



(RESEARCH ARTICLE)



## Enhancing honeybee breeding for sustainable agriculture through temperature and relative humidity monitoring

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### Abstract

Through their vital role in pollination, honeybee colonies play a crucial part in sustaining biodiversity and ensuring global food security. This paper aims to: 1) assess temperature and relative humidity variations within the hive and at the brood level; 2) determine genetic parameters for these traits; and 3) predict Breeding Values (PBVs) for honeybee colonies. Temperature and relative humidity data, during the period 2020-2023, were collected in Northern Tunisia using sensors placed inside hives and at the brood level. A dataset comprising 214,128 records for temperature and relative humidity within hives, sourced from 317 devices, was used in this study. Additionally, 20,740 records for temperature and relative humidity obtained from 78 brood-level devices were incorporated into the analysis. Phenotypic and genetic parameters were computed for the four examined traits, and using a BLUP Animal model, colony breeding values (PBVs) were predicted. Main results indicated a highly significant influence ( $p < 0.01$ ) of the month effect on the four temperature and relative humidity traits. Heritability estimates for in-hive temperature, in-hive relative humidity, brood relative humidity, and brood temperature were 0.14, 0.12, 0.16, and 0.28, respectively. Positive correlations were observed between relative humidity inside hives and at the brood level, as well as between temperature within beehives and at the brood level. Colony breeding values were predicted to select the best adapted bee queens to enhance honeybee's sustainable use under Southern Mediterranean climatic conditions.

**Keywords:** Honeybees; Temperature; Relative Humidity; Selection

### 1. Introduction

Numerous studies have shown that temperature and relative humidity are key traits in controlling honey bee colony activity [1,2]. Temperature translates the activity of bee workers within the hive and at the brood level. Maintaining a proper temperature range of 32 to 36 °C was explicitly reported to be critical for the development of broods [1,2]. The length of immature honey bee development, emergence rate, coloration of emerged bees, adult brain function, learning capacity, disease prevalence, and wing morphology are just a few of the variables that can suffer from deviations from this range [3,4,5]. However, foraging activity is positively influenced by ambient temperature [6]. Bee foraging is negatively impacted by high temperatures, whereas flying activity is hampered by low temperatures [7].

The honey bee colony places a great deal of attention on relative humidity. In beehives, a suitable range of humidity is thought to be above 75% [8], with the growth of brood generally requiring high humidity [9,10]. Relative humidity does not directly affect honey bees during outdoor activities like foraging. However, honey bee workers use a variety of actions, such as nectar water evaporation and water collecting, to increase humidity when relative humidity levels

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inside the hives fall [10]. According to studies, bees subjected to high temperatures show an increased thirst for water [11]. Therefore, integrating relative humidity and temperature is essential for tracking honey bee activity.

It is significant to note that different honey bee breeds and strains may respond differently to heat stress and relative humidity conditions. Because they have evolved responses to ecological pressures, certain honey bee breeds can flourish in specific areas. The main goals of the current study were to: 1) Assess honeybees' capacity to maintain optimal hive and brood conditions in the Southern Mediterranean region; 2) Estimate genetic parameters for critical honeybee and brood temperature and relative humidity traits; and 3) Predict breeding values (PBVs) of honeybee colonies for these traits.

## 2. Material and methods

Recorded temperature and relative humidity generated by sensors from a total of 317 devices in Northern Tunisia (<https://www.ugfsnorthafrica.com.tn/fr/portfolio/iris-technologies-2/>) were used in this study. The original data were edited by SAS software and analyzed using model (1):

$$Y_{ijkl} = \mu + byd_i + m_j + bt_k + e_{ijkl} \dots\dots\dots(1)$$

where:

$y_{ijkl}$ : The hive temperature, the hive relative humidity (RH<sub>c</sub>) and the brood temperature and the brood relative humidity (RH<sub>b</sub>);

$\mu$ : The population mean;

$y$ : The effect of the beekeeper-year-day class which represents the beekeeper management on measured variables;

$m_j$ : The effect of the month of the year;

$t_k$ : Covariate time when the record is taken;

$b$ : The linear regression coefficient;

$e_{ijkl}$ : The random residual error.

