# **Effects of Heat Stress on Reproduction**

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### **ABSTRACT**

When dairy cattle are subjected to heat stress, reproductive efficiency declines. Cows under heat stress have reduced duration and intensity of estrus, altered follicular development, and impaired embryonic development. However, the extent of reduction in reproductive efficiency during summer in the United States or elsewhere is difficult to assess unless reproductive measures are calculated monthly based on results of actual inseminations rather than as rolling annual averages. Common methods to ameliorate effects of heat stress have been to provide cooling in the form of shades, soakers, fans. or evaporative coolers. Because negative effects of heat stress have been identified from 42 d before to 40 d after insemination, continual cooling must be provided. Tools for synchronization of estrus have been developed that greatly reduce the need for detection of estrus. Pregnancy rates were more consistent over season when timed artificial insemination programs were used compared with artificial insemination after detected estrus, although negative impacts of heat stress were still observed. Recently, calving in summer has been reported to reduce the success of a timed first insemination between 60 and 66 d postpartum, although some researchers have found calving in the summer to have a positive impact on days open. Other techniques that have been investigated to reduce the negative impact of heat stress on reproduction include embryo transfer, induction of accessory corpus lutea, and crossbreeding. Each technique has potential advantages but not without limitations or costs. Comparative economic evaluations of various combinations of strategies to attenuate negative effects of heat stress on both milk production and reproduction are needed.

(**Key words:** heat stress, reproduction)

**Abbreviation key:** CL = corpus luteum; CR = conception rate (percent pregnant of cows inseminated); EDR = detection of estrus rate; ME = mature equivalent; PR = pregnancy rate (percent pregnant of eligible cows in

a group); TAI = timed artificial insemination; <math>THI = temperature humidity index.

### INTRODUCTION

Summer heat stress has long been recognized as reducing both the productivity and reproductive efficiency of dairy cattle in the Southeastern and Southwestern portions of the United States. The impacts of heat stress on reproductive efficiency have been well documented (Thatcher, 1974; Fuquay, 1981) and reviewed (Gwazdauskas, 1985; Hansen and Aréchiga, 1999; Wolfenson et al., 2000; Rutledge, 2001; Hansen et al., 2001). Heat stress has been shown to alter the duration of estrus (Gangwar et al., 1965), colostrum quality (Nardone et al., 1997), conception rate (CR; Ingraham et al., 1976), uterine function (Collier et al., 1982), endocrine status (Collier et al., 1982; Wolfenson et al., 1988b; Wise et al., 1988; Howell et al., 1994), follicular growth and development (Wilson et al., 1998), luteolytic mechanisms (Wilson et al., 1998), early embryonic development (Biggers et al., 1987), and fetal growth (Wolfenson et al., 1988a). Although season alters endocrine profiles and fertility in males (Gwazdauskas, 1985), this discussion will focus on the female because most of the impacts on male fertility can be circumvented with AI.

Climatic factors that may influence the degree of heat stress include: temperature, humidity, radiation, and wind (Gwazdauskas, 1985). In an Israeli study, evaluating the relationship between ambient temperature and body temperature, the upper critical temperature for heat stress to begin was between 25 and 26°C (Berman et al., 1985). Conception rate declined from 61 to 45% when rectal temperature 12 h postbreeding increased 1°C (Ulberg and Burfening, 1967). Furthermore, cattle with rectal temperatures of 40°C as a result of exposure to 32.2°C ambient temperatures for 72 h after insemination had conception rates of 0% compared with a conception rate of 48% when rectal temperature was 38.5°C for cows in an ambient temperature of 21.1°C (Ulberg and Burfening, 1967). Although changes in rectal temperature are the ultimate indicator of heat stress in cattle, such data are not always available. Therefore, heat stress is frequently defined as occurring whenever the temperature humidity index (THI) exceeds 72

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Table 1. Summary data of DHI records (mean ± SD) from Dairy Metrics (Dairy Records Management Systems; Raleigh, NC, July, 2002).

