

New strategies for controlling heat stress in New Zealand White (NZW) Rabbits in Egypt

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Abstract: The present work was planned to study the reproductive performance of New Zealand White (NZW) rabbits under hot subtropical environmental conditions of Egypt. 36 sexually mature New Zealand White (NZW) rabbits (32 does and four fertile bucks) were all located into equal four groups (eight does and one buck each). The rabbits in the first group (C) were kept as a control. The rabbits in the second group (HB) were supplemented by honey bee (20 ml/L of the drinking water). The rabbits in the third group (FR) were treated by feed restriction. While, The rabbits in the fourth group (BL) were reared under blue light color. Some performance data were calculated throughout the experimental period; The initial and final body weights (BW), feed intake (FI) and water intake (WI), body weight gain (BWG) for adult and young rabbits, feed conversion ratio (FCR) and the mortality rate (MR). Also, some reproductive data were recorded; the conception rate (CR), gestation period (GP), the amount of milk yield for each doe during the first 3 days post kindling, litter size, litter weight, individual kit weight at kindling and weaning and fetal losses. Hematological analysis; Red blood cells (RBCs) count, packed cell volume (PCV), total and differential leukocytic count and hemoglobin were determined by colorimetric method. The respiratory rate and rectal temperature were recorded. The obtained results revealed that, the treated groups recorded the higher values for the most of performance parameters than those that in the control group. The mortality rate was higher in the control group (15%) than that in the treated groups; HB, FR and BL (5, 11 and 8%, respectively). There was a significant ($P < 0.05$) decrease in the reproductive traits of NZW rabbits; conception rate, gestation period, litter size at birth, litter weight at birth and weaning, and three days post kindling estimated milk yield in the control group in comparison with treated groups. The honey bee group represented as the much better group in most of the performance and reproductive traits; conception rate (86%), fetal losses (0.046%), dead kits before weaning (0.81%), Litter weight at birth (320.4g.), Kits body weight at weaning (50.4g), Kits weight gain (240g) and milk yield (140.1g/day), followed by FR and BL treated groups. There was a significant ($P < 0.05$) differences in core temperature ($^{\circ}\text{C}$) and respiratory rate as they were higher in the control group in comparison with treated groups. From the obtained results, it could be concluded that the harm effect of heat stress can be mitigated in reared rabbits during summer season in Egypt by the addition of honeybee in the drinking water or applying 30% feed restriction. But, using blue light color had a little role in the mitigation of heat stress in rabbits, as it has a little positive effect on rabbit's performance and reproductive traits if compared with honey bee and feed restriction.

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1. Introduction

Egypt is located within the humid tropics and is subjected to extended periods of high ambient temperature and humidity. In these regions, rabbits are playing an important role in solving meat production deficiency. But, under the Egyptian summer conditions, rabbit reproductive problems sometimes appeared as a result of heat stress (Marai et al., 2002). Rabbits need to modify their behavior (ingestive and play) and physiological status (respiratory rate and peripheral temperature specially ear one) to maintain their body temperature constant under moderate

thermal stress. In rabbits, most of the sweat glands are not functioning well (Okab et al., 2008) and perspiration (evacuation of water via the skin) is low due to the presence of fur. The only controlled way of latent heat evacuation is done by altering the respiratory rate (Abdel-Samee, 1987). Rabbits are being very sensitive to heat stress which is considered as the most important factor influencing badly on their fertility, reproductive and physiological traits (Askar & Ismail, 2012). Due to their narrow comfort zone (15 to 20 $^{\circ}\text{C}$), rabbits can withstand the cold weather than warm one. The feed intake of rabbit does reared

under high environmental temperature (27-33°C) was 11 to 23% lower than when they were reared at 14-19°C (Mousa-Balabel, 2004).

In the intensive farms, rabbit producers used ad libitum feeding system to get high growth rates and to maximize the performance (Maertens, 2010), but on some farms, feed restriction is often applied to reduce daily changes in feed intake and the possible negative consequences on digestive health and epizootic rabbit enteropathy occurrence (Maertens and Gidenne, 2016). A moderate feed restriction can be used to improve the feed conversion and reduce nitrogen excretion without negative effects on growth (Birolo et al., 2017).

