not significant and not shown (*P < .05; ** P < .01; *** P < .001)

Characterization of Teat End Changes or Lesions Especially During Winter and Strategies For Prevention and Therapy

Leo Timms, extension dairy specialist; Marjorie Faust, extension dairy specialist, animal science, Mark Ackermann, assistant professor, College of Veterinary Medicine, Iowa State University; and Marcus Kehrli, Jr., associate professor, National Animal Disease Center

DSL-139

The objectives of this research are to: (1) Evaluate the prevalence and dynamics of teat end lesions; (2) characterize the tissue changes associated with lesion development and regression; (3)investigate environmental, infectious, and mechanical factors associated with teat end lesions; and (4) develop therapeutic interventions to prevent or decrease lesion development.

Materials and Methods

A longitudinal study to evaluate teat end dynamics was initiated in December 1996 and will continue until July 1998. All cows and late gestation heifers at the ISU Dairy Farm will be used. This includes 170 milking cows, and 40+ dry cows and late gestation heifers at any given time. Lactating cows are housed in two tie stall barns (55 cows each) and a free stall barn (60 cows). Holstein 2 yr olds and all five other dairy breeds are housed in one tie stall barn whereas aged Holsteins are housed in the other. Late lactation cows of all breeds are housed in the free stall barn. Dry cows and late lactation heifers are housed in open sheds.

Teat and teat ends were initially evaluated (every lactation also) for shape, length, and condition. Photographs of all teat ends were taken initially to serve as a baseline. Teat ends were scored every 3 days by 2 to 3 independent scorers by using a 1 to 5 system (0.5 increments) from December 1996 through April 1997. Photos of teat ends of 18 cows (72 teats) also were taken on the day of scoring for 8 consecutive weeks (two groups of 18 different cows photographed from December 1996 through March 1997).

The teat end scoring system was as follows:

1. smooth bottom – no callous or ring
2. slightly raised callous or ring
3. raised callous or ring (teat end as a pinpoint)
4. raised ring with cracking/cuts on teat end skin
5. lesions/scabs/excessive hyperakeratosis

(1 to 5 pt scale with 0.5 increments; 3.5 = skin break/cut)

A half udder design trial to evaluate post milking teat dipping with or without subsequent salve application was conducted for 8 weeks on free stall cows. A 2-month study to evaluate a novel glycol dip vs. a commercial chlorhexidine dip post milking also was conducted using all lactating cows January 1997 through March 1997.

Teat ends were evaluated weekly by one scorer from May 1997 through October 1997. Teats are currently being scored every two days and this will continue until April 1998 when weekly scoring will resume.

Preliminary Results

Data is currently being analyzed on all aspects of this trial. Teat end scoring data and dynamics from December 1996 through March 1997 is presented in the following two figures. There was significant variation within an individual teat end score over time ($P < .0001$) and significant shifts in cow and herds average scores over the three-month period possibly associated with temperature changes and fluxes. Variation between scorers was not significantly different ($P = .45$) nor was for a given day between udder halves(left vs. right $P = .87$). Variation between teat scores of different barns approached significance (0.09 with highest scores seen in barn with older early lactation Holsteins) and scores between front and rear teats of lactating cows were significantly different (0.13 score higher in front teats). There was a significant barn by day and animal by day effect indicating teat end changes were occurring during this period.

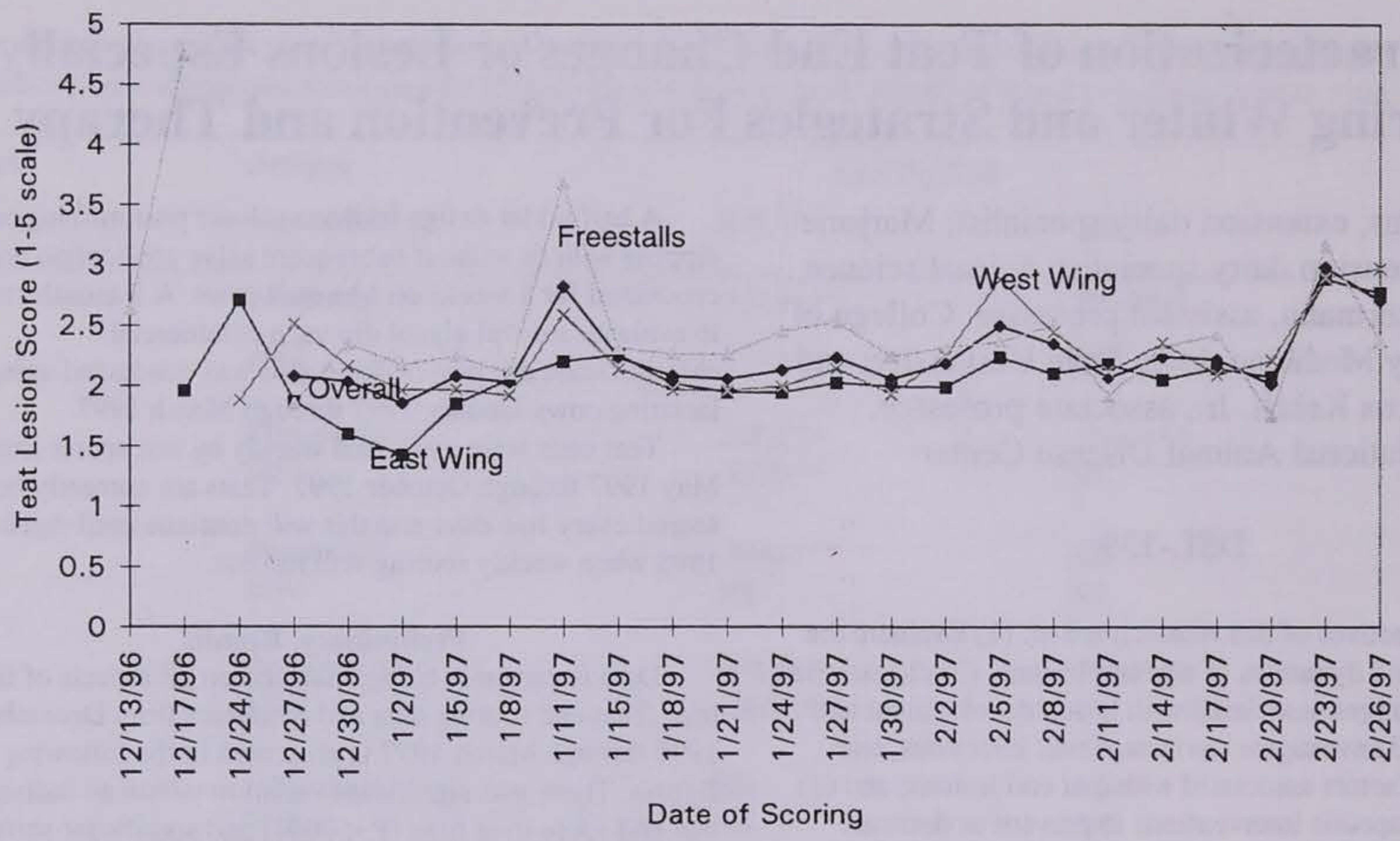


Figure 1. Mean teat end score for three barns and overall (December 1996 through March 1997).

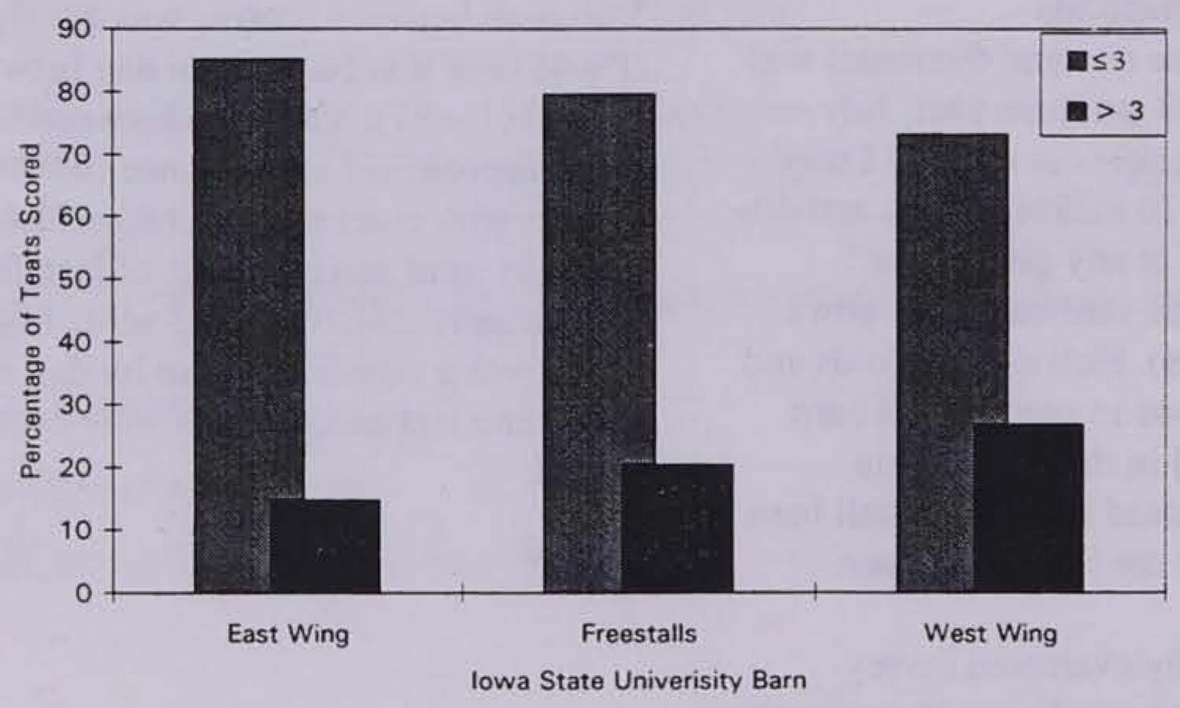


Figure 2. Distribution of teat end scores ≤3 and >3 for three different barns.