Variable	All	$Region^1$			
		East	Midwest	South	West
No. of Herds <sup>2</sup>	11,492	5626	4397	1287	177
Avg No. of Cows	$136 \pm 226$	$112~\pm~149$	$121~\pm~216$	$254 \pm 349$	$437 \pm 597$
Proj. calving interval	$14.7~\pm~1.4$	$14.2  \pm  1.1$	$15.1 \pm 1.6$	$15.1 \pm 1.5$	$15.8 \pm 1.6$
Act. calving interval	$14.1  \pm  1.2$	$13.8 \pm 1$	$14.2 \pm 1.3$	$14.4 \pm 1.3$	$15 \pm 1.6$
Voluntary waiting period	$56 \pm 6$	$58 \pm 5$	$54 \pm 7$	$57 \pm 6$	$54 \pm 8$
Avg. days to first service	$99 \pm 25$	$94 \pm 21$	$103  \pm  28$	$103 \pm 28$	$93 \pm 30$
Conception rate – first service <sup>3</sup>	$45 \pm 21$	$44~\pm~17$	$45 \pm 24$	$48  \pm  25$	$40 \pm 26$
Conception rate – second service <sup>3</sup>	$43 \pm 21$	$44 \pm 18$	$43 \pm 24$	$43  \pm  25$	$35 \pm 23$
Conception rate – third + service <sup>3</sup>	$40~\pm~21$	$42 \pm 18$	$39 \pm 24$	$38 \pm 22$	$34 \pm 21$
Percentage of heats observed	$39 \pm 15$	$43 \pm 14$	$35 \pm 15$	$34 \pm 17$	$36 \pm 18$
Percentage of herd bred to proven AI bulls	$54 \pm 34$	$59 \pm 32$	$48  \pm  36$	$47 \pm 36$	$58 \pm 35$
Percentage of herd bred to young sires	$15 \pm 20$	$18 \pm 21$	$13 \pm 18$	$9 \pm 15$	$4 \pm 10$
Percentage of herd bred to non-AI Bulls	$26~\pm~34$	$21~\pm~30$	$30 \pm 36$	$38 \pm 39$	$33 \pm 35$

<sup>&</sup>lt;sup>1</sup>States are allocated to regions as follows: East—ME, VT, NH, MA, RI, CT, NY, PA, NJ, MD, WV, VA; Midwest—ND, SD, NE, KS, MN, IA, MO, WI, IL, MI, IN, OH, KY; South—OK, TX, AR, LA, TN, MS, AL, NC, SC, GA, FL; and West—WA, OR, ID, MT, WY, CA, NV, UT, CO, AZ, NM.

(Armstrong, 1994). Ravagnolo et al. (2000) reported that THI, based on the maximum temperature and minimum humidity, most effectively accounted for the effect of heat stress on production.

### **CURRENT SITUATION IN THE SOUTHERN US**

With these documented differences in reproductive performance, one would expect to see differences in Southern regional reproductive measures recorded by DHI associations. Key variables to evaluate reproduction, based on herd summary reports, were summarized by region in July 2002 using DairyMetrics (Dairy Records Management Systems, Raleigh, NC; Table 1). When those measures are examined, not a great deal of difference is found between regions. Actually, the Southern region has the highest numerical first service CR. However, such unedited data should be interpreted cautiously because the Southern region also has the highest percentage of cows bred to non-AI bulls. In many herds where natural service is used, the only breeding information reported is the estimated breeding date of the successful service. The third and greater service CR of 38% in the Southern region (Table 1) is similar to the 32% CR estimated for Holstein herds from 1997 to 1999 (Washburn et al., 2002), a study in which most herds with extensive use of natural service had been removed from the dataset. Data for herd average conception rates as currently reported through DHI are higher than actual values and, if not consistently higher across regions, then interregional comparisons are biased.

Also, current yearly summaries of DHI data do not allow adequate evaluation of seasonal changes in either CR or detection of estrus rate (**EDR**, the percentage of estruses observed relative to expected in a 21-d period). At a minimum, monthly summaries of EDR and CR should be available so that duration and magnitude of the negative impact of heat stress can be quantified. Furthermore, to fully assess the impact of heat stress on reproduction in the Southern United States or elsewhere, the type of cooling and its effectiveness in individual herds would be required, as well as a measure of the duration and intensity of heat stress. Monthly summaries would also be beneficial to producers in the Northern United States or elsewhere in evaluating the potential returns on cooling system investments.

Washburn et al. (2002) have reported that the average days open and services per conception increased in both Holstein and Jersey herds in the 10 Southeastern states from 1976 to 1999; concurrently with milk, fat, FCM, and herd size. In this study, time and the breed × time interaction affected days open, whereas services per conception was affected by time, FCM (linear), and herd size (linear). In the period from 1985 to 1999, EDR declined also. Although not specifically attempting to define the effect of heat stress on reproduction, Washburn et al. (2002) reported that reproductive measures generally were more unfavorable for states located further south; however, the changes across years were similar in all subregions. They hypothesized that as production levels increased, the negative effects of heat stress became more acute and the residual effects continued longer. In addition to the effect of heat stress

<sup>&</sup>lt;sup>2</sup>Total number of herds processed by region; the number of herds represented for other variables may differ.

<sup>&</sup>lt;sup>3</sup>Conception rates are not adjusted for use of natural service and may include some assumed pregnancies based on nonreturn rates, therefore reported values are likely higher than actual. Differential use of natural service could also be a confounding factor across regions.

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on reproduction, Washburn et al. (2002) proposed that as a cow's genetic merit for milk production increases, fertility may be negatively affected due to either the negative correlation with cow fertility, alterations in physiological processes (hormone production or metabolism), or inbreeding.