Heritability estimates, along with Colony Breeding Values for the different temperatures and relative humidity traits were derived using MTDFREML software [12]. The single-trait animal model (2) with its assumptions (3,4,5), expressed in matrix notation according to [13], was used.

$$Y = Xb + Z_1a + Z_1pe + e \dots\dots\dots(2)$$

where:

$Y$ : The vector of observations of beehives temperature, beehives and brood relative humidity;  $b$ : The vector of fixed and covariate effects (beekeeper-year-month class and day-time class);  $a$ : The vector of colony random additive genetic values with  $\sigma_a^2$ ;

$pe$ : The vector of random permanent environmental effects  $\sigma_{pe}^2$ ;

$Z_1$  and  $Z_1$ : known incidence matrices;

$e$ : The error vector

Assumptions:

$$\text{Var} (a) = G = I\sigma_a^2 \dots\dots\dots(3)$$

$$\text{Var} (P_e) = I\sigma_{pe}^2 \dots\dots\dots (4)$$

$$\text{Var}(e) = R = I\sigma_e^2 \dots\dots\dots (5)$$

### 3. Results and Discussion

#### 3.1. Means and variation of temperature and relative humidity in beehives and brood

In Southern Mediterranean environments (Tunisia), average temperature and relative humidity of *Apis Mellifera Intermissa* were 22.83 °C±7°C and 25.65°C± 7 °C inside hives and at the brood level, respectively (Table 1). When compared to the relative humidity inside hives (71.49 ± 16.23), the relative humidity at the brood level was greater (75.36 ± 17.45). Similar results were reported by [14].

**Table 1** Means and variation of beehives temperature, beehives and brood relative humidity

Trait	Number of hives	Number of records	Mean	Standard deviation
In-hive Temperature	317	204 504	22.83	7.03
In-hive Relative Humidity	317	203 525	71.49	16.23
Brood Temperature	78	20 118	25.65	7.05
Brood Relative Humidity	72	20 260	75.36	17.45

#### 3.2. Frequency of hives with optimal conditions

As per [1], the ideal temperature range for the brood level falls between 30°C and 36°C. The frequencies of the investigated brood instances falling within these optimal temperature intervals are detailed in Table (2). Notably, the highest frequencies were documented during the summer months, with July registering at 39% and August at 46%. In contrast, the lowest frequencies occurred during early winter, specifically in November (15%) and December (20%).

Examining relative humidity (RH) frequencies, a substantial proportion of hives (60-65%) exhibited optimal conditions from June to October. Notably, the brood level displayed its highest frequency in January. These findings shed light on the seasonal variations in both temperature and relative humidity that impact the brood level. The concentration of optimal temperature frequencies during summer months aligns with the natural climatic patterns, while the observed RH frequencies highlight specific months conducive to favorable conditions for hive development. The nuanced understanding of these patterns provides valuable insights for beekeeping practices and hive management, aiding in optimizing conditions for brood development throughout the year.

**Table 2** Frequency of hives and broods with optimal environmental conditions

Month	30°C≤T <sub>2</sub> *≤36 °C	50%≤RH <sub>1</sub> *≤75%	50%≤RH <sub>2</sub> *≤95%
1	24.75	30.19	93.76
2	52.55	30.72	27.32
3	18.99	35.84	42.11
4	24.48	43.99	35.54
5	14.41	47.43	39.85
6	17.66	58.29	45.61
7	38.86	60.50	36.45
8	45.53	65.89	42.11
9	22.40	65.35	48.99
10	25.09	62.86	35.69
11	15.06	48.44	27.40
12	19.92	34.59	28.40

$T_1^*$ : In-hive Temperature,  $T_2^*$ : Brood Temperature;  $RH_1^*$ : In-hive relative humidity,  $RH_2^*$ : Brood relative humidity.

For bee worker longevity, brood health, development, and egg hatching, relative humidity is a crucial microclimatic variable [8,10,15]. Compared to the rest of the hive, the brood needs a little bit more humidity. Studies reported by [16, 17], showed that the ideal brood humidity ranges from 50% to 90-95%. According to [18], honeybee eggs need a relative humidity of above 50% to properly hatch, with the best survival rates occurring between 90 and 95% [9].

Due to the high humidity, parasitic Varroa mites are able to reproduce less frequently [19]. However, it has been demonstrated that when humidity levels rise, mature honeybee longevity decreases [20]. According to [21], honey bee workers fare better under conditions of relative humidity of 75% than they do under conditions of low relative humidity, particularly 15% to 50%. Therefore, it is desirable to keep the beehive's total humidity between 50% and 75%. This range helps prevent both extremely wet conditions that can encourage the growth of mildew or fungus and overly dry conditions that can dehydrate bees and their food supplies. Therefore, honey bee workers would need to use a variety of behaviors, such as fanning behavior, carbon dioxide regulation [22], nectar dehydration, water collection and spreading in the nest [15], to regulate relative humidity to optimal levels in the brood nest.