Under heat stress, the pregnant rabbits had a high respiratory rate, water intake and with low feed intake (Abdal-Rahman and Baqey, 2016) and without any effect on daily milk yield during the lactation period (Piles et al., 2012). Survivability and growth performance of the kits up to weaning age depend mainly on their mother's milk production. Therefore, all the factors affecting doe performance during lactation will have a big influence on pre and post weaning growth and survivability of kits. Adding honeybee in the drinking water for heat stressed rabbits expressed a significant increase ($P < 0.05$) in body weight, body weight gain, feed intake, feed conversion ratio, water consumption, conception rate, litter size and weight and milk yield (El-Saidy et al., 2016).

Rabbits are very sensitive to high ambient temperatures since they have few functional sweat glands limiting their ability in eliminating the excess of body heat. Thermal stress affects the animal in different ways, such as reducing the feed intake (Morrow-Tesch et al., 1994), increasing disease susceptibility (Kamwanja et al., 1994), or affecting reproductive efficiency (Marai and Rashwan, 2004). However, stress induced reactions in animals include behavioral modifications aiming at coping towards the stressor. Although these behavioral changes could be more difficult to perform when animals are housed in typical cages of rabbit production than in natural environments.

Blue light can be used as a managerial practice to alleviate the negative effect of heat stress in broiler farms during summer season in Egypt (Mousa-Balabel et al., 2017). However, in our understanding, so far the effect of the blue light color on heat stressed rabbits' production has not been analyzed. According to a free choice test, rabbits show higher preference towards light colors (white and yellow) than for darker colors such as green or blue (Gerencsér et al., 2009). Studying the effect of light colors on the rabbit production can be perspective (Gerencsér et al., 2011). Only limited information can be found

describing this topic for this animal.

Accordingly, the aim of the present study was planned to compare between using blue light, feed restriction and adding honey bee in the drinking water in alleviation of heat stress in rabbit under Egyptian conditions.

2. Materials and Methods

General layout of the experiment: The present study was carried out in the laboratory of the Department of Hygiene and Preventive Medicine, Faculty of Veterinary Medicine, Kafrelshiekh University, Egypt, (from 20th June till 22nd August, 2017) and in accordance with our institutional recommendations of the guidelines of Animal Care and Use of laboratory animals in the research.

Animals, housing and management: a total of 36 sexually mature New Zealand White (NZW) rabbits (32 does with an average age of seven months and nearly similar body weight 2kg and four fertile bucks with an average age of eight months and average body weight of 2.5k g, which were proven to be fertile) were used in this experiment. Rabbits were individually housed in metal hutches of a commercial type (60×55×40 cm) provided with separate and clean facilities for feeding, watering and nest box (40×30×30 cm). Each rabbit was vaccinated by; Cunipravac RHD (inactivated vaccine against hemorrhagic disease, HIPR Acompany) (0.5mlS/C), formalized polyvalent rabbit pasteurellos vaccine (Veterinary Serum and Vaccine Research Institute, Cairo, Egypt) (2ml S/C) and Ivermectin 1% against Mange (Memphis for Pharmaceuticals and Chemical Industries, Egypt) (0.2mlS/C).

The cages were cleaned daily from dropping (urine and feces). All rabbits were kept under identical care and the same hygienic environmental conditions. Under this experiment, the rabbits were fed on a pelleted commercial ration (18% crude protein, 10.19% crude fiber, 2.8% crude fat and 2635 kcal/kg) (Supervisor Company, Egypt). The feed was provided twice daily at 8 a.m. and 5 p.m., while drinking water was provided 3 times/day at 8 a.m., 4 and 12 p.m. The climatic data were recorded during the experimental period according to Marai et al. (2001). The Rabbitry was naturally ventilated through windows and provided with automatically controlled side exhaustion fans.

Experimental design: The rabbits were randomly allocated into equal four groups (eight does and one buck each) and housed in a separate room under the same environmental temperature ranged from 33-36±2°C and relative humidity ranged from 64-79±3% (Mousa-Balabel, 2004). The rabbits in the first group (C) were Kept as a control. The rabbits in the second group (HB) were supplemented by honeybee

(20ml/L of the drinking water) according to **El-Saidy et al. (2016)**. The rabbits in the third group (FR) were treated by 30% feed restriction (150 g/day/rabbit) according to **Oliveira et al. (2012)**. While, The rabbits in the fourth group (BL) were reared under blue light color (**Mousa-Balabel et al., 2017**). All does were identified and bred by natural mating using fertile bucks. Each doe was palpated 10 days post mating and if there was no pregnancy, she rebred until she became pregnant. The nest boxes were provided with a rice straw to get a comfortable warm nest for the kits and applied for kindling on day 28 of pregnancy. The kits were weaned at 4 weeks of age. The light/ dark cycle during the experimental period was 12/12 hours as suggested by **Lebas et al. (1984)**.