One of the potential reasons for seeing an increase in average days open and services per conception in herds subjected to heat stress is increased milk production potential, which was reported for Southern herds by Washburn et al. (2002). Al-Katanani et al. (1999) reported that the depression in fertility caused by heat stress was exacerbated by increasing mature equivalent (**ME**) milk yield. In their analysis of 90-d nonreturn rates, cows were grouped based on level of ME milk yield. They reported that cows having their first service in July had a 44.9% nonreturn rate if they produced <4536 kg of milk, but the nonreturn rate declined to 13.5% for those producing 4536 to 9072 kg and to 5.3% for those cows producing >9072 kg of milk.

Al-Katanani et al. (1999) also demonstrated the increasing influence of heat stress on cows based on their location. Least squares means for 90-d nonreturn rates were  $18.9\pm7.2$ ,  $7.0\pm2.8$ , or  $4.7\pm3.7\%$ , for cows located in south Georgia, north Florida, or south Florida, respectively. The number of months that 90-d nonreturn rates were  $\leq 20\%$  increased from 2 mo in south Georgia to 7 mo in south Florida. Al-Katanani et al. (1999) reported 90-d nonreturn rates were reduced when average temperatures exceeded  $20^{\circ}$ C on d -10, 0, or 10 relative to insemination.

The impact of season of calving on subsequent reproduction has been evaluated in several studies with variable results. Ray et al. (1992) reported that cows calving in the spring (March to May) or summer (June to August) in Arizona had the longest calving intervals and required the most services per conception. However, the data set was edited to include only cows with a calving interval between 300 and 600 d; therefore, cows conceiving very early postpartum or extreme problem breeders were excluded. No indication was made as to whether these outliers were proportional in all seasons, nor were measures of actual reproductive successes or failures culled before subsequent calving included. In a study evaluating bST, Silvia et al. (2002) reported that cows calving in the summer (July to September) in Kentucky actually had the fewest days open, while cows calving in the spring (April to June) had the greatest number of days to first service and those calving in the winter (January to March) had the fewest days to first service. This dataset may also be skewed in that cows with reproductive problems postpartum that had not fully recovered by 60-d postpartum, as well as thin cows, were excluded; nor did they indicate how they accounted for cull animals.

Average temperatures exceeded 20°C from April through October in Arizona (Ray et al., 1992), but were not reported by Silvia et al. (2002). Thus the differing results regarding the influence of season of calving on subsequent reproductive efficiency between the Arizona and Kentucky studies may be due to any of several factors: differences in months chosen for spring and summer; duration and intensity of heat stress in Arizona; differences in milk yield between locations; differences in culling criteria; differences in summer cooling techniques used in the regions; or an anomaly of the way datasets were edited.

Cartmill et al. (2001) reported that whenever the THI was ≥72, fewer cows were detected in estrus and CR were lower. Similarly, Ingraham et al. (1976) reported that CR declined from 66 to 35% when the THI increased from 68 to 78 the second day before breeding. When Jordan et al. (2002) evaluated season of calving and its influence on CR at a timed AI (TAI) first service in a herd with a 60-d voluntary waiting period, cows calving in the summer had the lowest first-service CR. This reduction in fertility may subsequently result in more days open than cows calving in other seasons. Cows at first service may be influenced by their season of calving; however, after the first or second insemination most summer calving cows, which have not conceived, would be under more moderate climatic conditions although residual effects of heat stress could be expected. This is supported by the work of Moore et al. (1992), who reported that days open was higher for cows calving in Mississippi in July than for those calving in August or September.

Research has provided insight into potential means of mitigating the effects of heat stress on dairy cows. These methods include cooling, using TAI programs, incorporating embryo transfer, inducing an accessory corpus luteum (CL), and crossbreeding cattle.

# **COOLING**

Providing cows with supplemental shade and cooling to mitigate summer heat stress has typically been evaluated economically using increases in milk production as the endpoint (Armstrong, 1994). Bucklin et al. (1991) summarized the results of adding sprinkler and fan cooling from Florida, Kentucky, Missouri, and Israeli locations; where measured, feed intake increased (7.1 to 9.2%), milk production increased (7.1 to 15.8%), rectal temperature decreased (-0.4 to -0.5°C), and respiration rates decreased (17.6 to 40.6%); however, reproductive measures and their benefits were not reported. Roman-Ponce et al. (1977) reported that cows provided shade

had lower respiration rates (54 vs. 82 breaths/min), reduced rectal temperatures (38.9 vs. 39.4°C) and improved CR (44.4 vs. 25.3%) compared with cows with no shade. Although milk production was increased from 15 to 16.6 kg/d by adding shade, those production levels are well below those expected today.

In a Texas field survey, Thompson et al. (1996) found that providing shade in the lounging area, holding pen, or dry cow areas and fans in the lounging area improved chances for summer pregnancy; whereas fans in the dry cow area decreased the chances of summer pregnancy. Sprinklers did not, however, influence the chance of pregnancy in the Texas situation. Whether this was the result of how the sprinklers were managed or was the result of reduced effect of evaporative cooling under the humid conditions in portions of the state was not determined.