Besides the function of bees, there are many beekeeping techniques designed to get the temperature inside hives to the ideal level, especially during extreme weather conditions. In order to help the hive retain heat in low-temperature situations, the entrance size can be decreased or an entrance reducer can be used. In order to monitor beehives continually, it is crucial to install specialized equipment such hive thermometers, temperature and humidity sensors (SmartBee devices), and digital temperature control systems

### 3.3. Sources of variation

According to the sources of variation analysis (Table 3), the effect of the month, the beekeeper-year-day contemporary group and the time when the records were taken were highly significant sources of variation ( $p < 0.01$ ). These results demonstrated that beehive management and practices have a significant impact on the temperature and relative humidity traits inside beehives and at the brood level, beside colonies genetic makeup.

**Table 3** Sources of variation of temperature and relative humidity inside hives and at the brood level

Source of variation	In-hive Temperature	In-hive Relative Humidity	Brood Temperature	Brood Relative Humidity
Beekeeper-year-day	**	**	**	**
Month	**	**	**	**
Time	**	**	**	**
Error df	199 338	198 382	19 194	19 341
R <sup>2</sup>	0.66	0.54	0.85	0.84

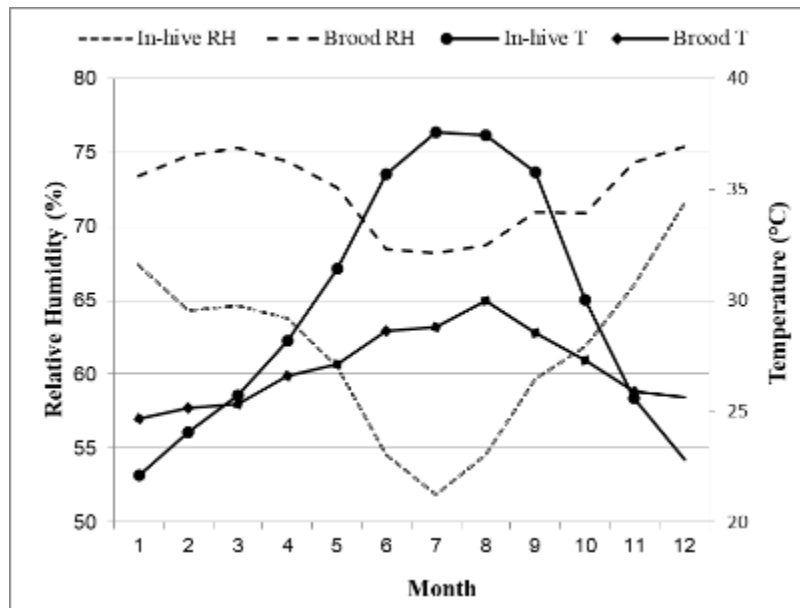
\*\*  $P < 0.001$

### 3.4. Effect of the month of the year

The month effect on temperature and relative humidity inside hives and at the brood level is shown in figure (1). These results were obtained by using the Least Squares Solutions from model (1). They were in agreement with those obtained by [14]. The temperatures inside beehives were rather low from November to March. January, which is the coldest month, had a temperature of  $15.42\text{ }^\circ\text{C} \pm 4.93$ . The difference between January and March was determined to be about  $3\text{ }^\circ\text{C}$ . The in-hive temperature then continues to rise from March and reaches its peak in July ( $30.37\text{ }^\circ\text{C} \pm 3.68$ ), after which it begins to decrease again from July to December ( $16.34\text{ }^\circ\text{C} \pm 4.86$ ). There was a  $16\text{ }^\circ\text{C}$  difference between the coldest and warmest month. The temperature at the brood level was always moderate and lower than that inside the hive to guarantee a relatively high relative humidity conducive to the brood development. This clearly shows that bees play an essential role in regulating the exterior conditions to obtain a favorable environment in the brood.

In December, the brood recorded its highest relative humidity ( $82.88\% \pm 14.16$ ). Then, until August, their values decline before gradually increasing once more. At the brood level, July and August recorded the lowest relative humidity levels ( $64.49\%$  and  $63.46\%$ , respectively). This period's high temperatures may be the cause of the resulted low humidity. Although at the brood level there is less nectar available as a water source and a tendency for evaporation due to the high temperature maintained, the relative humidity was always greater than that recorded inside hives (Figure 1). These

results are in agreement with those obtained by [9]. They indicated that bee workers control relative humidity by ensuring: 1) A high brood humidity through controlling evaporation and moisture distribution; and 2) A lower humidity in the remaining hive areas, such as the honey storage cells and the entrance, to dehydrate nectar to honey and prevent microbial growth.



