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### ► To cite this version:

S Mattalia, Aurélie Vinet, J Vandenplas, M J Carabaño, C Diaz, et al.. Towards new breeding tools in a context of climate change: first results of the RUMIGEN project on new phenotypes for heat tolerance traits. 45th ICAR Annual Conference, ICAR, May 2022, Montréal Québec, Canada. pp. 195-202. hal-04478350

**HAL Id: hal-04478350**

**<https://hal.inrae.fr/hal-04478350>**

Submitted on 27 Feb 2024

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## Towards new breeding tools in a context of climate change: first results of the RUMIGEN project on new phenotypes for heat tolerance traits

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RUMIGEN is a project financially supported by the EU that aims to develop breeding programs capable of managing the trade-offs between efficient production and resilience to extreme climate conditions. RUMIGEN is designed under a multi-disciplinary approach that mixes competencies in both genetics and social sciences. The genetic approach aims to enhance genomic selection using three levers: quantitative genetics, genome editing, and epigenetics.

One of the objectives of RUMIGEN is to enlarge selection criteria and to provide genomic tools to select dairy cows tolerant to heat stress. Studies are dedicated to the definition of heat-tolerance traits based on production, reproduction and health records, as well as to the study of the trade-offs between these traits, and with those already selected. These analyses are based on performances recorded in commercial herds in France, Spain and the Netherlands (i.e., milk production traits and somatic cell scores recorded by Milk Recording Organizations, and fertility traits derived from AI events), in combination with meteorological data obtained from the corresponding Meteorology Agencies. Records are associated to meteorological information at the farm level, in order to measure the impact of heat stress. First results obtained for different breeds and in a large range of farming and climatic scenarios showed that the combination of both types of information is relevant to measure the decline of performances due to heat stress and to define heat stress indicators and new traits for future breeding tools.

*Keywords: Dairy cattle, animal breeding, heat tolerance, climate change, new phenotypes.*

### Abstract

## Introduction

Social expectations are more and more pushing for the development of sustainable breeding programs and linked technologies, as well as of breeding for adaptation to climate change while taking into account genetic diversity. The RUMIGEN program (<https://rumigen.eu/>), financially supported by the EU, aims to produce robust and efficient cattle able to manage the trade-offs between production and adaptation to extreme climate conditions.

To reach this goal, RUMIGEN gathers a large panel of competences. Research in quantitative genetics are focused on the study of heat tolerance traits and of the impact of climate change on the trade-offs between traits. The partners also study new approaches to maintain genetic diversity using genome information and the potential use of epigenomics information to enhance genomic predictions. Another ambition of this program is to improve the understanding on how genetic and epigenetic processes shape the phenotypes. Several experiments will be conducted to determine the impact of environmental stressors (metabolic demands during the gestation, heat, immunity challenge and pathogens). Epigenotyping tools will be used on a large scale to study the variability of the epigenomic profiles, their transmission between generations, and their impact on phenotypes. Regarding new biotechnologies, genome editing has been also suggested as a promising tool to speed up selection and design disease resistance alleles. However, it raises technical and social issues since only few data are available regarding the genome integrity (off-targeting, rates of de novo mutation). This is why RUMIGEN aims to assess the potential of genome editing to speed up introgression and the security of genome editing in different conditions. Finally, animal breeding brings not only biological, technical and economical issues, but also societal concerns with respect to breeding objectives, biodiversity and the potential use of new breeding technologies. Therefore, a work-package of RUMIGEN is dedicated to the social acceptance of breeding and the related technologies.

One of the main objectives of RUMIGEN is to provide breeding tools to face selection under the harsh environmental conditions generated by climate change. For this purpose, France (Idele, INRAE), Spain (INIA—CSIC, IRIAF) and the Netherlands (Wageningen University and Research) collaborate to define new traits related to heat tolerance. Large-scale data from commercial farms are combined with meteorological information in order to highlight mid-long term impact of heat stress on performances and to define new traits related to heat tolerance.