The influence of dry cow cooling on subsequent fertility is an area that warrants further investigation. When Collier et al. (1982) reported effects of shade immediately prepartum, they found no statistical differences (shade vs. no shade) in days to first progesterone rise to >1 ng/ml (13 vs. 12 d), days to first observed estrus (28 vs. 36 d), days open (114 vs. 92 d), nor services per conception (2.6 vs. 2.4); although those variables, except for days to first observed estrus, were numerically higher (i.e., less favorable) for cows provided shade than for cows with no shade before calving.

Flamenbaum et al. (1995) evaluated the effect of post-partum cooling on cows of different BCS (six-point scale, 0 to 5) prepartum. Cows, which were cooled, consumed 1.6 kg more DM/d. Cooled cows with low BCS (2.65) lost 0.5 units of condition during the first 4 wk postpartum, compared with cows with high BCS (3.80) that lost 1 unit. Cooled cows reached higher levels of peak milk production, and cows with low BCS attained peak milk production earlier than cows with high BCS. Uncooled cows with low BCS had the lowest milk and fat production. In that experiment, effects of cooling and body condition were not additive and higher energy reserves at calving did not substitute for effects of subsequent cooling.

Attempts have been made to identify whether cooling at critical times during follicular and embryonic development could enhance fertility. Al-Katanani et al. (2002b) compared oocyte competence of nonlactating Holstein cows in the winter and summer. During summer, cows were housed in a heat stress (shades only) or cooled environment (free-stall barn with fans and foggers) for 42 d before slaughter and subsequent oocyte recovery. Although rectal temperatures were lower in the cooled cows (39.2°C), they were still higher than for cows during winter (38.7°C). Despite being cooled for 42 d before oocyte collection, there was no difference in

the proportion of oocytes that developed to blastocysts by d 8 of culture between cooled and uncooled cows in summer; however, a lower proportion developed to blastocysts during summer than during winter.

When Her et al. (1988) cooled cows nine times per day with a combination of sprinkling and forced ventilation from 1 d before a synchronized estrus to 8 d post-AI, more cooled cows exhibited standing behavior during estrus and fewer were classified as anestrus than noncooled cows. However, no improvement in CR was reported after the short duration cooling, despite a reduction of 0.5 to 0.9°C in the typical diurnal rise in body temperature. In a Florida trial, Ealy et al. (1994) provided cooling to cows from 2 to 3 d before, until 5 to 6 d after AI and reported a CR of 16.0% for cows cooled using sprinklers, fans, and shades, compared with 6.2% for cows with access to shade only. Ealy et al. (1993) evaluated embryos from superovulated cows that had been subjected to heat stress on 1, 3, 5, or 7 d after breeding and found embryos collected from the cows exposed to heat stress on d 1 of pregnancy had decreased viability and development compared to control cows. Thus, bovine embryos become more resistant to heat stress as they develop from single cells through morula and blastocyst stages. Putney et al. (1988a) have shown that d-17 conceptuses incubated at high temperatures synthesize heat shock proteins, which may enhance their resistance to heat stress.

Other researchers have reported that summer season or heat stress during oocyte maturation and near ovulation (Thatcher, 1974; Ingraham et al., 1976; Putney et al., 1989b; Al-Katanani et al., 1999; Cartmill et al., 2001; Al-Katanani et al., 2002b) and from 8 to 16 d (Biggers et al., 1987) or 27 to 40 d (Cartmill et al., 2001) of pregnancy have deleterious effects on embryonic development or CR. Putney et al. (1988a) showed that in vitro endometrial tissue subjected to elevated temperatures increased release of prostaglandins into culture medium. If such an effect occurred in vivo, it might initiate premature luteal regression or compromise the function of the CL. In addition, hyperthermia on d 17 of pregnancy increased uterine production of prostaglandin  $F2\alpha$  in response to oxytocin (Wolfenson et al., 1993). Thus, it appears that cooling is needed from at least 42 d before ovulation (Al-Katanani et al., 2002b) to over 40 d postinsemination (Cartmill et al., 2001) if negative impacts of heat stress are to be mitigated. Current methods of cooling may improve reproductive efficiency, but most do not maintain THI below 72 or body temperatures <39°C; therefore some compromise in fertility should be expected. With the development of implantable electronic temperature recording devices, increased accuracy in determining the magnitude and duration of elevated body temperature will allow imE108 JORDAN

proved assessment of when heat stress occurs and allow for more effective evaluation of abatement methods.