Measurements: The initial and final body weights (BW), feed intake (FI) and water intake (WI), body weight gain (BWG) for adult and young rabbits and feed conversion ratio (FCR) were calculated weekly throughout the experimental period (**Marai et al., 2006**). The mortality rate (MR) for kits from birth until weaning was calculated according to **Yassein et al. (2008)**.

The conception rate (CR), gestation period (GP), the amount of milk yield for each doe during the first 3 days post kindling, litter size, litter weight and individual kit weight at kindling and weaning were

recorded according to **Ahmed et al. (2005)**. The respiratory rate and rectal temperature were recorded according to **Marai et al. (1999)**.

Blood samples were collected from three rabbits of each group on a weekly basis, in the morning from the margin of ear vein, into clean test tubes containing EDTA for hematological analysis. Red blood cells (RBCs) count, packed cell volume (PCV), total and differential leukocytic count and hemoglobin were determined by colorimetric method (**Coles, 1986**). Blood samples were centrifuged for 10 min at 3000 rpm after clotting. Serum was aspirated and stored frozen at -20°C until assaying.

Statistical analysis: The obtained data were tested for distribution normality and homogeneity of variance. Data were reported as mean \pm standard error of the mean and analyzed by ANOVA using SAS (Statistical Analysis Software), Institutes INC (**2005**). The significance of difference among the different treatments was evaluated by Tukey's test. The significance level was set at $P < 0.05$.

3. Results and Discussion

Climatic data in table 1 indicated that the rabbits were exposed to severe heat stress during the experimentation as its estimated values exceeded than 25.6°C (**Marai et al., 2000; Abdel-Moneim, 2001**).

Table (1): Weekly maximum and minimum averages of ambient temperature (°C) and RH % during the experimentation.

Weeks	Temperature (°C)		Relative humidity (RH %)	
	Minimum	Maximum	Minimum	Maximum
1	64.2	39.1	34.4	24.3
2	67.3	38.6	34.3	23.6
3	70.2	40.2	34.2	24.2
4	70.2	40.5	35.2	24.5
5	73.5	41.6	35.1	25.1
6	74.2	39.5	34.1	23.8
7	68.5	42.2	34.2	23.4
8	70.3	42.3	34.7	23.2
9	71.6	44.1	35.6	24.1

The performance data of rabbits; final body weight, body weight gain, feed intake, feed conversion ratio and water consumption; are represented in table 2. They were inversely and significantly ($P < 0.05$) affected by high ambient temperature, as it was noticed that the treated groups recorded the higher values for the most of performance parameters than those that in the control group. These results are in agreement with that recorded by **Ayyat et al. (2004)** and **Villalobos et al. (2008)**. However, the addition of honeybee in the drinking water for heat stressed rabbits improved most of the traits concerning growth performance, especially body weight gain

(BWG) and FCR in comparison with other treated groups. These findings are similar to that recorded by **Bonomi et al. (2001)** who observed that the body weight gain improved by about 11% and 15%, while, feed intake improved by 8.5% and 12.5% when a honey bee secretion was used in rabbits feeding at 15 and 20 ppm for 30 to 90 days.

Furthermore, **Han et al. (2009)** mentioned that the BW was increased in piglets with honey bee venom injection. However, feed intake in the case of feed restriction group recorded the higher numerical values if compared with other treated and control groups. These results were in agreement with

(Gidenne et al., 2010; Maertens and Gidenne, 2016) who reported that high digestibility was recorded after using a mild feed restriction diet post weaning in rabbits.

The mortality rate was higher in the control group (15%) than that in the treated groups; HB, FR and BL (5, 11 and 8%, respectively). These results are in the same line of **Marai et al. (2002)** and **Mousa-Balabel (2004)** who reported that rabbits are very sensitive to heat stress since they have few functional

sweat glands, which means they have difficulties in eliminating excess body heat when the environmental temperature is high. **Habeeb et al. (1997)** reported that MR from birth up to weaning was significantly ($P<0.05$) increased in response to an ambient temperature which increased from 19.5 °C in January to 34.8 °C in July. In addition, the same authors estimated MR in adult rabbits in the summer to be 18%, while no mortality was recorded during winter.