Modifications to Enhance Performance of a Novel Persistent Barrier Teat Dip for Preventing Mastitis During the Dry Period

Leah Allen, undergraduate, and
Leo Timms, extension dairy specialist,
Animal Science Department

DLS - 140

Summary and Implications

A novel persistency barrier teat dip developed at ISU was shown to reduce dry period and calving intramammary infections (IMI) in a 13 month natural exposure field trial. However, problems with dip visibility on teat ends and decreased dip persistency due to a shearing effect of the dip when temperatures were >55° F were encountered. Trials were conducted to evaluate color and viscosity modifications, with the outcome being a 12% polyurethane dip with pink pigment (no dip shearing up to 100°F and visible from 30 yards). The formulation was licensed to West Agro, Inc., K.C., and became commercially available in May 1997. Current enhancements being evaluated are germicide inclusion and different adhesion promoting compounds. One prototype dip has shown >24 hr increased protection compared to the commercial dip. A 14 month field study to evaluate the commercial dip for preventing dry period IMI and its potential to substitute for dry cow antibiotic therapy is also currently underway.

Introduction

Mastitis research has shown that 40 to 50% of intramammary infections (IMI) are contracted during the dry or non-lactating period with the greatest percentages of these occurring during the first and last two weeks of the dry period. At these times, the mammary gland is in a transitional state. Immunological factors are preoccupied or suppressed, milk is no longer being flushed from the gland, and increased mammary pressure distends the teat, thus allowing for easier bacterial penetration through the streak canal. The primary goal for mastitis control during the dry period is to minimize bacterial exposure on teat ends. A novel persistent barrier teat dip developed at ISU was shown to significantly reduce dry period and calving environmental Streptococcal and total intramammary infections by 63% and 37%, respectively, with no harmful effects on teat tissue in a 13 month natural exposure field trial. Although the dip persistency was evaluated daily during that trial, dip visibility was poor from >5 feet due to the opaque nature of the dip. Also, decreased dip viscosity associated with temperatures >55°F resulted in increased shearing of the dip film and significant decreases in teat end persistency and protection. The objectives of the following trials were to make modifications to enhance visibility, maintain an optimum viscosity across all temperatures, incorporate a germicide, and further enhance teat end persistency and protection.

Materials and Methods

Visibility: Barrier teat dips containing orange, yellow, or pink pigments were applied to forty teats each of dry cows. Visual and teat end persistency appraisal was performed every 12 hours. Dip visibility was scored from 20 feet away using a 1-3 scoring system with one being very hard to see (like original dip) and three being easy to see and evaluate. Appraisals were performed by two evaluators independently.

Viscosity: Viscosity trials were conducted at West Agro, Inc., Kansas City, Missouri. Viscosity was measured using a VL adapter and measured as cps. Initially, the original formula was evaluated at temperature ranges from 30-100°F. This curve was used to estimate the viscosity at 50-55° where dip began to shear and was used as a minimum baseline threshold for modified dips. The original formula contained 10% polyurethane. Modified dips included 12.5 or 15% polyurethane, or a .5% thixotropic (non-dripping) agent. Test tubes also were dipped to evaluate product usage rates/dip. A trial using twelve cows (48 teats) and four dips (3 modifications plus original) randomized within cow was conducted to evaluate dip persistency.

Adherence: Two modified dips containing both a germicide and different adhesion promotors were tested against the commercially available dip which now contains the color and viscosity enhancements achieved in the first two experiments. Twelve cows (48 teats) were dipped in a randomized fashion with two teats dipped in the commercial dip (double blind) and each of the other two dipped with a modified dip. Teat end persistency and protection was evaluated every 12 hrs.

Results and Discussion

Visual appraisal of three modified dips with different coloring agents is shown in Table 1. Teats dipped in the pink formulation had superior visibility compared to yellow and orange and scored 2.8 or better up to 156 hours post dipping. The pink dip was also visible from 100 feet. Persistency of these dips is shown in Table 2. Teats dipped with the pink formulation had the highest percentage of teats protected >3 days. A separate trial using five cows and four dips (three colored and original) randomized within cow was conducted. The dip containing the pink formulation showed superior visibility to all other dips and persistency was equal (40%) or better (60%) compared to the other dips. Based on these results, the pink pigment was chosen to maximize dip visibility.

Viscosity data on the original dip (10% polyuretane (PU)) is shown in Table 3. Problems in the field associated with dip viscosity between 50-70°F correspond to a drop in viscosity from 154 to 119 cps in the laboratory. It was decided that modified products had to have at least 154 cps at 100°F to assure product integrity and eliminate shearing. Viscosity data on the original dip (10% PU) and three modified dips (12.5% and 15% PU, .5% thixotropic agent) is shown in Table 4. All three modified dips showed viscosities >154 cps at 100°F. Teat dip persistency or protection of these dips is shown in Table 5. All three modified dips showed greater persistency than the original dip. The 15% PU was too viscous at lower temperatures making dipping and uniform coverage very hard. The .5% thixotropic dip had stability problem with the dip increasing viscosity over time. Based on these results, 12.5% PU was chosen to optimize dip flexibility and persistency.

Adherence data on the commercially available persistent barrier teat dip product (original formulation with color and viscosity change) compared to two new formulations containing a germicide and different adhesion promoting compounds is shown in Table 6. Dip D had greater persistency than the other three dips with a 18 to 30 hr greater average minimum retention time, and had the highest persistency within cow 75% of the time. Different modifications of dip D are now under evaluation.

Table 1 Visual appraisal of persistent barrier teats modified to enhance visibility.

	Average Visual Score Post Dipping*					
Color	12 hrs	24	36	48	72	96
Orange	2.6	2.4	2.2	2.2	2.0	1.7
Yellow	2.9	2.8	2.6	2.2	2.2	1.7
Pink	3.0	2.9	2.9	2.9	2.9	2.8

*Forty teats/product evaluated from 20 feet away; 1 = very hard to see; 2 = fair to see; 3 = easy to see and evaluate; appraisal by two evaluators independently.

Table 2 Teat end persistency or protection using modified persistent barrier teat dips with different coloring agents.

	Teat end persistency or protection (%)	
	2-3 days	>3 days
Orange	56	44
Yellow	56	44
Pink	20	80

Table 3 Viscosity of a persistent barrier dip containing 10% polyurethane at different temperatures.

Temperature	Viscosity (cps)
30°F	182
50°F	154
70°F	119
90°F	94
100°F	87

Table 4 Viscosities and average dip adhering/dipped test tube using persistent barrier teat dips containing 10, 12.5, or 15% polyurethane (PU), or .5% thixotropic agent (T).

Temperature	Viscosity (cps)			
	10% PU	12.5% PU	15% PU	.5T
40°F	180	378	1086	358
70°F	126	236	662	252
100°F	90	180	462	182
Avg dip adhering(g)	.025	.032	.038	.032

Table 5 Teat dip persistency or protection using persistent barrier teat dips with 10, 12.5, or 15% polyurethane or .5% thixotropic agent.

Dip	Teat End Persistency or Protection %	
	2-3 days	>3 days
10% Polyurethane	50	50
12.5% Polyurethane	41	59
15% Polyurethane	25	75
.5% Thixotropic	34	66

Table 6 Teat dip persistency or protection using a commercially available persistent barrier teat dip and 2 modified dips containing germicide and different adhesion promoting compounds.

	Teat end protection (%)	
Dip*	2-3 days	>3 days
A	58	33
B	25	50
C	50	42
D	25	75

*Dips A and C are commercially available dip (Stronghold, West Agro, Inc.) while B and D are modified dips containing germicide and different adhesion promoting compounds.

Evaluation Of Teat Dip Usage Comparing A Two Chamber Siphon Dipper Against A Pressurized Dip Gun System

Leo Timms, extension dairy specialist, Animal Science Department; and Ron Nyman, ISU dairy farm parlor manager

DSL - 141

Summary and Implication

A comparison of two different methods to apply postmilking teat dip was conducted. The two methods were: A) conventional two chamber siphon dipper; and B) pressurized dip gun. Using the pressurized dip gun resulted in a 44% decrease in dip usage compared to the two chamber siphon dipper. Returning to the two chamber siphon dipper following the pressurized gun system resulted in an 11% increase in dip usage. However, this was a 37% decrease in dip usage compared to the same method used prior to using the pressurized system emphasizing the educational role that the pressurized system played in milker awareness of dip usage and wastage.

Introduction

The concept on postmilking teat dipping has been around since 1916. The original main purpose of teat dipping was to apply an effective germicidal product to kill contagious mastitis pathogens potentially found on the residual milk on teat skin following milking. This technology, in conjunction with dry cow antibiotic therapy, was highly adopted in the 1960's as two major keys to controlling contagious mastitis. As contagious mastitis has been controlled, environmental mastitis has surfaced as the major mastitis problem in many herds. With this has come the development of specific dips, like barrier dips, to target these problems. Also, with the concerns of teat chapping and teat end lesions resulting from winter weather, new dips such as powders and glycol based dips, and methods such as drying teat ends after dipping have been developed. As a result, there are a myriad of dips for different conditions with similar or different germicide content and percentages, skin conditioners, efficacies, and prices, often making it a complicated decision for many producers.

Profitability is key to the success of a dairy operation. Controlling costs is a major factor in this. Teat dips, in relation to feed and labor costs, are not a major expense to the dairy operation, but often times receive considerable scrutiny concerning cost, and many teat dip decisions are often made in pursuit of a lower cost product. Also, many times dips are blamed for mastitis problems, and it is a very easy decision to change teat dips to improve efficacy and/or lower costs. As a result, many teat dip decisions result in similar or lower efficacy, even though cost may be decreased.

Most attention has been spent on the dip product cost, rather than the efficiency or dip usage rate. The advent of sprayer systems for dip application has resulted in significantly higher teat dip costs to achieve similar results to dipping, and poorer mastitis control when spray is not applied properly. High usage rates and costs often lead to decisions to purchase cheaper dips. The focus of effective and efficient teat dipping is to select the most efficacious product for that specific farm operation and apply the dip in a manner that maximizes coverage yet minimizes usage and cost. The objective of this study was to evaluate dip usage rates comparing conventional dipping to a pressurized dip system.

Materials and Methods

This research was conducted at the ISU Dairy Teaching Farm. Cows are milked in a single eight herringbone parlor. Number of cows milked daily ranged from 150 to170 over the trial and cows were milked 3x from January until October 1997 and 2x from November to December. Milking was performed by three full time milkers and many part time student milkers. A conventional two chamber siphon dip cup was used for post milking teat dipping from January to April. A portable PowrDipper teat dipping system (RJB, Inc., Modesto, CA) was put at the farm in April with all milkers required to use it. The system consists of a one liter bottle coupled to a dip gun that has a pistol grip that can be pressured using an air bulb. The dip gun was researched and developed to provide adequate dip for four teats (4 to 5 ml) and maximize displacement properties to get effective coverage yet minimize spillage. The bottle is set in a container holder and is attached to a belt that can be worn by the milker. The belt also contains a metal holder to clip on the dip gun. This system was used from May to August. In early September, the herd returned to the two chamber dip cups due to a hole in the PowrDipper hose from September to October. The PowrDipper was reinstituted at the farm from November to December 1997. Number of cows milked and dip usage rate was monitored for the entire year.