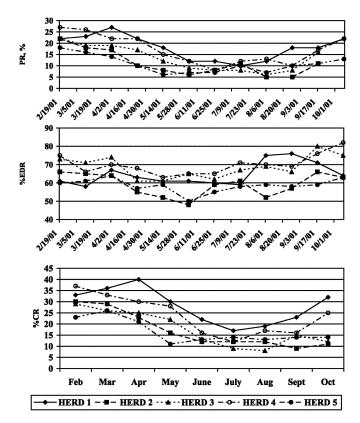
# **USE OF TAI PROGRAMS**

Recently, TAI programs based on follicular recruitment to synchronize ovulation have been developed (Pursley et al., 1995). In experiments conducted during summer heat stress, cows inseminated using a TAI program had a higher percentage of cows pregnant by 90 d (Aréchiga et al., 1998) or 120 d (de la Sota et al., 1998) postpartum than cows inseminated after first observed estrus, although CR at first service did not differ (Aréchiga et al., 1998). The beneficial effects of the first service TAI during the summer were maintained over the course of a year with fewer cows being culled (12.9 vs. 22.0%) and more cows eventually conceiving (87.0 vs. 77.9%) if TAI was used for first service compared with cows first inseminated at detected estrus (de la Sota et al., 1998). They estimated the increase in net revenue from implementing a first-service TAI program in the summer to equal \$118 per cow. Although TAI programs have been shown to improve pregnancy rate (PR) of cows suffering heat stress, increased subsequent embryonic losses have sometimes occurred. For example, Cartmill et al. (2001) showed that submission rates and PR at d 27 to 30 were enhanced when a TAI program was used; however, the advantage in PR was lost by d 40 to 50 due to increased embryonic mortality in cows bred using TAI.

Follicular dynamics of the heat stressed cow are altered when compared to control cows (Wolfenson et al., 1995). Although the average size of the first wave dominant follicle was similar between heat stressed and control cows, it decreased in size sooner, and the second wave dominant follicle emerged earlier. The second wave dominant follicle in heat stressed cattle may then be an aged follicle at ovulation; thereby reducing fertility. This may partially explain why the enhanced PR observed by Cartmill et al. (2001) 27 to 30 d postinsemination was not maintained to the subsequent pregnancy diagnosis.

When Jordan et al. (2002) evaluated two different first-service TAI regimens over the course of 11 mo, the season of first insemination was not significant. Burke et al. (1996), Aréchiga et al. (1998), and Britt and Gaska (1998) have also reported that PR were more consistent over seasons when synchronization programs were used compared with inseminations after detected estrus.

As discussed previously, herd average DHI data are summarized on an annual basis; therefore, it is difficult to identify seasonal trends in reproductive measures. By using data recorded using DairyComp 305 (Valley



 $\label{eq:Figure 1.} \textbf{Figure 1}. Twenty-one day pregnancy rate (PR), estrous detection rate (EDR), and conception rate (CR) in five large herds using first cycle timed artificial insemination and resynchronization.$ 

Ag Software, Tulare, CA), seasonal patterns can be illustrated. Key reproductive measures from five southern herds, containing 8300 cows, that have implemented TAI programs for first service, as well as resynchronizing cows when they have been diagnosed not pregnant are depicted in Figure 1. Herd 1 was housed in dry-lot conditions with shades and soakers. Herds 2, 3, and 4 were housed in free-stall barns equipped with fans and soakers, except late-lactation cows, which were in dry lots. Herd 5 was housed in free-stall barns equipped with fans and soakers. When evaluating the 21-d PR for 2001, it was evident that all herds experienced a depression in reproductive performance during summer. With the implementation of the TAI and resynchronization programs, overall EDR declined very little in summer; however, all five herds exhibited a marked decrease in summer CR. Thus, on-farm implementation of TAI programs has removed much of the typical decrease in EDR in summer because cows can be inseminated independent of expressed estrus, thereby helping to maintain PR; however, TAI programs can not overcome the negative impacts of heat stress on oocyte maturation, conception, and embryonic development.

# USING EMBRYO TRANSFER TO ENHANCE SUMMER FERTILITY

One potential method of mitigating the negative impacts of heat stress on CR is to use embryo transfer to circumvent those circumstances that reduce oocyte maturation, fertilization, and early embryonic development. In a retrospective study of a commercial embryo transfer program, Putney et al. (1988b) did not detect summer depression in pregnancy rates among recipients of transferred cattle embryos. They followed that study with an experiment to test the hypothesis that embryo transfer would increase the number of pregnancies attained in Florida cows under heat stress compared with AI (Putney et al., 1989a). Embryos were collected from superovulated heifers that were housed under shade after insemination. Mean maximum temperature for the 7-d period from insemination to embryo recovery was not related to numbers of induced corpora lutea, ova recovered, ova fertilized, or transferable embryos; however, the mean maximum temperature was correlated with an increased percentage of embryos classified as severely retarded (≤16 cells), a decreased percentage of embryos classified as morulae, and an increased percentage of embryos graded poor to fair quality. The percentage of cows becoming pregnant was higher for embryo recipient cows than AI cows at 21 d postestrus based on milk progesterone (47.6 and 18.0%, respectively).