The objective of this paper is to present the first results of using performances of Spanish, French and Dutch dairy cows recorded by Milk Recording Organizations (MROs) and associated meteorological data to measure the impact of heat stress period on production and health traits.

## Material and methods

### *Performances of the cows used in the analyses*

Test-day (TD) records for milk yield (MY, kg/d), fat yield (FY, g/d), protein yield (PY, g/d), fat content (FC, %), and protein content (PC, %) for French, Spanish, and Dutch cows, were extracted from the respective genetic evaluations for production and health traits. In addition to these traits, TD records for somatic cell score (SCS; defined as  $SCS = 3 + \log_2 (SCC/100,000)$ , with SCC being somatic cell counts in cells/ml) were available for French and Dutch cows.

Three breeds were involved in the analyses: the Holstein population, studied in the three countries, and two regional breeds, that is Montbéliarde studied in France, and Meuse-Rhine-Yssel (MRY) studied in the Netherlands.

For all countries, the extracted datasets covered approximately a period of 10 years, starting from the 2010s. Different edits were applied across countries. For example,

after editing, the analyses carried out cover out the period 2016 - 2020 for France, and the period 2010 to 2020 and 2021 for Spain and the Netherlands, respectively. All countries analysed the different lactations separately with univariate models. Common edits were applied by all three countries on parity, age of calving at different parities (age of calving between 23 and 42 months and 35 to 60 months for 1<sup>st</sup> and 2<sup>nd</sup> parities respectively) and extreme phenotypic values. In all countries, only cows with known parents were considered. Only Days in Milk (DIM) between 5 and 305 were retained in France and in the Netherlands, between 5 and 400 in Spain.

Briefly, the datasets with production and health traits (MY, PY, FY, PC, FC, and SCS) for Holstein included around 36 million test-day records associated with around 7 million French first- and second-parity Holstein cows, around 20 million TD records associated with around 1 million Spanish first- to third- parity cows, and around 7 million TD records associated with around 500 thousand Dutch first- and second-parity Holstein cows. These cows were distributed in around 45,000 French herds, around 4,600 Spanish herds, and around 1,500 Dutch herds.

Meteorological data were provided by Météo-France (Safran database) for France, by the National Meteorological Agency (AEMET) for Spain, and were extracted from the Koninklijk Nederlands Meteorologisch Instituut (KNMI) website for the Netherlands. Weather records were available for the French territory in the form of a grid of 9,892 8x8km squares, for 1,993 Spanish weather stations, and for 34 Dutch weather stations. Each herd was connected to these meteorological information through the (partial) ZIP code of the farm.

#### Weather data

For each daily record measured in each weather station, a Temperature – Humidity Index (THI) was computed using the NRC (1971) formula:

$$THI = (1.8 \cdot T + 32) - (0.55 - 0.0055 \cdot RH) \cdot (1.8 \cdot T - 26)$$

with T being the average daily temperature (degrees Celsius) and RH the average daily relative humidity.

The phenotypes (MY, FY, PY, FC, PC and SCS) of each lactation were analysed in separate studies. THI was calculated as the average THI of 3 days (in Spain and in the Netherlands: 3 days before the record; in France: day of TD and 2 days before).

#### Models

For each population, the effect of THI on phenotypic performances at the population level was estimated using the following model:

$$y = X\beta + Z_1 a + Z_2 p + e$$

where y,  $\beta$ , a, p and e are respectively the vectors of phenotypes, fixed effects, additive genetic random effects, random permanent environment effects and the random residuals respectively, and X, Z<sub>1</sub> and Z<sub>2</sub> are the incidence matrices for the listed effects.

The fixed effects were almost the same in all countries, excepted the gestation stage that was included in the French and in the Dutch model but not in the Spanish one. However their combination differ from one country to the other:

- France: Herd-Year, THI, DIM, gestation stage, month of calving and age at calving.
- The Netherlands: Herd-Year, THI, DIM, gestation stage, age at calving-year- season.
- Spain: Herd-Year-Season, THI, DIM and age.