Results and Discussion

Dip usage results are shown in Table 1. Initial usage rates using the doubled chamber dipper (Jan to April) were 8.9 ml or .31 oz/dip (dip = all 4 quarters or one cow). This would result in an annual dip usage/cow of 1.8 or 2.7 gallons/cow (2x or 3x). Dip usage rates using the PowrDipper system (May-Aug) were 5.06 ml or .174 oz/dip. This equates to a 44% reduction in teat dip usage compared to the double siphon dipper. Dip usage rates using the

double chamber dipper (Sept-Oct) were 5.6 ml or .193 oz/dip. This equates to an 11% increase compared to resulted in dip usage rates of 4.96 ml or .171 oz/dip (similar to previous PowrDipper rates).

Using the pressurized dip system resulted in a 44% decrease in dip usage compared to the original dipper. Returning to the original dipper following the pressurized dip system resulted in an 11% increase in dip usage. However, this was a 37% decrease in dip usage compared to PowrDipper but a 37% decrease from the original dipper results. Using the PowrDipper after the hose was fixed the same dipper used prior to the pressurized system, emphasizing the educational aspect of dip usage or wastage accomplished by the pressurized system as well as the smaller dip chamber. This data emphasizes the importance of evaluating efficiency of dip usage (wastage) and its importance in evaluating overall teat dip cost factors.

Table 1 Teat dip usage rates using a conventional two chamber siphon dip cup and a portable pressurized dip cup system.

Dip Method	Dates	Usage Rates/Dip*	
		ml	oz
Two chamber siphon dipper	January to April	8.9	.31
Pressurized dip gun	May to August	5.06	.174
Two chamber siphon dipper	September to October	5.61	.193
Pressurized dip gun	November to December	4.96	.171

* Dip = all 4 teats of 1 cow

Evaluation of Recommended and Extended Pirlimycin Mastitis Therapy for Recent and Chronic High SCC Cows in Two Herds

Leo Timms, extension dairy specialist,
Iowa State University

DSL-142

The objectives of these field investigations were to: (1) examine the etiology and mastitis infection dynamics in two herds during the winter of 1997 whose normal herd bulk tank somatic cell counts (SCC) were between 350 to 500,000 cells/ml but had jumped to 1 million and 2.3 million in the two herds, respectively; and (2) evaluate the efficacy of recommended and extended therapy with pirlimycin hydrochloride (Pirsue, Pharmacia/Upjohn Co. Kalamazoo, MI), for gram positive intramammary infections (IMI) based on duration of elevated individual cow SCC above 300,000 cells/ml.

Materials and Methods

Two commercial herds with herds bulk tank SCC normally between 350 to 500,000 were chosen for this trial. The SCC in these herds had elevated to 1 million (herd 1) and 2.3 million (herd 2) during mid-January 1997. Herd 1 had 57 cows but only 29 high SCC cows were initially sampled. Herd 1 was housed in a loafing barn and milked in a 12-stall flat parlor with a highline milking system. Milking sanitation included predipping and postdipping with an iodine dip and drying with individual paper towels. Milking machine analysis showed the machine and system to be functioning properly. Herd 2 originally had 120 cows but only 86 were sampled initially due to culling 20 animals and drying off the rest to try and lower SCC. Herd 2 was housed in free-stall barns and milked in a double six parlor with weigh jars. A previous visit to this farm in August 1996 showed milking equipment problems (regulator, pulsators) but these problems had been fixed and the system was functioning properly. Milking sanitation included pre- and post-teat spraying, washing udders with a rag and water after pre-spraying, and drying with paper towels. Both herds had evidence of winter teat end lesions and SCC increase was seen following a cold snap.

Aseptic duplicate quarter milk samples were taken at milking time for subsequent bacteriological analysis at the start of the trial and at 40-days post treatment. Quarters with gram positive IMI were treated intramammary 2X with one 10-ml plastet containing 50 mg of pirlimycin HCl with the second treatment 24 hr after the first if that individual animal's SCC had been >300,000 for <60 days prior to the trial. Animals with chronic high SCC had this therapy strategy repeated three times in succession with each series of treatments starting 48 hr post last 2X treatment series.

Results

Seventeen cows and 27 quarters of the initial 29 cows sampled were treated with the recommended treatment protocol in herd 1. *Staphylococcus aureus* was found in 14 cows and 19 quarters (17 new high SCC, two chronic) and Streptococcus uberis was present in eight quarters of seven cows. Cure rates are shown in Table 1. Cow and quarter cure rates for *S .aureus* were 7 and 21%, respectively (8 and 24% if the two chronic high SCC quarters were excluded from the data). Cow and quarter cure rates for *S. uberis* were 71 and 63%, respectively.

Herd 2 had 71% of cows (61 of 86) and 46% of quarters (158 of 344) infected with *S. aureus*. A total of 46 cows and 123 quarters was chosen for treatment. Sixty-three quarters of 27 cows whose SCC had been >300,000 for <60 days received the recommended treatment protocol. Cure rates are shown in Table 2. Cow and quarter cure rates for *S. aureus* were 0 and 3%, respectively with a 3% quarter new infection rate during that period. Sixty quarters of 19 cows were presented for extended therapy (48 quarters of 16 cows with chronic high SCC, 12 quarters of 3 cows with recent SCC increase (<60 days)). Cow and quarter cure rates for *S. aureus* were 0 and 6%, respectively, for chronic high SCC cows, and 0 and 50% for recent high SCC cows.

Cure rates for *S. uberis* in herd 1 were similar to previous studies. Quarter cure rates for *S. aureus* in herd 1 were higher (24%) compared with herd 2 (3%) in cows with recent high SCC and recommended treatment strategy, although cow cure rates were both <10%. Lower herd prevalence and multiple quarters/cow infected with *S. aureus* combined with better milking management may explain these differences. Cure rates for recent high SCC cows was less than expected based on research showing high cure rates for *S. aureus* if the infection is treated within 60 days post-infection. Our cure rates may substantiate that elevation in SCC may not be a good indicator of initiation of infection, especially in high prevalence herds. Quarter cure rates for extended therapy were higher in recent high SCC cows (50%) compared with chronic high SCC cows (6%) although cow cure rates were both zero. Extended therapy of recent high SCC cows showed greater cure rates than recommended therapy. Further studies to evaluate if extended therapy for recent high SCC cows in a high prevalence *S. aureus* is cost beneficial are warranted.

Table 1. Cure rates for herd 1 (all treated with recommended pirlimycin therapy*).

	*S.uberis***	*S.aureus****	
	%	%	%
Cow cure rate	71	7	8*****
Quarter cure rate	63	21	24*****

* Treated intramammary 2X with one 10 ml plastet containing 50 mg of pirlimycin HCl 24 hrs apart.
** Seven cows; eight quarters. *** Fourteen cows, 19 quarters.
***** Cure rates for 17 quarters with SCC >300,000 for < 60 days (two chronic quarters excluded).

Table 2. Cure rates for *S. aureus* (herd 2) using recommended or extended pirlimycin therapy.*

	Recommended therapy**	Extended therapy (%)	
	%	chronic***	recent**
Cow cure rate	0	0	0
Quarter cure rate	3	6	50
Quarter new infection rate	3		

*Recommended therapy = treated intramammary 2x with one 10 ml plastet containing 50 mg of pirlimycin HCl 24 hr apart; extended therapy = three series of recommended therapies 48 hr apart.
** Cows with SCC >300,000 for < 60 days; (recommended 27 cows, 67 quarters; recent extended: three cows, twelve quarters).
*** Cows with SCC >300,000 for > 60 days (16 cows, 48 quarters).

Development of an Improved Model for *Mycoplasma bovis*-induced arthritis, pleuritis, and tenosynovitis

A.L. Menke, veterinary student

DSL-143

Abstract

Seven clean caught Holstein calves, fed pasteurized colostrum were raised in hutches located in a clean pasture. Of these, five calves were inoculated intrathoracicly (ITh) with 4×10^8 CFU/ml. and two calves were inoculated subcutaneously (SQ) with 4×10^8 or 1×10^8 CFU/ml. of *Mycoplasma bovis* (MB) isolate from a recent field case. Two calves were necropsied at five days and three calves at eight days post-inoculation. Macroscopic lung lesion scores ranged from 12 to 17% of the lung affected in MB-inoculated calves. Microscopic lung lesions in the ITh-inoculated calves were characterized as fibrinosuppurative and histiocytic pleuritis. The three calves necropsied at eight days had moderate to severe clinical lameness and marked swelling in the hocks. These calves exhibited severe necrosuppurative periarthritis, arthritis, and tenosynovitis. Histopathological examination of the joint synovium revealed fibrinosuppurative and histiocytic inflammation with the formation of abscesses surrounding areas of caseous necrosis. *M. bovis* was cultured from all lung lobes, rear limb joints, tracheobronchial lymph nodes, gastrointestinal pools (abomasum, small intestine, and colon), and organ pools (liver, kidney, and spleen) from MB-inoculated calves. Calves inoculated subcutaneously lacked any clinical illness other than measurable subcutaneous foci of inflammation, hemorrhage, and early abscess formation ranging from 1.5 cm. x 3.0 cm. to 5.5 cm. x 5.5 cm. in size at the inoculation sites. Histologically, these inoculation sites were characterized by multifocal vasculitis with a fibrinosuppurative and histiocytic reaction in the subcutaneous portion of the dermis. The intrathoracic model described should be useful for study of MB-induced arthritis and pleuritis.