Drost et al. (1999) compared the percentage of cows pregnant following AI to those receiving embryos collected from superovulated nonheat-stressed donors or embryos produced from nonheat-stressed donors in vitro. They reported that the percentage of cows pregnant at d 42 in heat stressed cattle was enhanced by the transfer of embryos collected from nonheat-stressed donors (35.4%); however, the percent pregnant following transfer of frozen embryos (18.8%) produced from in vitro fertilization did not improve results compared to AI (21.4%).

Ambrose et al. (1999) investigated the use of in vitro-produced embryos by comparing timed embryo transfer of either fresh or frozen in vitro-derived embryos (produced predominantly in Wisconsin) transferred to cows housed in cooled barns with TAI in Florida in August to October. The percentage of cows becoming pregnant was greater for cows receiving a fresh embryo (14.3%) than for cows receiving either a frozen embryo (4.8%) or TAI (4.9%). One must be cautious in interpreting the results of these two trials as indicating superiority of embryos collected from nonheat-stressed donor embryos to in vitro derived fresh embryos because Drost et al. (1999) conducted their experiment in northern Florida, while Ambrose et al. (1999) used cattle housed

further south in Lake Okeechobee County. Al-Katanani et al. (1999) have reported depressions in fertility as cow location changes from south Georgia, to north Florida, and to south Florida; thus location may influence absolute success rates.

Recently Al-Katanani et al. (2002a) attempted to improve the success rate when using in vitro produced cryopreserved embryos by incorporating vitrification as the cryopreservation method. The cows for this experiment were again located in the Lake Okeechobee area, while the in vitro derived embryos were produced in Wisconsin. Despite using vitrification, more cows receiving fresh embryos in a timed embryo transfer program were pregnant (19.0%) than those receiving TAI (6.2%) or vitrified embryos (6.5%). Although in most studies lower body condition results in lower fertility, in this experiment more cows with a BCS  $\leq$  2.5 were pregnant (15.6%) than cows with a BCS  $\geq$  2.5 (5.5%). There was also a nonsignificant trend for reduced pregnancies at higher levels of milk production.

Although embryo transfer of good quality embryos appears to provide a methodology to enhance summertime fertility, it is not without problems. During periods of heat stress, the number of embryos produced following superovulation may be reduced as a result of fewer oocytes released in response to superovulatory drug therapies, lower fertilization rates, or reduced embryo quality (Hansen et al., 2001). These adverse effects appear more pronounced in dairy cattle than beef cattle, particularly when maximum air temperature exceeds 32°C (Hansen et al., 2001). The recommendations put forth to mitigate the negative effects of heat stress on superovulation are similar to those discussed herein, namely: reduce heat stress with cooling, alter the genetic make-up of the cattle involved, use lower producing or nonlactating donors, and enhance detection of estrus by using synchronization schemes or other detection aids (Hansen et al., 2001).

In his review of embryo transfer to mitigate effects of heat stress, Rutledge (2001) identified the following factors, which have prevented widespread commercial adoption of embryo transfer, to bypass effects of heat stress: 1) using dairy heifers as donors may delay their first parturition, and hence productivity; 2) the negative relationship between embryo quality and ambient temperature means the fewest good to excellent quality embryos are available when the most are needed; 3) embryo recovery is highly technical requiring experienced labor and supplies that add costs; 4) for embryos that have been frozen, so far only those produced in vivo produced frozen embryos have increased the percentage of animals pregnant compared with AI; and 5) the cost of in vitro-produced embryos is reduced compared to in vivo production of embryos; however, in E110 JORDAN

vitro-produced embryos currently only enhance results when transferred fresh not frozen.

Thus, the potential for commercial adoption of embryo transfer to increase reproductive success depends on enhancing the outcome while minimizing cost. One method of potentially improving the success rate of embryo transfer under heat stress is to induce heat shock proteins. When Ryan et al. (1992) applied a pulse heat shock treatment to embryos in culture, they found an increase in the rate of embryos hatching compared with embryos that had been chronically exposed to heat shock, which may be related to the induction of heat shock proteins. Further investigation is needed to determine whether incorporating a brief pulse heat shock treatment on embryos destined to later be transferred into heat stressed cows might improve the success rate of embryo transfer during times of heat stress.

# INDUCING ACCESSORY CORPUS LUTEUM

Rosenberg et al. (1982) and Wolfenson et al. (1988b) have indicated that plasma progesterone concentrations may be reduced in dairy cattle under heat stress, although Wise et al. (1988) showed no difference in plasma progesterone concentrations. In a recent review, Wolfensen et al. (2000) concluded that chronic heat stress reduces progesterone concentrations, although progesterone concentrations may be elevated after acute heat stress. Schmitt et al. (1996) have shown that injection of a GnRH agonist or hCG on d 5 of the estrous cycle results in ovulation of the first wave dominant follicle and formation of an accessory CL, with subsequent elevation of plasma progesterone levels. However, lactating dairy cows receiving injections of hCG on d 5 or 6 after insemination did not have improved CR during heat stress (Schmitt et al., 1996). In this experiment, cows were inseminated based on signs of estrus following synchronization. Further studies following TAI are needed to determine whether inducing an accessory CL might reduce or prevent pregnancy losses that occurred from pregnancy diagnosis via ultrasound at d 27 to 30 to rectal palpation at d 40 to 50, reported by Cartmill et al. (2001), although the negative effects of heat stress on the developing conceptus may prevent elevated progesterone levels from having a positive impact on pregnancy maintenance.