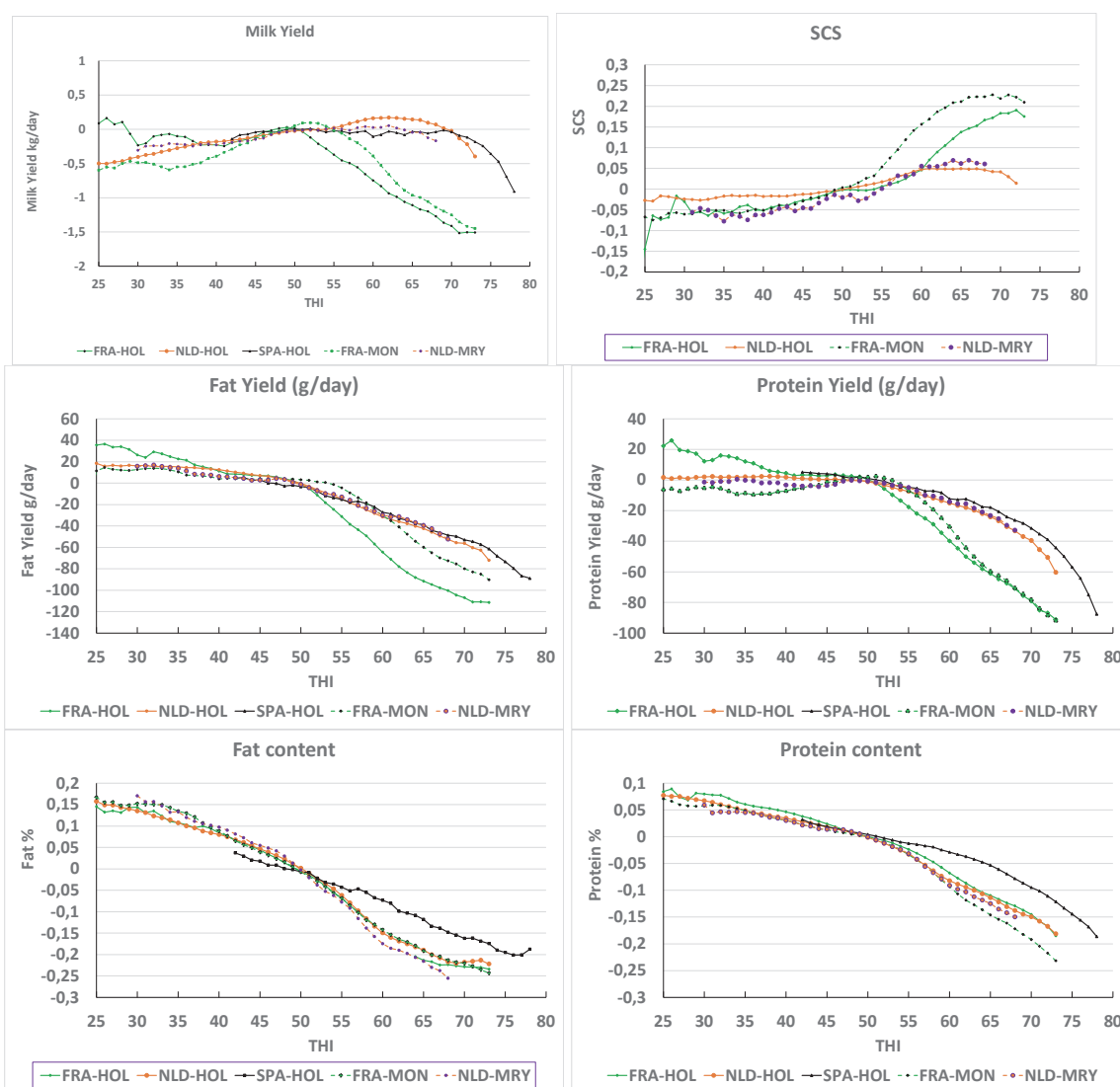