Key words: arthritis; intrathoracic inoculation; decubital abscesses; *Mycoplasma bovis;* pleuritis

Mycoplasma bovis (MB) is associated with a variety of bovine diseases, including arthritis and pneumonia of calves and feedlot cattle, mastitis, and genital tract infections.[4,6,7,9,13] Due to the variety and frequent occurrence of such diseases, MB is considered the most virulent bovine mycoplasma in the United States.[11] Several reports of MB co-infections exist where calves were concurrently inoculated with MB and *Pasturella haemolytica*, respiratory syncytial virus, and/or bovine viral diarrhea virus and *P. haemolytica*.[8,14,19] Lesions associated with MB alone were described as suppurative bronchiolitis with varying degrees of lymphoreticular hyperplasia and focal areas of caseous necrosis surrounded by mononuclear cells.[1] In addition, a dramatic increase in the severity of illness and extent of pneumonic consolidation occured when MB was inoculated prior to *P. haemolytica*.[8]

Research efforts have resulted in various methods to detect MB both antemortem and postortem. *M. bovis* has been cultured from blood, transtracheal washes, and nasopharyngeal swabs, and has been demonstrated in the lungs, liver, kidneys, joint synovium, milk and brisket regions utilizing nucleic acid probes,[10,15] polymerase chain reaction (PCR),[10] immunohistochemistry,[2] and polyclonal serum-based immunoperoxidase techniques.[18] With such techniques in place, MB is more commonly diagnosed today in diagnostic laboratories and clinics than before when such techniques did not exist. Moreover, MB is a common isolate from arthritic joints in diagnostic laboratory cases that do not always accompany bovine pneumonia, and the reason for this unique association is unknown.[12]

The upper respiratory tract is thought to be a natural reservoir of MB.[3,11] Although the pathogenesis of MB remains unknown, it has been suggested that ingestion of MB-infected mastitis milk, direct contamination of devitalized tissues with nasal discharges due to licking, and direct nasal contact are probable causes.[3,12]

Calf pneumonia resistant to antibiotic therapy is frequently encountered in the United States and Canada.[17] With increased efforts and success in the diagnosis of MB-induced disease, it is now more evident that MB is present in many of these cases and that few therapeutic regimens, efficacious vaccines, or specific preventative measures are available to combat MB-associated diseases.

It was the intent of this research project to develop a predictable and reproducible animal model that can be utilized to further study the epidemiology, pathogenesis and treatment of MB-induced disease. Several calf models have been previously described for MB-induced disease. Many of these models did not prove to be reliable. Calves inoculated with an intravenous route did not have consistent sites and numbers of joints exhibiting arthritis.[6] Intraarticular and intratracheal inoculations utilized unrealistically high dosage levels.[16,19] Pneumonia or severe illness was not produced by calves infected by an intranasal route.[9]

Two routes were chosen for this study based on the type of disease desired (pneumonia and abscess formation), incubation time, and dosage levels. Rather than using gnotobiotic calves, which are costly and not readily available,[19] precautions were taken to obtain calves in a sterile manner by a clean catch into sterile canvas bags and immediate removal from the farm. Intraperitioneal inoculations have been found to have no effect given in low

dosages, therefore low dosages were introduced intrathoracicly into the lung tissue (Rosenbusch, unpublished). A subcutaneous route is useful in the study of cellular response to MB and allows for palpation of the progression and regression of abscesses in the live animal.[5]

Materials and Methods

Seven Holstein calves were vaginally delivered into sterile canvas bags and immediately removed from the supplying dairy farm and placed into calf hutches in a pasture. Colostrum was obtained from the dairy farm supplying the calves, pooled, heat treated to 56°C for 30 minutes, and frozen at -30°C for later use. Although the dairy farm supplying calves had been continuously monitored for mycoplasmal diseases, colostrum samples were cultured before and after heat treatment and were found to be negative. Following off-site transportation, each calf was tube fed two liters of colostrum and administered 10 cc of *Clostridium perfringens* type C & D antitoxin subcutaneously, 100 mg. of gentocin orally and 100 mg. intramuscularly, one ml. of vitamins A & D, and 2.5 ml. of (one mg. of selenium and 50 mg. of vitamin E) of BO-SE®, intramuscular (Schering Plough). Subsequent feedings of a commercially available, antibiotic-free milk replacer were given twice per day. Rectal temperatures and fecal scores were monitored twice per day.

Experimental Inoculation. Five calves were inoculated intrathoraccicly (ITh). *M. bovis* 428E was isolated from a 1994 field case submitted to the Iowa State University Veterinary Diagnostic Laboratory (ISU-VDL) from a 400 lb. calf exhibiting pneumonia and arthritis. The calves inoculated ITh received 4 x 10^8 MB 428E passage six suspended in five ml. of sterile phosphate buffered saline (PBS). Calves were restrained and an area over the fifth and sixth ribs was surgically prepared, and locally anesthetized. The inoculate was introduced into the thoracic cavity via a three-and-a-half inch, 18 gauge trocar. The trocar was inserted four inches below the thoracic vertebra wings between the fifth and sixth ribs, in a caudal-ventral direction. The inoculation was repeated on the opposite side of the calf with a new trocar. A control calf received sterile PBS in the same manner.

Two calves were inoculated subcutaneously (SQ). These calves were inoculated with 4 x 10^8 or 1 x 10^8 CFU/ml. of MB per site suspended in two ml. of sterile PBS or sterile PBS alone. A surgical preparation was made on the dorsum of the calf. Beginning at the withers, one inch off the midline, inoculations were made two inches apart in the subcutaneous layer beginning with 4 x 10^8 CFU/ml., followed by 1 x 10^8 CFU/ml., and sterile PBS. The sequence was repeated two times per side between the withers and the tail head so that there were four replicates on each calf. A total of six inoculation sites on each side of the calf were given.

Antemortem Sample Collection and Analysis. Blood and nasal swab samples were cultured on day -2, day 0 (day of inoculation), and daily after inoculation until necropsy. Blood and nasal swab samples were cultured on Friis agar with three inhibitors (thallium acetate, ceflobid, and bacitracin) and incubated for 48 hours at 37°C in five percent CO_2.

Nasal swabs were vortexed in 0.9 ml. of sterile PBS and removed. The PBS was diluted in Friis broth with three inhibitors. Samples were also cultured in Friis broth with the three inhibitors at 10^{-1}, 10^{-2}, and 10^{-3} concentrations and incubated for 48 hours. At the end of incubation the broths were examined for growth and the 10^{-2} dilution was cultured on Friis agar with three inhibitors and incubated for 48 hours.

Postmortem Sample Collection. Two of the ITh-inoculated calves were necropsied at five days post inoculation (DPI) and three were necropsied eight DPI. The SQ-inoculated calves were necropsied at six or eight DPI. The tracheobronchial lymph node (TBLN), each lung lobe, gastrointestinal pool (abomasum, ileum, jejunum, duodenum, and colon), and an organ pool (liver, kidney, and spleen) were homogenized in 0.9 ml. of sterile PBS. The suspension was cultured directly and diluted in broth as previously described. Joint aspirates were collected on each limb from long bone joints and cultured. In addition, the subcutaneous reaction sites from the two calves inoculated SQ were each homogenized and cultured. Tissues collected for histopathology included the brain, nasal turbinate, tonsils, TBLN, all lung lobes, heart, abomasum, ileum, jejunum, duodenum, colon, liver, kidney, spleen, and synovial membranes from each limb. In the SQ-inoculated calves, injection site tissues were also collected and labeled for individual identification. Tissues were fixed in 10% neutral buffered formalin and routinely processed to paraffin blocks in an automated tissue processor. Blocks were sectioned and stained with hematoxylin and eosin.

Results

Intrathoracic Inoculation. No clinical signs or fever were observed in the control calf receiving sterile PBS alone and no evidence of pneumonia or arthritis was observed at necropsy. Clinical signs were present in all four inoculated calves with MB 428E (Table 1). All ITh-inoculated calves exhibited signs of depression. The three calves that were necropsied on day eight also exhibited moderate lameness. Lameness was most pronounced in the rear limbs and marked swelling was observed in the rear hock joints. The swelling was bilateral in all but one calf. Upon palpation the hocks were warm and painful. By day eight the calves were usually reluctant to stand or move. The calves appeared anorexic, dehydrated, and rough-haired.

M. bovis was isolated from the cultures of blood and nasal swab samples of all ITh-inoculated calves, and was not cultured from the control calf (Table 2). *M. bovis* was cultured from all lobes of the lungs, TBLN, and joints from all MB-inoculated calves. In addition, two calves (104 and 105) had MB cultured from the organ and gut pools. Lung suspensions and joint aspirates from each calf were pooled and submitted to the Iowa State University Veterinary Diagnostic Laboratory for microbiological examination. No bacteria other than MB were isolated from the joint pools of

any calves. The lung pool suspension from one calf (101) had growth of *P. haemolytica* in low numbers. No other pathogenic bacteria were isolated.

Macroscopic lung lesion scores ranged from 12 to 17% involvement in the ITh-inoculated calves, while the control calf had no visible lesions (Table 3). The most prominent lesion in the ITh-inoculated calves was locally extensive pleuritis most evident on the right and left caudal lobes near the inoculation sites. In calf 102, the craniolateral portion of the left hock contained a 5 cm. x 3 cm. abscess. Incision into the skin overlying the hock revealed a straw-yellow, viscous exudate that extended the length of the tendon sheath. Similar exudate was recovered from calves 104 and 105 but no abscesses were found. Upon examination of calf 104, similar exudate was found in the brisket region. Both connective tissue and muscle were involved in the lesion although no abscesses were visible grossly.

Upon histopathlogic examination of the calves, no abnormal findings were reported in the control calf. The lung specimens from MB-inoculated calves revealed moderate, locally extensive, fibrinosupprative and histiocytic pleuritis. Neutrophilic inflammatory cells were present in high numbers (Figure 1). No changes were observed in the alveoli or praibronchiolar regions. Examination of the synovial membranes showed focal areas of caseous necrosis surrounded by fibrinosuppurative and histiocytic exudate, and early proliferation of connective tissue. The formation of abscesses was evident in calf number 105 (Figure 2). The brisket area of calf number 104 was also submitted for histopathology. Fibrinosuppurative inflammation and edema separated muscle fibers. Multifocal areas of caseous necrosis were also present in the brisket (Figure 3).

Subcutaneous Inoculation. No clinical signs were evident in the calves given subcutaneous injections of MB 428E or sterile PBS. Blood and nasal swab samples collected did not culture positive for MB. Calf 201 had palapable swelling near inoculation sites on day two. Calf 202 was sensitive to touch on day three after inoculation, on day four swelling could be palpated and visually observed. No measurements were taken antemortem because the perimeter was not well defined.

