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EFFECTS OF HEAT STRESS ON REPRODUCTIVE ACTIVITY IN DAIRY COWS BRED IN THE POTENZA DISTRICT.

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ABSTRACT - The effect of heat stress (HS) on the reproductive performance in dairy cows reared in Apennines areas of Southern Italy was evaluated. Reproductive parameters obtained from three farms during the period 2007-2012 were related to either season variations or the temperature-humidity index (THI), i.e., a complex climate parameter obtained by the maximum temperature and minimum relative humidity. The THI was able to assess the HS effects on parameters as conception rate on an annual basis (CRY) ($R=-0.437$; $P<0.01$) but was less efficient for parameters as the conception rate (CR). Whereas (i) CRY is influenced by both heat detection rate (HDR) and CR; (ii) an indirect analysis detected a significant ($P<0.001$) reduction in the HDR along with THI increase; and (iii) CR was only partially affected by either THI or season, it follows that the main cause of reduced fertility in the farms surveyed was the HDR. The number of days open was significantly larger in the animals calved from January to July than in those calved between August and December (163±33 *vs* 123±36 days; $P<0.001$); this increase may be because of the rescue of reproduction activity in the cows calved during the former period coincides with HS occurrence.

INTRODUCTION - The decrease of the productive and reproductive performance of livestock in conditions of high environmental temperatures is a phenomenon known for some time that is, for example, the main problem related to breeding high production dairy cattle in tropical and subtropical areas (Kadzere et al., 2002). Global warming and the breeding of selected animals that are more and more sensitive to environmental effects have made this phenomenon, named heat stress (HS), particularly relevant even in temperate areas (De Rensis and Scaramuzzi, 2003). The origin of HS comes from trying to counteract hyperthermia induced by the high temperature with mechanisms of thermal dispersion. These mechanisms are, however, also influenced by other parameters, such as the relative humidity, the ventilation, the radiant energy and the rainfall (Igoro et al., 1992). Complex indexes, which combine several of the above parameters involved, have been proposed to monitor the effects of HS. The temperature-humidity index (THI), that combines the maximum temperatures with the minimum relative humidity, is the most used one (Rovagnano et al., 2000).
The infertility caused from the HS is due to both direct and indirect causes that affect reproductive performance. In particular, HS may act directly by reducing the quality of the gametes. In the male, there is a reduction in the number of spermatozoa and an increase of oligo-astheno-teratospermia (Meyerhoeffer et al., 1985). In the female, several alterations are described either in the processes of folliculogenesis, as prolongation of follicle dominance, co-dominance phenomena, or luteogenesis, as a delayed luteolysis (Wilson et al., 1998). The follicle and oocyte maturation are compromised and the mature oocyte has a reduced competence for fertilization and

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embryonic development (Roth, 2008). The lowering of the levels of estrogen produced by the growing follicles translates into a lower intensity of estrous signs (Roth, 2008). The effects attributable to HS are prolonged up to 40-60 days from the end of the high environmental temperature (Roth, 2008) and are reduced with the progress of the embryo-fetal development (Biggers et al., 1987). Indirect effects due to HS are attributable to a reduction of dry matter intake, which is responsible for a deepening and extension of the negative energy balance postpartum and results in reproductive failures (Lucy et al., 1992). Several techniques have been proposed in order to counteract the effects of HS on reproductive activity, as the use of awnings, showers, fans (West, 2003); however, they are able to alleviate the productive inefficiency without solving reproductive failures. New strategies to combat infertility are geared to accelerate the follicle renewal by hormonal treatments or ovum pick-up as well as by using fixed-time artificial insemination and embryo transfer.
The purpose of this study was to evaluate in an Apennine area of Southern Italy, such as the province of Potenza, the magnitude of the effects exerted by HS on reproductive activity in high producing dairy cows and the compliance of the THI for HS evaluation.

MATERIALS AND METHODS - Reproductive data from January 2007 to December 2012 were collected from a total number of 1,743 Holstein Friesian (HF) cows bred in three dairy farms in the province of Potenza. Based on information collected on the date of birth, the calving date, the date of the inseminations and the culling date, reproductive parameters were developed such as age at first calving, conception rate on a per year basis (CRY), conception rate (CR), the number of artificial inseminations on a per year basis (NAI), the number of days open (DO) in relation to the calving month.
Meteorological data were collected from weather stations site nearby (less than 3 km) and at the same altitude of examined farms (source, ALSIA). The parameters considered were: maximum, minimum and average temperature and maximum, minimum and average relative humidity. As proposed by Ravagnolo et al. (2000), the parameter maximum temperature (T) has been combined with the parameter minimum humidity (H) in order to obtain a new complex parameter, i.e., Temperature-Humidity Index (THI), as follows: $THI = (((9/5) * T) +32) - ((11/2) - (11/2) * H) *(((9/5)*T)-26))/100$
This new parameter was found to vary on average from 22 to 157. THI classes were, then, ranged according to arbitrary intervals in order to simplify the analysis of the data. Statistical analysis was performed by Linear Regression and ANOVA (Systat 11.0 release) by including in the model variables as year and farm. Season variations were evaluated on a month basis. Before statistical processing, all parameters expressed as annual incidence rate were arcsine transformed and the homogeneity of variance was assessed.