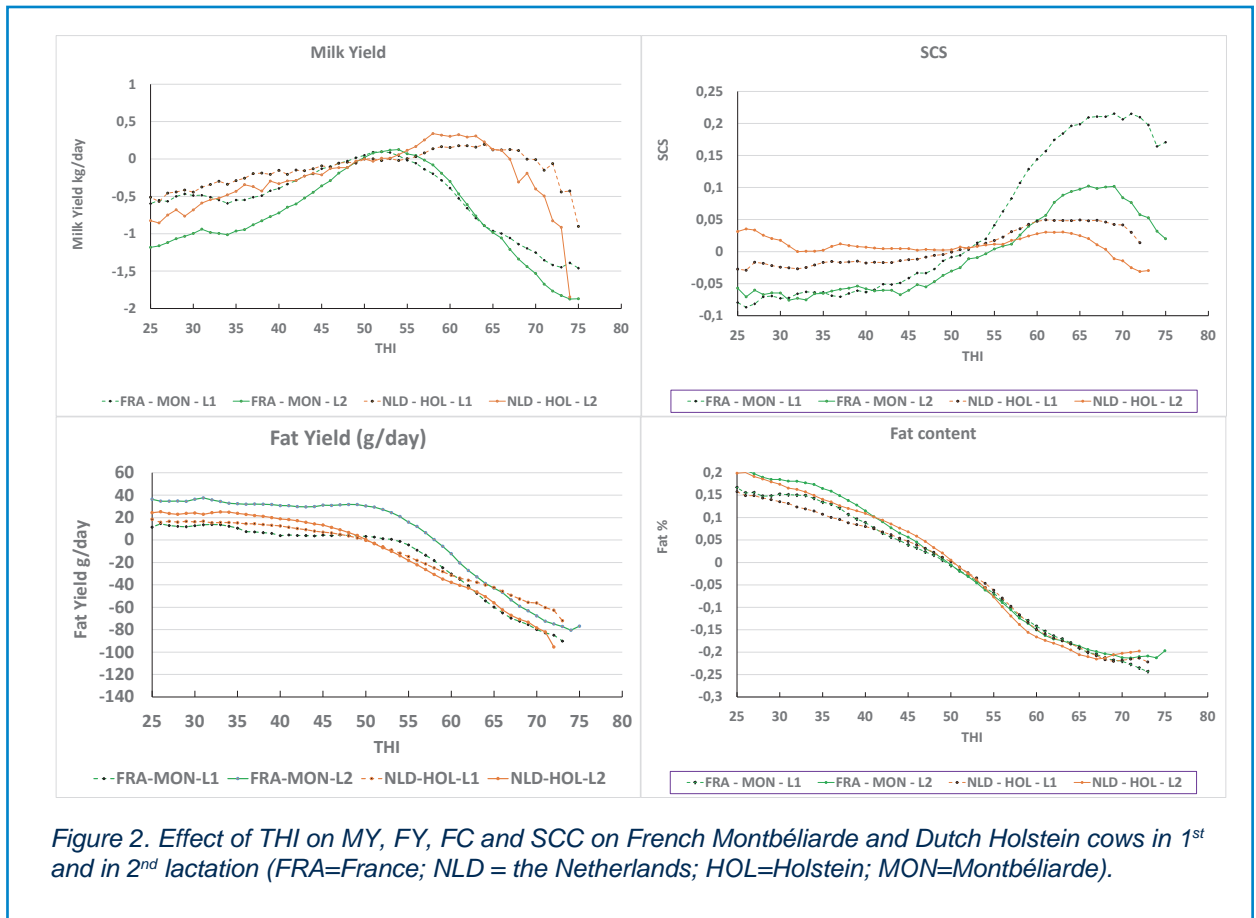