Calf 201 and 202 were SQ-inoculated calves that were necropsied on days six and eight post-inoculation, respectively. When the carcass was skinned, abscesses and discrete SQ foci of hemorrhage and edema were evident (Table 4). These foci ranged in size from 5.5 cm. x 5.5 cm. to 1.5 cm. x 3.0 cm. No gross or microscopic lesions were noted in the lungs or joints of the SQ-inoculated calves. Histopathologic observation of injection sites revealed foci of caseous necrosis surrounded by accumulations of inflammatory cell infiltrates. The subcutaneous layer of the dermis was the limit of involvement. Abscesses were noted due to the fibrinohistiocytic reaction (Figure 4).

Discussion

Both the SQ and ITh models described will be used for future MB pathogenesis research. The MB 428E strain used here induced characteristic tenosynovitized arthritis lesions as well as pleuritis similar to what was observed in the field.[1,12] We are unable to produce the characteristic pulmonary bronchiectasis and abscessation as seen with increasing incidence in field cases. The pulmonary lesion in all ITh inoculated calves were limited to a moderate fibrinosuppurative pleuritis with focal areas of necrosis and these lesions were mostly limited to the areas near injection sites. Low dosages and short incubation times may be two explanations why these lesions were absent in the experimental animals of this study. We are confident that the pleuritis present was caused by the MB colonization. Although calf 101 had *P. haemolytica* cultured from homogenized lung samples, the appearance of the lungs and the extent of involvement was not different than the lungs of other inoculated calves. *P. haemolytica* is a common inhabitant of the nasal cavity of cattle.[8] The serotype of the isolate was not determined and it is not known if the isolate was virulent. Adegobye, et al. 1995, reported that of 45 fatal chronic pneumonia cases in calves, 56% stained specifically for MB and 52% of those did not have lung abscesses. This study seems to support that reporting. Although major lung involvement was not evident, MB was cultured from normal and involved lung areas.

Three of the five calves had clinical lameness and purulent arthritis involving the hocks. Of the two calves not exhibiting similar lesions, one calf (103) was a control inoculated intrathoracicly with sterile PBS and the other (101) was necropsied at five days post-inoculation. We speculate that the incubation period (five days) was not long enough for arthritis to develop in calf 101. Perhaps if this calf has been necopsied at day eight or later, an arthritis may have been evident.

The arthritic lesion was unexpected upon ITh inoculation. This model appears to be similar to intravenous inoculations due to the production of arthritis. The MB 428E gained access to the circulatory system to reach the joints, as MB was cultured from the blood of the three arthritic calves. Growth of the isolate in the pleural space may have allowed for expression of virulence factors that permitted the MB to escape phagocytocytosis in the reticuloendothelium system. It may be possible that a subpopulation of more virulent MB was selected for in the pleural space in inoculated calves. In addition, the cell population in the pleural space may also be less effective in clearing MB.

The lesion in the brisket of calf 104 is strikingly similar to field case reports.[12] It is not known why other calves were not similarly affected with decubital abcesses in the brisket or how the lesion arose in this area. No other lesions were noted in calves in addition to those described in the lungs, hocks, and brisket, although several other tissues cultured positive for MB. MB has been previously cultured from the lymph nodes, liver, kidney, spleen, and abomasum. The once the organism was introduced into the lung at inoculation. Blood was not aspirated during inoculation but

route of entry into the regions described in Table 5 are unknown but it is thought to be via the circulatory system

it is reasonable to conclude that blood vessels were penetrated with the trocar. MB has been known to gain access to the circulatory system and as previously stated, MB virulence factors may have assisted in the possible entrance to the circulatory system.[13]

This project was successful in that one animal model was demonstrated that contained several components of field cases - pleuritis, decubital abscesses, and multiple tissue MB isolation. As additional field cases become available more information regarding the epidemology of MB-associated diseases is evident. However the availability of an animal model mimicking field cases is useful for pathogenicity studies. Together all available information may elucidate the significance and mechanism of MB action in feedlot and dairy operations.

References

1. Adegboye DS, Halbur PG, Cavanaugh DL, Werdin RE, Chase CCL, Miskimins DW, Rosenbusch RF: Immunohistochemical and pathological study of *Mycoplasma bovis*-associated lung abscesses in calves. J Vet Diagn Invest **7:**333-337, 1995

2. Adegboye DS, Ulrike R, Halbur PG, Andrews JJ, Rosenbusch RF: Monoclonal antibody-based immunohistochemical technique for the detection of *Mycoplasma bovis* in formalin-fixed, paraffin-embedded calf lung tissues. J Vet Diagn Invest **7:** 261-265, 1995

3. Bennett RH, Jasper DE: Nasal prevalence of *Mycoplasma bovis* and titers in young dairy animals. Cornell Vet **67:** 361-373, 1977

4. Boughton E: *Mycoplasma bovis* mastitis. Vet Bull **49:** 337-387, 1979

5. Chima JC, Wilkie BN, Nielson KH, Ruhnke HL, Truscott RB, Maxie G, Chick B: Synovial immunoglobin and antibody in vaccinated and nonvaccinated calves challenged with *Mycoplasma bovis*. Can J Comp Med **45:** 92-96, 1981

6. Chima JC, Wilkie BN, Ruhnke HL, Truscott RB, Curtis RA: Immunoprophylaxis of experimental *Mycoplasma bovis* arthritis in calves. Protective efficacy of live organisms and formalized vaccines. Vet Microbiol **5:** 113-122, 1980

7. Doing PA, Ruhnke HL: Effects of mycoplasma and acholeplasma infection on bovine reproduction. *In*: Current therapy in theriogenology, ed. Marrow DA, pp. 228-291. WB Saunders Co., Philadelphia, PA, 1986

8. Gourlay RN, Houghton SB: Experimental pneumonia in conventionally reared and gnotobiotic calves by dual infection with *Mycoplasma bovis* and *Pasteurella haemolyticea*. Res Vet Sci **38:** 377-382, 1985

9. Gourlay RN, Thomas LH, Wyld SG: Increased severity of calf pneumonia associated with the appearance of *Mycoplasma bovis* in a rearing herd. Vet Rec **124:** 420-422, 1989

10. Hotzel H, Demuth B, Sachse K, et al: Detection of *Mycoplasma bovis* using in vitro deoxyribonucleic acid amplification. Rev Sci Tech Off Int Epizoot **12:** 581-591, 1993

11. Jasper DE, Al Aubaidi JM, Fabricant J: Epidemiologic observations on mycoplasma mastitis. Cornell Vet **64:** 407-415, 1974

12. Kinde H, Daft BM, Walker RL, Charlton BR, Petty R: *Mycoplasma bovis* associated with decubital abscesses in Holstein calves. J Vet Diagn Invest **5:** 194-197, 1993

13. Lauerman LH: Mycoplasmas of the bovine respiratory tract. *In:* Mycoplasmas in animals: laboratory diagnosis, ed. Whitford HW, Rosenbusch RF, Lauerman LH, pp. 50-56. Iowa State University Press, Ames, IA, 1994

14. Lopez A, Maxie MG, Ruhnke L, et al.: Cellular inflammatory response in lungs of calves exposed to bovine viral diarrhea virus, *Mycoplasma bovis*, and *Pasteurella haemolytica*. Am J Vet Res **47:** 1283-1286, 1986

15. McCully MA, Brook KV: Development of hybridization probe for the detection of *Mycoplasma bovis*. J Vet Diagn Invest **4:** 464-467, 1992

16. Ryan MJ, Wyand DS, Hill DL, Tourtellotte ME, Yung TJ: Morphologic changes following intraarticular inoculation of *Mycoplasma bovis* in calves. Vet Pathol **20:** 472-487, 1983

17. Stalheim OHV: Failure of antibiotic therapy in calves with mycoplasmal arthritis and pneumonia. J Am Vet Med Assoc **169:** 1096-1097, 1975

18. Sternberger LA, Hardy PH, Cuculis JJ, Meyer HG: The unlabled antibody-enzyme method of immunohistochemistry. J Histochem Cytochem 18: 315, 1970

19. Thomas LH, Howard CJ, Stott EJ, Parsons KR: *Mycoplasma bovis* infection in gnotobiotic calves and combined infection with respiratory syncytial virus. Vet Pathol **23:** 571-578, 1986

Table 1 Calves inoculated introthoracicly with M. bovis 428E.

Calf Number	Peak Temperature	Necropsy Day [+]	Clinical Signs [++]
101	103.0	5	D
102	103.6	8	D P L
104	104.8	8	D L
105	104.9	8	D L
103 (control)	102.1	5	-

[+] necropsy day post-inoculation

[++] D = depression, L = lameness P = panting

Table 2. *M. Bovis* isolation from blood and nasal swabs.

	day -2		day 1		day 2		day 3		day 4		day 5		day 6		day 7		day 8	
	blood	nasal	blood	nasal	blood	nasal	blood	nasal	blood	nasal	blood	nasal	blood	nasal	blood	nasal	blood	nasal
calf 101	-	-	+	-	-	-	-	-	-	-	+	-	ND	ND	ND	ND	ND	ND
calf 102	-	-	-	-	+	-	+	-	-	+	+	-	-	-	+	-	-	-
calf 104	-	-	-	-	-	-	+	-	+	-	+	+	+	+	+	+	+	+
calf 105	-	-	-	-	+	-	-	-	-	+	-	+	-	-	-	-	-	-
calf103 (control)	-	-	-	-	-	-	-	-	-	-	-	-	ND	ND	ND	ND	ND	ND

ND = not done

Table 3. Macroscopic scores and culture results from ITh inoculation.

	lung pool	TBLN	joint pool	organ pool	gut pool	Macroscopic score lung score % involvement	joint score
calf 101	+	+	+	+	-	17	none
calf 102	+	+	+	+	+	11	severe
calf 104	+	+	+	+	+	15	severe
calf 105	+	+	+	+	+	12	severe
calf 103	-	-	-	-	-	0	none

Table 4. Subcutaneous lesion measurements from Calf 202.

dose	site 1 size (cm)	site 2 size (cm)	site 3 size (cm)	site 4 size (cm)	culture results
4×10^8	3.5 x 5.5	4.5 x 4.5	5.5 x 5.5	3.5 x 4.5	+
1×10^8	3.0 x 4.0	1.5 x 3.0	5.5 x 3.0	2.5 x 2.0	+
PBS	none	none	none	none	-