RESULTS - The data analyzed clearly shows the repetition frequency of calving with seasonal peaks. Subtracting the duration of gestation equivalent to 284 days (source ANAFI, 2013) to the calving date, we obtained the conception date. This datum was related to the total conceptions per year and a new parameter, called "conception rate on a per year basis" (CRY), was obtained. Evaluating the performance of this parameter in relation to THI, we observed that CRY and THI followed an inverse pattern. CRY was significantly affected by either THI ($P <0.001$) or month ($P <0.001$). Creating classes of THI and comparing them to CRY, a significant ($P <0.01$) decrease was observed along

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with THI increase (Figure 2); this relationship was expressed by a straight linear regression with R=-0.437 (P <0.001). The annual incidence of the number of inseminations (NAI) showed a significant effect of both the THI (P<0.04; R=0.329) and months (P<0.001). Comparing the number of conceptions with the number of inseminations on a per month basis, we obtained the actual conception rate (CR). CR was significantly affected by the year (P <0.003) but not significantly affected by either months or THI, in spite of a significant decrease in the period between April and August with respect to the other months of the year (27.3% *vs* 35.4%; P <0.01) and a sharp drop between 90 and 100 THI (R=-0.303). The age at first calving did not show statistically significant differences in relation to the year, month, THI and farms, ranging in relation to the latter parameter from 26 to 27 months. Evaluating the number of DO in relation to the calving month, we found a significant effect of the month (P <0.001), but not of the THI. In relation to the month effect, there was a significant reduction in the number of DO between August and December compared to January and July (123 *vs* 163 days, P <0.001), ranging among farms from 35 to 55 days (Figure 3).

Figure 1. Mean patterns of the conception rate evaluated on a per year basis (CRY) and temperature humidity index (THI) in the three evaluated farms.

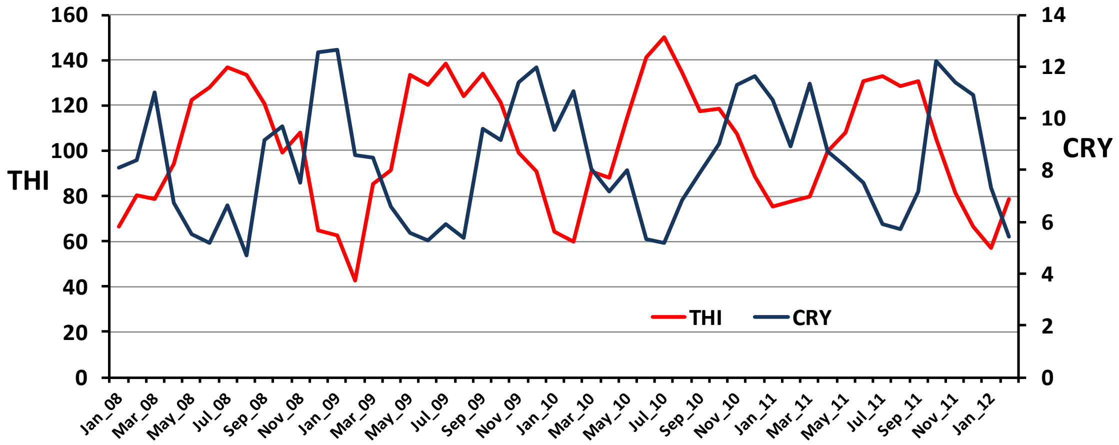


Figure 2. Relationship between CRY and THI.

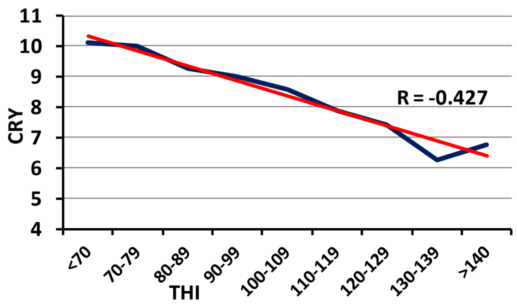
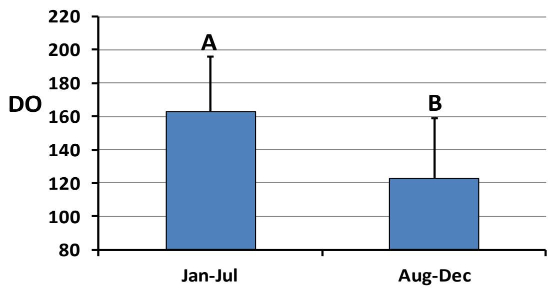


Figure 3. Mean (+SD) number of days open (DO) in relation to the calving month.