In the summer of 2001, Gandy et al. (2002) synchronized a group of cows using a TAI protocol and divided them into three groups. The first group received no further treatment after the TAI, while the remainder of the cows received 100  $\mu g$  of GnRH on either d 5 or d 11 post-TAI. Progesterone levels in the cows receiving supplemental GnRH were 1 ng/ml higher from d 5 through 17 post-TAI than in the control cows. In addi-

tion, PR were greater for the cows receiving GnRH on either d 5 (32.4%) or d 11 (38.2%) postinsemination compared with control cows (18.9%) that did not receive any supplemental GnRH. The difference between the conception results from this experiment and that of Schmitt et al. (1996) may be the degree of heat stress the cows were under in Florida compared with those in North Carolina or Mississippi (Gandy et al., 2002).

A second potential mechanism by which hCG might enhance conception rate is by minimizing estrogen during pregnancy recognition. Using hCG to induce accessory CL in heifers, Diaz et al. (1998) found that not only was progesterone concentration increased but three-wave follicular cycles were induced when treatment occurred on d 5 after estrus. Furthermore, the dominant follicle did not reach 9 to 10 mm in size until approximately d 20 of the estrous cycle. Additional research is needed to define the range of heat stress conditions under which induction of an accessory CL may enhance CR.

There are two potential drawbacks to commercial application of hCG on d 5 postinsemination. First, hCG is a potential immunogen; therefore, repeated use should be avoided. The second concern is the additional handling of animals. One potential method to circumvent both problems is the incorporation of Deslorelin implants (long-acting GnRH agonist) into the TAI protocol. Ambrose et al. (1998) treated lactating cows (n = 16) with a BCS of 2.5 (five-point scale, 1 to 5) with GnRH (Cystorelin) on d -9 and PGF2 $\alpha$  on d -2. On d 0, half the cows (n = 8) received GnRH and half (n = 8)received a Deslorelin implant. The cows receiving the Deslorelin implant had higher progesterone concentrations. In addition, five of eight cows in the Deslorelin group became pregnant, while only one of eight cows treated with GnRH became pregnant. In general, cows with low BCS (<2.5 on a five-point scale) have been less fertile (Moreira et al., 2000) just as cows under heat stress are less fertile. Whether improvements would be seen under heat stress conditions if Deslorelin implants are incorporated into a TAI program has not been determined. Caution in implementing this protocol is warranted, as Santos et al. (2002) did not find an improvement in CR in a large experiment; however, the interval to reinsemination for cows treated with the Deslorelin implant was increased and CR to a resynchronized TAI was decreased. These negative effects could easily counter any gains in conception rate at the TAI.

### **CROSSBREEDING**

With over 90% of all US cattle Holsteins, an opportunity exists to introduce greater heat tolerance into the current population of dairy cattle by crossbreeding.

Swan and Kinghorn (1992) have reported that heterosis for production traits ranges from 0 to 10%, while fertility ranges from 5 to 25%; thus, there may be opportunities to improve fertility and production simultaneously. According to McAllister (2002): "... only two studies have attempted to characterize the additive and nonadditive genetic influences on multiple lactation or lifetime production." Because those studies were conducted in Illinois and Canada they did not have the opportunity to assess the impact of chronic heat stress on heat tolerance and reproductive performance.

Touchberry (1992) summarized a number of early crossbreeding studies; however, in recent years only a limited number of studies have evaluated the impact of using crossbred European cattle for dairy purposes. In the Illinois crossbreeding experiment of Holstein and Guernsey cattle conducted from 1949 to 1969, milk yield and fat were the traits considered most important; however, various reproductive measures, body size measurements, health events, and milking characteristics were also assessed as part of total performance (Touchberry, 1992). The crossbred cattle had a slightly higher rate of survival than the purebred Holsteins, while the Guernseys had the lowest survival rate. Touchberry reported that crosses of Holsteins and Guernseys actually were older at first calving (9.3 d), required more services per conception, and had longer calving intervals (9.4 d) than purebreds. Although the total dollar value produced per cow per year was 11.4% greater for crossbreds than the average of the purebreds, the total value produced by Holsteins still exceeded that of crosses of Holsteins and Guernseys.