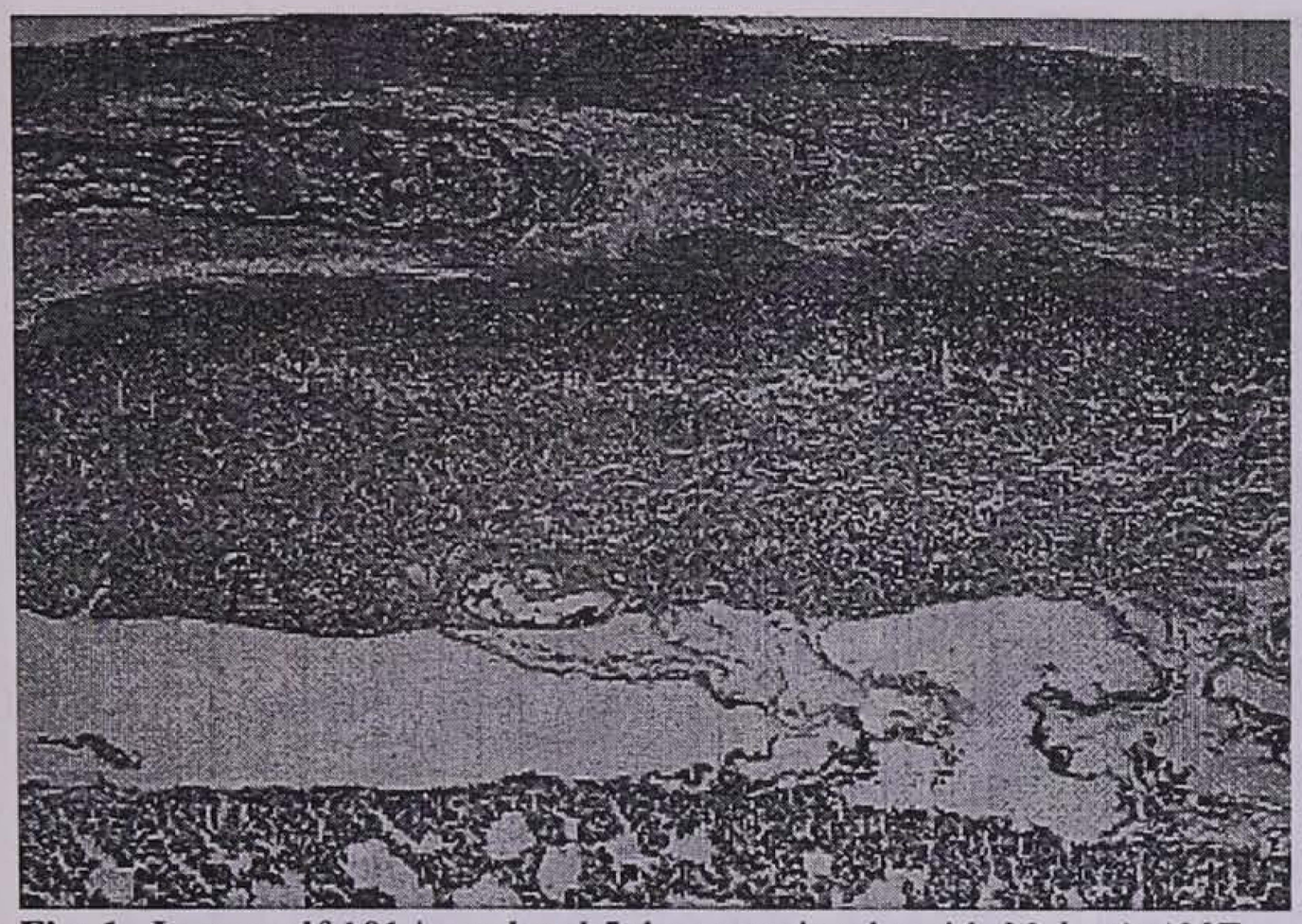


Fig. 1. Lung; calf 101 inoculated 5 days previously with *M. bovis* 428E. Severe fibrinosuppurative pleuritis near the site of MB inoculation. 10x.

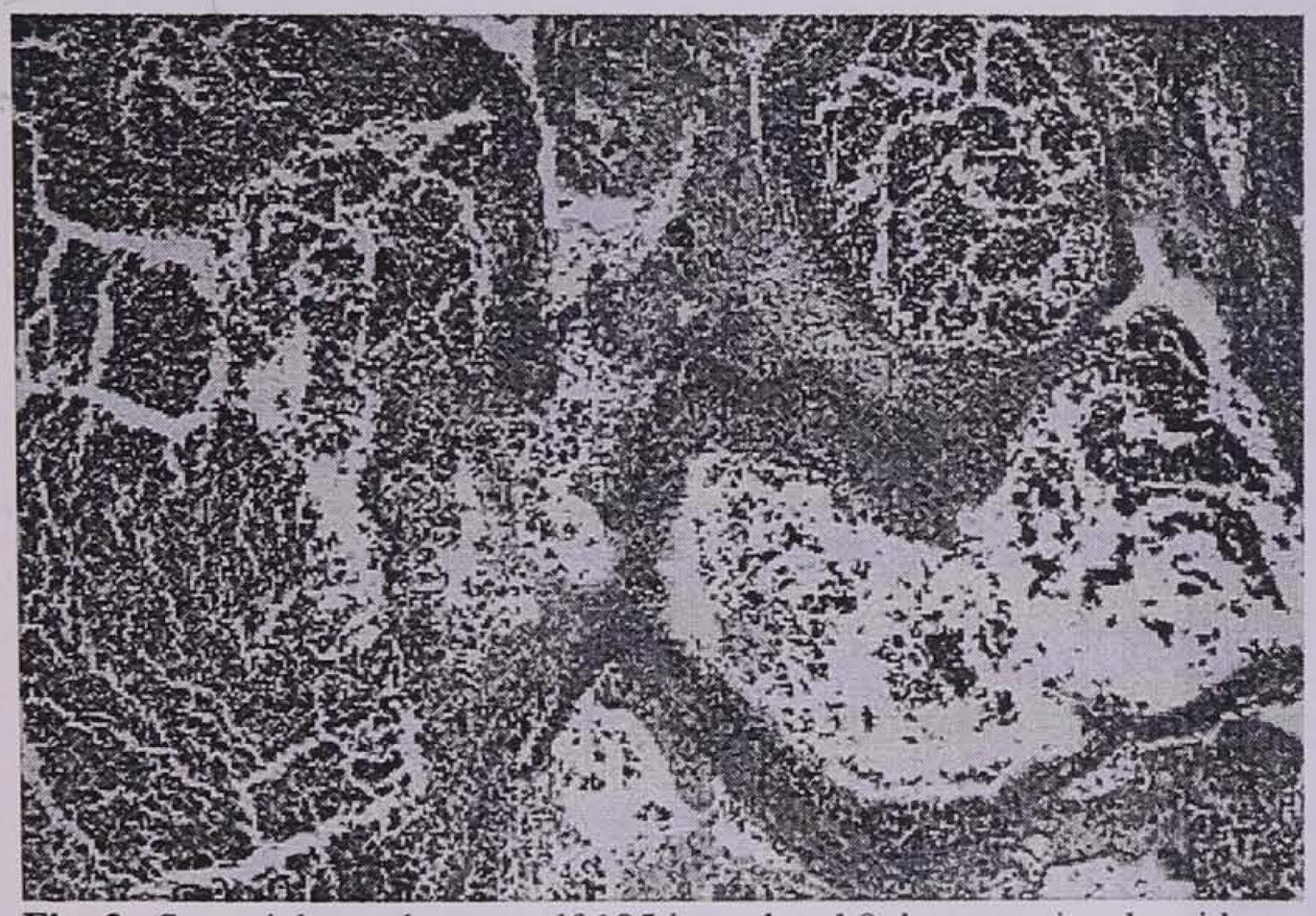


Fig. 2. Synovial membrane, calf 105 inoculated 8 days previously with *M. bovis* 428E. Formation of abscesses with caseous necrotic centers is evident. Note the accumulation of neutrophils, macrophages and plasma cells. 40x.

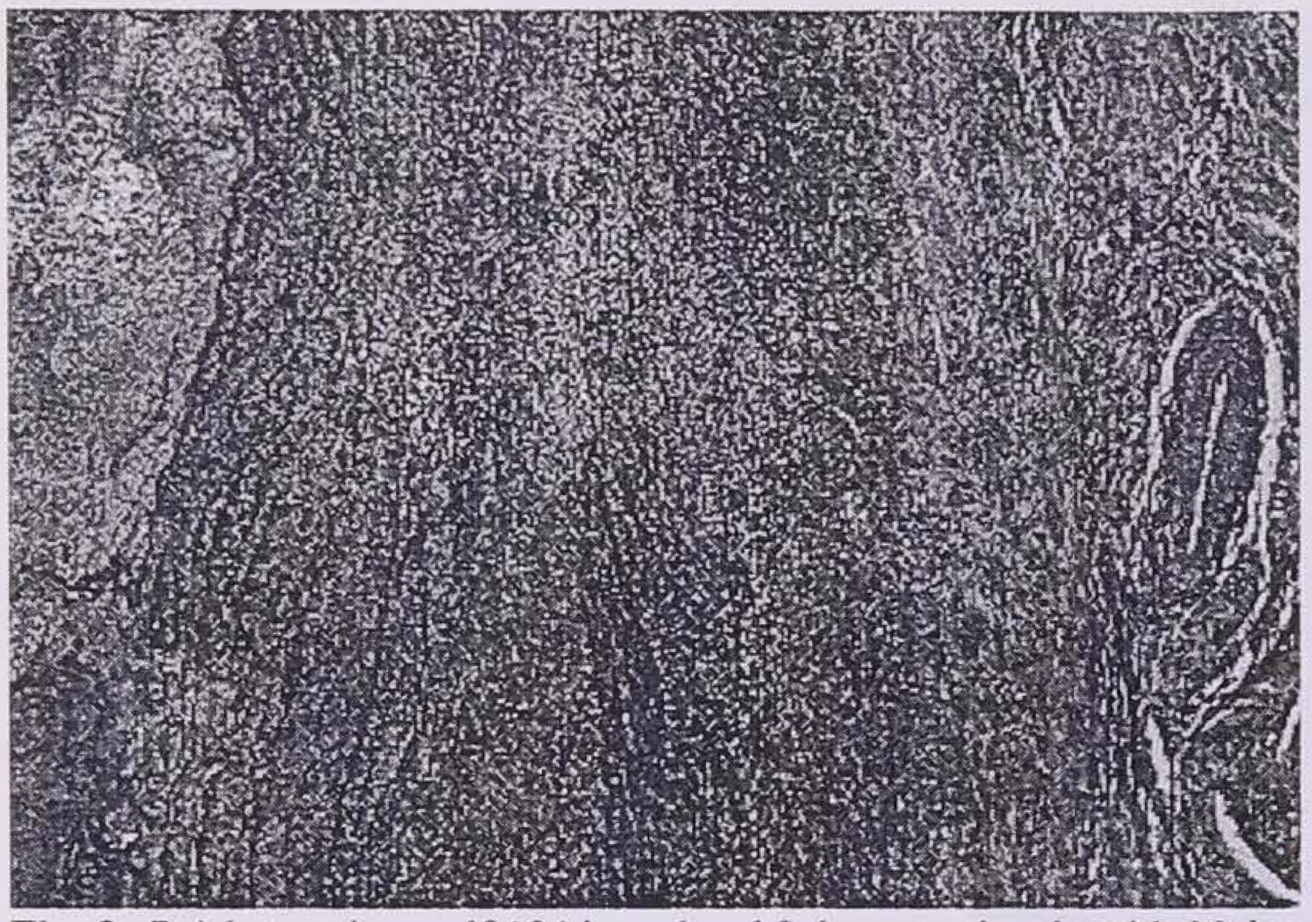


Fig. 3. Brisket region, calf 104 inoculated 8 days previously with *M. bovis* 428E. Muscle fibers are separated by mixed inflammatory cells. Foci of coagulative necrosis are evident. Fibrin accumulation is also noted. 10x.