**Figure 1** The effect of month on temperature and relative humidity inside beehives and at the brood level

### 3.5. Heritability estimates

In-hive Temperature, in-hive RH, brood Temperature and brood RH heritability estimates were 0.14, 0.12, 0.16 and 0.28, respectively (Table 4). The investigated traits' medium to high heritability values indicate that they are heritable and could thus be enhanced via genetic selection methods. These results are expected since honey bees engage in thermoregulation by fanning their wings to cool the hive or clustering together to generate heat, which is why the brood temperature is thought to be fascinating. The obtained brood temperature heritability was higher than that of the in-hive temperature. One reason for these results is because the environment inside the hives is significantly impacted by factors that are outside of the beekeepers' control. Moreover, the ability of honey bees to control the temperature inside the hive can be impacted by the ambient temperature as well as the presence of heat sources or cooling systems in the immediate area.

The estimate of heritability for brood relative humidity was higher than that obtained for the in-hive humidity. Sudarsan et al. [23] reported that honeybee workers primarily use an air ventilation system to maintain various humidity levels in the various regions of the hive. This is crucial to not only remove heat that has accumulated inside the hive to prevent overheating but also to lower the levels of CO<sub>2</sub> and water vapor and replace them on a consistent basis with new oxygen.

**Table 4** Estimates of heritability, residual and animal effects of in-hives temperature, in-hives and brood humidity traits

Trait	Heritability	Residual effect	Animal effect
In-hive temperature	0.14	0.69	0.16
In-hive humidity	0.12	0.18	0.70
Brood Temperature	0.16	0.11	0.73
Brood humidity	0.28	0.71	0.01

### 3.6. Genetic and phenotypic correlations

Table (5) displays genetic (upper diagonal) and phenotypic (lower diagonal) correlations between temperature and relative humidity inside hives and at the brood level. The observed genetic correlations ranged from -0.46 to 0.06, with the relative humidity inside hives and the brood relative humidity showing the strongest genetic correlation. The lowest

genetic correlation was observed between the temperature and the relative humidity at the brood level. The obtained positive and weak genetic correlation (0.06), suggests that the two traits are genetically related but not strongly so. It is possible that the same or nearly related genes are responsible for defining these traits given the positive genetic association between them. Favorable phenotypic correlations were found between in-hive and brood temperatures (0.34) and between in-hive and brood relative humidity traits (0.45). The remaining phenotypic associations were all negative indicating that there is little to no systematic association between traits at the phenotypic level. Therefore, changes in one feature may not always predict or account for changes in the other trait.

**Table 5** Genetic (above diagonal) and phenotypic (below diagonal) correlations (with standard deviation in parentheses) for in-hive temperature, in-hive and brood humidity

	In-hive T	In-hive RH	Brood T	Brood RH
In-hive T	1	-0.4	0.051	-0.3
In-hive RH	0.42	1	-0.08	0.06
Brood T	0.34	-0.13	1	-0.46
Brood RH	-0.31	0.45	-0.26	1

### 3.7. Prediction of breeding values

The calculated Breeding Values present a promising avenue for future applications, specifically in the strategic selection of Bee queens ideally suited for sustainable use within the Southern Mediterranean regions. Harnessing the insights derived from Breeding Values, beekeepers and researchers can make informed decisions to identify and propagate colonies with heightened adaptability to the unique environmental challenges prevailing in the Southern Mediterranean context.

By integrating Computed Breeding Values into the selection process, the aim is to refine and elevate the quality of Bee queens. This approach not only contributes to the preservation of biodiversity but also aligns with the broader goal of fostering sustainable beekeeping practices in the face of climatic and ecological variations characteristic of the Southern Mediterranean regions.

## 4. Conclusion

This study has shown that Tunisian bee colonies are able, under Southern Mediterranean climate conditions, to preserve optimal temperature and relative humidity in their hives and broods. The medium to high obtained heritability estimates indicated the possibility of genetic selection to enhance the studied traits. Implementing a recording system, based on sensors, has many promise for improving bee genetics and making well-informed decisions to improve bee management and productivity. It is important to acknowledge the possibility of achieving more accurate genetic evaluations by improving pedigree information and implementing reliable breeding strategies for queens.

## Compliance with ethical standards

### *Disclosure of conflict of interest*

No conflict of interest to be disclosed.

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