A,B (P< 0.01)

DISCUSSION – The results of this study clearly demonstrated that the reproductive performance of dairy cows are greatly influenced by the season and that the parameter THI allows only partly to highlight the relationship between HS and reproductive

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efficiency. CRY, similarly to the Pregnancy Rate (PR) (Ferguson and Skimore, 2013), combines the heat detection rate (HDR) and conception rate (CR). The lower CRY during the hot season may result from either a decreased detection of estrus or a lower quality of gametes and embryos; both these failures are involved in determining the effects of HS in cattle and the latter causes a lower CR (Roth, 2008). We did not record the given number of heats detected but the number of inseminations. These two parameters, in practice, are equivalent since each heat detected was followed by one artificial insemination; hence, we may postulate that NIA=HDR. During the summer period the frequency of heats detected was significantly reduced either when related to months (P< 0,001) or THI (P< 0.04). On the other hand, CR did not show statistically significant differences in relation to either the month of the year and the THI. This clearly indicates that the reduction in fertility observed during the summer season, as assessed by CRY, may be mainly due to a lower identification of animals in heat. The large temperature excursion, which is typical of the Apennines areas, may attenuate the negative effects of HS on the quality of gametes responsible for the lowering CR, as recorded in other geographical areas (Roth, 2008). This result is comforting and shifts the focus to a possible improvement in reproductive efficiency of cattle bred in the territory examined by using different strategies to contrast the low HDR, as heat-watch, pedometer, fixed-time artificial insemination.

The lack of significant effects on the age at first calving suggests a relative refractoriness of heifers to HS, as found in previous studies (West, 2003). A good management of the calving timing of the heifers could be a reliable strategy to reduce the seasonal variability of milk production and/or to direct the calvings towards the period that provides the lowest number of DO.

Another consequence of HS may be the large variation of the number of DO in relation to the calving month with a larger number of DO recorded from January to July with respect to the August-December period. Considering that in this study the minimum mean number of DO, calculated on a monthly basis, was 118 days, the animals born in January outweighed their negative energy balance postpartum and were able to rescue their reproductive activity at the month of April-May, when temperatures responsible for HS occur. Conversely, those who delivered after July overcame the problem by matching their reproductive recovery with a more favorable climate. Since in the period from August to December the animals calved were on average 57.1% of the total calved animals in the five years evaluated, it follows that the HS affected the calving interval with an average annual increase of 17.1 days.

In conclusion, heat stress caused many failures on reproductive activity in dairy cows reared in Apennines areas of Southern Italy, i.e., a lower heat detection rate and a larger number of days open. Strategies should be ranged to counteract the effects of heat stress on reproduction in order to attenuate the relevant economic losses caused by high environmental temperatures.

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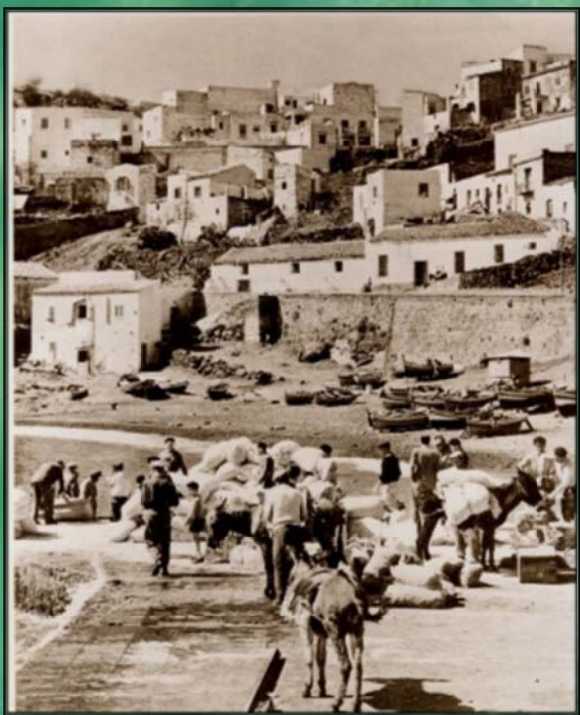

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