Figure 1. Effect of THI on MY, FY, PY, FC, PC and SCS on cows in 1<sup>st</sup> lactation, according to the country and to the breed (FRA=France; NLD=the Netherlands; SPA=Spain; HOL=Holstein; MON=Montbéliarde, MR Y=Meuse-Rhine-Yssel).



The estimated effects of THI on MY, FY, PY, FC, PC for 1<sup>st</sup> parity cows of the three breeds and of the three countries, and on SCS for Dutch and French populations are presented in figure 1. The estimated THI effects on MY, SCS, FY and FC in 1<sup>st</sup> and 2<sup>nd</sup> lactations are compared on figure 2 for French Montbéliarde and Dutch Holstein cows. Estimated THI effects presented on both figures were averaged using a rolling basis of 5 THI units to smooth the fluctuations.

## Results and discussion

This study covered several breeds, climatic conditions and farming systems. A common approach was defined between countries and breeds to identify heat stress indicators and to measure their impact on the cow performances.

Dutch results of estimated effects of high THI must be interpreted carefully since the Netherlands are exposed to oceanic climatic conditions, with moderate daily variations. Only 66 days with a daily THI over 72 were observed within 10 years (2010-2019). However, THI over 60 were observed on average 120 days each year during the same decade, which means that the effect of a moderate heat stress can be measured using Dutch data. The frequency of days with very high THI was much higher in France and in Spain (eg. in France, pending on the region, between 88 and 345 days with a THI over 72 observed within 5 years).

This study showed that increasing THI had a negative impact on all studied traits, but more prominent for production than for SCS. To illustrate that, the estimated decrease in FY between 50 and 70 units of THI (ie., 9 and 22 °C of daily average temperature for

the relative humidity in summer in France) corresponds to 11% of the daily production of 1<sup>st</sup> parity French Holstein cows. The pattern of the curves of THI effects depended on the trait: for MY, SCS and PY, the THI effects were almost stable below 50 (with some exceptions such as the French Holstein cows for PY), while a decline was observed all along the THI scale for FY, FC and PC.

Some differences of magnitude of the estimated effects and of THI-thresholds were observed between countries, more than between breeds, particularly for the yields and for SCS. For MY, FY and PY, moderate THI (50-70) had a stronger impact on the performances of French females than for the other populations. For SCS, the effect of increasing THI over 50 was stronger for the 1<sup>st</sup> parity French cows. The comfort regions observed in France were relatively low when compared to the literature (Carabaño *et al.*, 2017) and to the two other countries. This could be explained by differences of farming systems, that are often based in France on pasture. Thus French cows are often exposed to outside temperatures (even if they are kept inside during very hot periods) which is almost not the case in Spain. In the Netherlands, a large proportion of females included in this study were raised in farms equipped with automatic milking systems, with less grazing and potentially equipped with barns more adapted to heat than the average Dutch farms. Brügemann *et al.* (2012) also reported lower milk yield losses in feeding systems based on crop production than on pasture. The exposure to wind, more frequent in summer in oceanic regions than in continental ones may also explain some differences. In Spain, a probable acclimatization along the large periods of high temperatures during the summer and the use of heat abatement devices and mitigation practices in many farms could also explain some differences with the two other countries.

## Conclusion

This study showed that heat stress indicators could be defined, combining performance and meteorological data. Increasing THI, mostly due to increasing temperatures, had a negative impact on all studied traits and all breeds. The estimated impact differed from one trait to the other and it was more prominent for production than for SCS. Therefore, heat stress tolerance is a complex trait. The studies are going on with reproduction traits in order to get a global view on the major traits included in breeding goals in dairy cattle.

Differences of magnitude of the effects of heat stress and of THI-thresholds were observed between countries, more than between breeds. This could probably be due to differences in farming systems and, in the case of Spanish cows, an acclimatization along the summer periods.

## Acknowledgment

This study received funding from the European Union's Horizon 2020 research and innovation program under grant number 101000226 (Rumigen). This project adheres to EuroFAANG (<https://eurofaang.eu>). The CAICalor project was funded by APIS-GENE. The authors thank Meteo-France, AEMET and KNMI for the meteorological data and INRAE-CTIG, CONAFE and CRV for the performance and pedigree data.

The use of the high-performance cluster was made possible by CAT-AgroFood (Shared Research Facilities Wageningen UR, Wageningen, the Netherlands).

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