Fig. 4. Calf 202 inoculated subcutaneously with 4 x 10^8 CFU/ml of *M. bovis* 428E 5 days previously. Enlargement of the subcutaneous layer of the dermis is due to fibrinosuppurative and histiocytic reaction. 10x.

Is Milkfat from All Cows Atherogenic?

G. L. Lindberg, assistant professor; and
G. Bobe, research assistant
Department of Animal Science
Iowa State University

DSL-144

Summary and Implications

The fatty acid composition of milkfat helps to determine nutritional, processing, and storage properties of milk products. We examined the fatty acid composition of milkfat from 233 Holstein cows to determine the range of fatty acid composition and to relate the composition of milkfat to the potential to cause atherosclerosis. We found that, even after seasonal effects and stage of lactation are calculated, milkfat from individual cows varies significantly in its percentages of the fatty acids myristic acid (C14:0, range = 3.3 to 13.7%), palmitic acid (C16:0, range = 22.9 to 36.2%), and oleic acid (C18:1, range = 19.7 to 44.3%). When the fatty acid compositions of milkfat from individual cows are used to calculate the atherosclerotic potential of milkfat, the values for milkfat averaged 2.25 and ranged from 0.77 to 3.97 in comparison with cocoa butter (13.6) and margarine (0.6). Dairy products that are made from milkfat at the extremes of the values observed here will vary considerably in their fatty acid composition and in their atherogenic potential.

Introduction

Americans currently consume 34 to 37% of food energy as fat, and dairy products account for 17% of the total fat in the American diet. Milkfat is frequently cited to increase blood cholesterol and LDL (low-density lipoprotein) cholesterol because it has relatively high concentrations of C14:0 and especially C16:0, relatively low concentrations of C18:2 and C18:3, and variable concentrations of oleic acid (C18:1). Foods such as milkfat contain mixtures of fatty acids and effects of mixtures of fatty acids on blood cholsterol are difficult to predict.

A formula for comparisons of the atherogenic potential of dietary fats has been devised. This formula is the Atherogenic Index (AI). The more atherogenic fatty acids are in the numerator of the formula and the less atherogenic fatty acids are in the denominator (Figure 1). According to this formula, cocoa butter has the highest AI, several vegetable oils have the lowest, and milkfat is relatively high among the several common fats.

The purpose of our study was to determine the range of fatty acid composition in milkfat of a herd of

Table 1. The major fatty acids in milk and their effects on human health.

Fatty acid	Abbrev.[a]	% in milkfat	Effects on human health
Butyric	C4	3.3	Possible colon cancer inhibitor
Hexanoic	C6	2.3	
Octanoic	C8	1.2	
Decanoic	C10:0	2.8	
Lauric	C12:0	3.4	Stimulates cholesterol synthesis
Myristic	C14:0	11.4	Stimulates cholesterol synthesis
Palmitic	C16:0	29.5	Raises blood cholesterol by decreasing liver removal of cholesterol
Palmitoleic	C16:1	3.4	Possibly cholesterol-lowering effect
Stearic	C18:0	9.8	Not absorbed efficiently and therefore influences cholesterol little
Oleic	C18:1	27.4	Possible cholesterol-lowering effect; abundant in olive oil
Linoleic	C18:2	2.8	Polyunsaturated; cholesterol-lowering effect
Linolenic	C18:3	0.8	Polyunsaturated; cholesterol-lowering effect

cattle maintained under constant nutritional and management conditions. A second purpose of our study was to determine the range of atherogenic index in the milkfat from individual dairy cows.

Materials and Methods

Milk was sampled 2 to 4 times from 233 Holstein cows in the ISU Ankeny Dairy Herd for a total of 800 samples. Fat, protein, and solids percentages were determined by infrared analysis. The fatty acid composition of milkfat was determined by extraction of fat with organic solvents, conversion of the fatty acids to ethyl esters, and separation of the fatty acid esters by gas chromatography. The atherogenic index of milkfat was calculated from the fatty acid composition of milkfat produced by each cow by using an atherogenic index formula (Figure 1).

Figure 1 Formula to calculate Atherogenic Index (AI).

$$IA = \frac{C12:0 + (4 \times C14:0) + C16:0}{C14:1 + C16:1 + C18:1 + C18:2 + C18:3 + \text{others}^a}$$

[a] others=C10:1, C12:1, C13:1, C15:1, C17:1, C20:1

Results and Discussion

The atherogenic index combines quantitative and qualitative information about the potential of the individual fatty acids in a fat source to cause atherogenesis. The numerically greater the atherogenic index, the greater is thought to be the atherogenic risk associated with the fat. Myristic acid (C14:0) is relatively atherogenic as revealed by its presence in the numerator of the AI equation and by its multiplier (Figure 1). The range of myristic acid concentrations in milk varied from 3.3 to 13.7%. The milk sample that contained 3.3% myristic acid had the highest concentration of oleic acid, 43.3%, whereas the milk sample with 13% myristic acid contained 19.9% oleic acid (Table 2). By applying the quantitative fatty acid data from multiple milk samples from 233 cows, an average AI score of 2.25 was observed (Tables 2 and 3). The lowest AI score of milkfat from any single cow was 0.77, which is similar in atherogenic index to margarine (Table 3). The AI scores from the highest and lowest cows are not unusual because the highest 5% and lowest 5% AI scores from the 233 cows differ greatly from one another (Table 3).

Approximately half of the milk fatty acids (C4 to C16:0) are made directly in the gland and the remaining fatty acids (C16:0 to C18:2) are transported from the blood to the milk. Synthesis of the intermediate chain fatty acids in the mammary gland (especially C12:0 to C16:0) seems to be influenced by the amount of longer chain fatty acids (C18) that are transferred into the gland from the blood. For example, when there is a greater abundance of the C18 fatty acids in milk, the mammary gland synthesizes less of the fatty acids of shorter and intermediate chain length. This reciprocal synthesis of fatty acids should increase the variability of fatty acids in milk.

The inverse relationship between C14:0 and C18:1 is not greatly surprising. In the bovine mammary gland, increased synthesis of longer chain fatty acids such as 18:0 and 18:1 causes decreased mammary synthesis of other fatty acids such as 12:0, 14:0, and a portion of 16:0. This tendency results in an inverse regulation between secretion of 18:1, which is less atherogenic, with secretion of 14:0 and 16:0, which are more atherogenic.

These results show that the milkfat produced by many cows has an atherogenic potential similar to that of margarine. Could this information be used to design novel milk products for people who are especially conscious about the fat composition of their diet? Alhough it would require separate handling of milk produced by individual cows, partitioning of milk of unique composition could allow milk processors to manufacture products of specialized composition for health-conscious consumers. Increased sales of milk products to new groups of consumers who are currently reluctant to consume milk products could be a result.

Acknowledgments

This research was supported in part by the USDA Center for Designing Foods to Improve Human Nutrition.

Table 2 Comparison of fatty acid contents of milkfat from 233 cows ranked according to AI index.

	Fatty Acid			
AI ranking	14:1	16:0	18:1	AI index
	---------- % by weight ----------			
Lowest cow	3.3	26.8	44.3	0.77
Median cow	11.3	28.6	26.3	2.29
Highest cow	13.6	36.2	19.9	3.97
Lowest 5% cows	5.2	25.3	37.8	1.06
Middle 5% cows	10.7	28.0	25.8	2.30
Highest 5% cows	12.6	36.2	20.3	3.31

Table 3 Comparisons of AI index of milkfat from selected cows with other fat sources.

Source	AI index
Chicken	0.5
Margarine	0.6
Lowest AI milkfat	0.77
Palm oil	0.9
Beef roast	1.0
Mean AI milkfat	2.25
Greatest AI milkfat	3.97
Cocoa butter	13.6

Feeding of Vitamin D_3 is a Potential Method to Improve Tenderness of Beef

D. Beitz, distinguished professor of agriculture; A. Trenkle, distinguished professor of animal science; F. Parrish, professor of animal science; J. Montgomery, research assistant, now at Texas Tech University; and R. Horst, research physiologist, U.S. Department of Agriculture, Agricultural Research Service, National Animal Disease Center, Metabolic Diseases and Immunology Research Unit, P.O. Box 70, Ames, Iowa 50010-0070

DSL-145

Summary and Implications

Feeding 5 million IUs of vitamin D_3 per day for 9 consecutive days increased tenderness of strip loin and top round steaks from beef cattle. Improved tenderness was determined by the Warner-Bratzler shear method and by Western blot analysis of a muscle protein degradation product. We are optimistic that a similar vitamin D_3 supplementation program could be used to improve the tenderness of beef derived from dairy animals.

Introduction

One of the major challenges of the beef industry is to provide a consistently tender meat product for consumers. Tenderness has been identified as the single most important factor affecting consumers' satisfaction and perception of taste. One way of improving beef tenderness is the injection of calcium chloride solution into postrigor and prerigor beef carcasses and cuts. This exogenous calcium chloride evidently activates both μ-calpain and m-calpain, which are intracellular calcium-dependent proteases and are responsible for tenderization. Supplemental dietary vitamin D_3 will increase blood calcium markedly via actions of additional 1,25-dihydroxyvitamin D that promotes absorption of dietary calcium. Vitamin D_3 also increases uptake and transport of calcium by the mitochondrial, sarcoplasmic, and plasma membranes of skeletal muscles. Because supplemental dietary vitamin D_3 causes increased calcium concentrations in blood and skeletal muscles, we hypothesized that supplemental dietary vitamin D_3 would increase calcium-activated protease activity in bovine muscles during postmortem storage and thereby cause improved beef tenderness.