Although not assessing lifetime productivity, Ruvuna et al. (1983) evaluated differences in productivity and reproductive performance between first-lactation purebred and crossbred (Holstein, Jersey, and Brown Swiss) animals based on whether the cows calved in the cool or warm season. In the cool season only the 3/4 Holstein-1/4 Swiss crossbred group exceeded the purebred Holsteins for milk production, while during the warm season both the ½ Holstein × ½ Swiss and the 5% Holstein × 1/4 Swiss × 1/8 Jersey produced more milk. Reproduction was poorer for all breeds and crosses during the warm season. Holsteins had more days open than Jerseys and Brown Swiss, but the differences were not significant. However, during the warm season, Ruvuna et al. (1983) proposed the differences might become of practical significance due to their increased magnitude. They also concluded that crossbreds had superior reproduction with "some indication of even more favorable heterosis in the warm season, especially for Holstein crosses."

Lopez-Villalobos et al. (2000) simulated the potential effects of various crossbreeding scenarios with Holstein, Jersey, and Ayrshire dairy cattle in New Zealand. They

did not report simulation of potential reproductive consequences of the various crossbreeding schemes. Lopez-Villalobos et al. (2000) concluded that upgrading to Jersey cattle resulted in the greatest fat and protein production, but least milk production per hectare. With the New Zealand payment structure, the greatest milk income is achieved from upgrading to Jerseys; however, in the Southeastern United States milk income is derived primarily on a volume basis rather than on a fatprotein basis. In addition, the New Zealand climate is much more temperate than that found in much of the Southern United States; therefore, their model is not directly applicable to the situation in the Southeastern or Southwestern United States.

Swan and Kinghorn (1992) attribute the lack of widespread crossbreeding in dairy cattle in temperate climates to "the notable merit of purebred Holstein strains for milk production." In a study assessing heat tolerance of temperate Bos taurus, tropical Bos taurus, and Bos indicus beef cattle, Hammond et al. (1996) reported that rectal temperatures in Senepol cattle were less than those in Brahman cattle, but the converse was true of respiration rates. Angus heifers on the other hand had the highest respiration rates and temperatures. Because the Hereford × Senepol crosses in this trial had rectal temperatures similar to the purebred Senepol, there appears to be a high degree of dominance associated with the genes responsible for controlling rectal temperature in this tropical Bos taurus breed. Preliminary results from Florida (Olson et al., 1997) provided evidence of a gene influencing hair length and heat tolerance in the Senepol breed. If this gene can be identified as the bovine genome is mapped, perhaps it could be introduced into dairy breeds. Alternatively, a crossbreeding program starting with Senepol, and selecting for high milk production and low rectal temperature during heat stress, might develop a dairy cow with greater heat tolerance without compromising productivity.

Crossbreeding European dairy breeds with Zebu or native cattle has been evaluated for enhancing milk production in the tropics and subtropics. McDowell et al. (1996) evaluated the economic viability of crossing Bos taurus and Bos indicus for dairying in warm climates. Although these cattle have increased growth rates, milk yield, and reproductive rates compared with local cattle in warm climates; they usually have received rations that were inadequate for them to produce to their genetic potential. In general, Bos taurus-Bos indicus crosses do not meet the milk production expectations of US dairy producers and, depending on the management level of the farm in other regions of the world, may result in performance declines when animals are more than one-half European breeds (Mada-

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lena et al., 1990). Furthermore, if the depressing effect of pregnancy on milk yield is greater in Zebu cattle as hypothesized by Madalena et al. (1990), then the beneficial effects of *Bos indicus* influence on heat tolerance would be countered by the negative impact on productivity.

Additional research is needed to determine whether crosses between Holstein, Jersey, Ayrshire, Brown Swiss, Milking Shorthorn, or Guernsey cattle, perhaps with some genes from Senepol, can be developed that will meet the milk production expectation of US producers, while capitalizing on hybrid vigor in survival traits such as calf viability, disease resistance, and reproductive rate. This research must assess not only first-generation crossbreds, but explore viable alternatives for subsequent generations as Freitas et al. (1998) have done for the European dairy breed crosses on Zebu and native cattle. The evaluation should include climatological influences and consider the various milk pricing mechanisms used in different Federal Milk Marketing Orders to determine which purebred or crossbred scenario provides the greatest economic opportunities.

### **CONCLUSIONS**

Heat stress reduces reproductive efficiency of dairy cattle through a variety of different mechanisms. To accurately quantify the magnitude of the reduction in reproductive efficiency occurring during the summer in the United States, calculation of DHI reproductive management variables must be available on a monthly basis rather than a rolling annual basis. Use of various cooling methods can improve fertility of cows under heat stress compared with uncooled cows. Because negative effects of heat stress have been identified from 42 d before insemination to 40 d after insemination, continual cooling must be provided. Although TAI programs eliminate failure to detect estrus as a reason for reproductive inefficiency under heat stress, CR is not enhanced. Two methods that show promise in enhancing CR in the summer are transfer of embryos collected from superovulated donors and induction of accessory CL. Research is needed to evaluate the potential of crossing traditional and nontraditional dairy breeds to enhance reproductive capability, while maintaining productivity at levels acceptable to US dairy producers. Comparative economic implications of the various strategies are needed as well, perhaps using modeling techniques to examine multiple scenarios.

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