Materials and Methods

Thirty crossbred steers approximately 23 months of age and predominantly of large-frame Continental X British breeds were allotted randomly to three treatment groups: a placebo, 5 million IUs of vitamin D_3, and 7.5 million IUs of vitamin D_3. All 30 steers were fed a high-concentrate finishing diet consisting of a dry basis of 78.2% whole shelled corn, 14.2% chopped alfalfa hay, 4.1% soybean meal, and 3.5% of a 40% crude protein liquid supplement. For the last 4 weeks, the steers were fed a high-concentrate diet consisting of 85% cracked corn and 15% oats along with 0.45 kg of commercial supplement (42% crude protein, 5% calcium, 1.1% phosphorus, 1.6% salt, and added vitamins A, D, and E) per head per day. The supplement provided 225 mg of monensin per head per day. The steers had free access to water and large bales of hay consisting of predominantly alfalfa with some mixed grasses. The steers were grouped together in one pen. Also, starting 10 days before slaughter, steers were given intraruminally a bolus containing ground corn or ground corn containing the 5 or 7.5 million IUs of vitamin D_3 in gelatin capsules before the morning feeding for 9 consecutive days. On the morning of day 10, the steers were transported to a commercial beef packing plant and slaughtered that afternoon.

Three days after slaughter, carcasses were transported to a beef breaking plant. *Longissimus lumborum* (strip loins) and semimembranous muscle (top round) were placed in anaerobic vacuum bags and transported to the Iowa State University Meats Laboratory. For Warner-Bratzler shear evaluation, strip loin and top round steaks were cut 2.54 cm thick, placed in plastic bags, and wet-aged at 1°C. Both types of steaks were aged 7, 14, and 21 days; striploin steaks also were aged for 3 days. After aging, steaks were frozen at -20°C until subsequent analysis.

Steaks were thawed slowly at 2°C for 24 hours and then broiled until they reached an internal temperature of 71°C. After cooling to 25°C, six 1.27-cm-diameter cores were removed parallel to the muscle fiber direction of each of the steaks. Cores were sheared perpendicular to the fiber direction through the center of the core by using a Warner-Bratzler Shear head attached to an Instron Universal Testing Device (model 4502) controlled with a Model 4500 computer-assisted module (Instron, Canton, MA). Peak shear force values were recorded as kilograms per 1.3-cm-diameter core. The six shear values per steak were averaged, and the means for treatments were analyzed for statistical significance.

Whole muscle preparations for SDS-PAGE were performed on strip loin steaks postmortem aged for 14 days

to detect amounts of the 30 kDa component, a proteolytic degradation product from troponin T.

Data were analyzed as a completely randomized design with individual steers serving as the experimental unit.

Results and Discussion

As shown in Table 1, steaks from steers orally administered vitamin D_3 preceding slaughter had numerically lower Warner-Bratzler shear values. When the Warner-Bratzler shear force means for the four postmortem aging times were averaged, the decrease in shear force for both strip loin and top round steak that was caused by both dosages of vitamin D_3 was significant ($P > 0.0001$ and $P > 0.0003$, respectively). Although shear force data suggest that supplemental vitamin D_3 improved steak tenderness at all postmortem aging times, the maximal improvement was noted for those steaks postmortem aged for 14 days. Moreover, the 5 million IUs dose per day was equally effective to the 7.5 million IU dose per day in causing improvement in beef steak tenderness.

Degradation of troponin T to a 30-kDa component is related to improved tenderness. Strip loin steaks postmortem aged 14 days from the 5 million IU-treated cattle had significantly ($P = 0.03$) more proteolytic degradation as evidenced by higher content of the 30-kDa component observed by diffusing Western blots than did steaks from the control group (Table 2). Proteolytic degradation products of steaks from the 7.5 million IU-treated cattle tended to be different ($P = 0.052$) from the 5 million IU-treated group but were not significantly different from controls ($P > 0.05$).

Based on this and previous studies, postmortem increase in tenderness is most likely the result of the degradation of myofibrillar proteins responsible for the integrity of the myofibril. Degradation of troponin T and the simultaneous appearance of polypeptides migrating at about the 30-kDa region are correlated strongly to beef tenderness. A protease called m-calpain degrades purified bovine troponin T to produce polypeptides in the 30-kDa region. This result of the degradation of troponin T to a 30-kDa component recently has been confirmed by using Western blotting techniques. Because calpains are considered the major system in postmortem proteolysis, and by our detection of an increase in the 30-kDa component demonstrated that proteolysis is greater in muscles of cattle fed supplemental doses of vitamin D_3.

Feeding supplemental daily doses of 5 or 7.5 million IUs of vitamin D_3 to feedlot cattle improves tenderness of beef strip loin and top round steaks. Therefore, feeding vitamin D_3 offers a simple, low-cost, short-term, and effective way to improve tenderness within 14 days of postmortem aging of muscles.

Feeding 5 million IUs of vitamin D_3 per day for 9 consecutive days before slaughter could be implemented easily in a commercial feedlot system and offer a cost effective way of producing more tender strip loin and top round steaks that are within 14 days postmortem. Feeding supplemental vitamin D_3 also could be used in a value-based marketing system that is based on improved tenderness. Therefore, antemortem feeding of supplemental vitamin D_3 is an effective, easy, and inexpensive way to improve beef tenderness before slaughter and potentially increase consumer acceptance of beef.

Acknowledgments

The research was supported partly by the Iowa Beef Industry Council

Table 1. Effect of supplemental dietary vitamin D_3 on Warner-Bratzler shear force of strip loin and top round steaks at different postmortem aging times.

Steak/ Aging	Vitamin D3 Treatments: Control Shear force, kg	SE	5 million IUs/day Shear force, kg	SE	7.5 million IUs/day Shear force, kg	SE	P>F
Strip loin steak							
Aging time (day)							
3	3.58	.17	3.11	.18	3.17	.15	.1638
7	3.32	.09	3.20	.19	2.89	.16	.1873
14	3.25[a]	.09	2.80[b]	.08	2.78[b]	.14	.0015
21	3.38	.11	2.90	.17	3.02	.13	.1071
Mean	3.38[a]	.12	3.00[b]	.16	2.97[b]	.15	.0001
Top round steak							
Aging time (day)							
7	3.97	.23	3.56	.20	3.32	.17	.0685
14	3.91[a]	.15	3.37[b]	.14	3.37[b]	.15	.0366
21	3.74	.10	3.32	.17	3.56	.16	.1973
Mean	3.87[a]	.16	3.42[b]	.17	3.42[b]	.16	.0003

[a,b]Means (n=10) in the same row with a common superscript or no superscript are similar (P>.05).

Table 2. Effect of 5 million IUs of vitamin D_3 administered daily for 9 days to steers on amount of the 30-kDa component in 14-day postmortem aged steaks.

	Vitamin D_3 Treatments					
Steaks	Control		5 million IUs/day		7.5 million IUs/day	
	Mean[a]	SE	Mean[a]	SE	Mean[a]	SE
Strip loin steaks	.614[b]	.09	.889[c]	.09	.631[b]	.09
Top round steaks	.965	.16	1.207.16	1.029	.17	

[a]Means (n=10) represent relative values of amount of the 30-kDa band in Western blot analyses.
[b,c]Means in the same row with a different superscript letter differ (P<.05).

We wish to express appreciation to the following agencies or companies for their support of the dairy program at Iowa State University:

American Cyanamid
American Jersey Cattle Club
AMPI
Anderson Erickson Dairy Co.
Avecor Biomedical, Inc.
Beatrice Cheese, Inc.
Binational Agricultural Research and Development
Biotechnology Research and Development Corporation
Center for Crop Utilization Research
Center for Designing Foods to Improve Nutrition
Church and Dwight, Co.
Consortium for Plant Biotechnology Research
Dairy Lab Services
DDX, Inc.
DEC—Boumatic
DeKalb Plant Genetics
Eastern Artificial Breeders Cooperative
Eli Lilly
Foremost Farms
Fort Dodge Laboratories
Holstein Association
Huprich, Inc.
Iowa Agriculture and Home Economics Experiment Station
Iowa Ayrshire Breeders Association
Iowa Beef Industry Council
Iowa Brown Swiss Association
Iowa Corn Promotion Board
Iowa Dairy Products Association
Iowa Department of Agriculture and Land Stewardship
Iowa Farm Bureau
Iowa Guernsey Cattle Club
Iowa Holstein Association
Iowa Jersey Cattle Club
Iowa Milking Shorthorn Association
Iowa Pork Producers Association
Iowa Purebred Dairy Cattle Council
Iowa Sheep and Wool Promotion Board
ISU College of Veterinary Medicine
ISU Department of Animal Science
Lederle Laboratories
Leopold Center for Sustainable Agriculture
Lyon Jersey
Mid-America Dairymen, Inc.
Mid-States Dairy Record Processing Center
Midland Dairy Association
Milk Specialities
Monsanto
National Animal Disease Center
National Association of Animal Breeders
National Dairy Promotion and Research Board
Northern Ohio Breeders, Inc.
Pharmacia?Upjohn Corp.
Practical Farmers of Iowa
RJB, Inc.
Select Sires
Swiss Valley Farms
Synergen, Inc.
Trans-Ova Genetics
21st Century Genetics
U.S. Department of Agriculture
Wapsie Valley Creamery
Wells Blue Bunny
West Agro, Inc.

This report was coordinated by Leo Timms, Extension dairy specialist and Elaine Edwards, Extension communications specialist. Edited by Julie Todd, Extension program specialist. Front and back covers by Dean Biechler.

DyS-3357 | December 1997

. . . and justice for all

The Iowa Cooperative Extension Service's programs and policies are consistent with pertinent federal and state laws and regulations on nondiscrimination. Many materials can be made available in alternative formats for ADA clients.

Issued in furtherance of Cooperative Extension work, Acts of May 8 and June 30, 1914, in cooperation with the U.S. Department of Agriculture. Stanley R. Johnson, director, Cooperative Extension Service, Iowa State University of Science and Technology, Ames, Iowa.

File: Animal Science 5