Table 2. Performance parameters of New Zealand White rabbits under Egypt summer conditions,

Treatments	C	HB	FR	BL
Initial BW (g)	2.100±0.021 ^a	2.085±0.013 ^a	2.079±0.023 ^a	2.068±0.014 ^a
Final BW (kg)	2.254±0.024 ^a	2.506±0.053 ^b	2.510±0.054 ^b	2.520±0.046 ^b
BWG (g)	154.0±1.804 ^a	421.0±2.125 ^d	431.0±2.108 ^c	452.0±1.801 ^b
FCR	3.210±0.054 ^a	2.655±0.052 ^b	2.100±0.023 ^{ab}	2.000±0.052 ^b
FI (g/day)	48.5±2.013 ^a	115.4±2.512 ^b	130.0±2.652 ^c	96.6±2.534 ^d
WI (ml/day)	378.5±1.439 ^a	295.5±1.700 ^b	285.5±1.266 ^a	288.5±1.384 ^a
Mortality %	15	5	11	8

*Means which superscript with different small letters (a, b, c...) within the same row differ significantly at ($P<0.05$).

There was a significant ($P<0.05$) decrease in the reproductive traits of NZW rabbits; conception rate, gestation period, litter size at birth, litter weight at birth and weaning, and estimated milk yield three days post kindling in the control group in comparison with treated groups as shown in table 3. The results are in agreement with the results of **Mousa-Balabel (2004)**; **Marai et al. (2006)** and **Abdel-Moneim et al. (2013)** who proved that high ambient temperature had a negative effect on appetite and subsequently reduced the feed intake resulting in a slow growth rate and impairment of reproduction in rabbits. These results explained the decrease in the fertility and conception rate under high ambient temperature as a complex set of events was expressed in a significant reduction in total young born and in an increase in the percentage of young born dead (**Matassino et al., 1970**). These findings of lower productive traits of heat stressed NZW rabbits may emphasize the hypothesis of **EI-Masry et al. (1994)** who explained that the live sperm concentration could be decreased under heat stress condition resulting in low conception rate. The decrease in kit's weight at birth and weaning under high ambient temperature may be attributed to, the hyperthermic condition of pregnant does lower the feed intake and thyroid activity and hence, lower the metabolic rate, resulting in a decrease in the litter weight at birth, in addition, such does have a low milk yield resulting in less feed for the growing kits. The

honeybee group represented as the much better group in the most of the reproductive traits; conception rate (86%), fetal losses (0.046%), Litter weight at birth (320.4g.), Kits body weight at weaning (560.4g), Kits weight gain (240g) and milk yield (140.1g/day), followed by FR and BL treated groups (table 3). It explained by honeybee administration possesses better feed utilization, especially starch and mineral (**EI-Nagar et al., 2010**).

Hematological parameters were significant differed by high ambient temperature as shown in table 4, as measured data for RBCs, PCV, Hb, WBCs and neutrophils showed a significant ($P<0.05$) decreases in the control group in comparison with treated groups. On the other side, there was a significant ($P<0.05$) increases in the number of lymphocytes in the heat stressed rabbits. While, basophils didn't show any significant difference by elevated temperature. These results are similar to the recorded by **Ondruska et al. (2011)** who reported that heat stress in mammals decreased the level of ACTH, which might then result in decreases in RBCs count, PCV, and Hb concentration. In addition, the reduction in the PCV during the summer season was attributed to low cellular oxygen, which required for reducing metabolic heat production in order to compensate for the elevated environmental heat load (**Okab and El-Banna, 2003; Okab et al., 2008**).

Table3. Reproductive traits of White New Zealand rabbits under Egypt summer conditions.

Treatments	C	HB	FR	BL
Conception rate %	45	86	50	66
GP (days)	29.2±0.210 ^a	30.3±0.120 ^b	30.2±0.320 ^b	30.6±0.124 ^b
Milk yield (g/day)	95.4±0.214 ^a	140.1±0.261 ^b	132.5±0.230 ^b	106.3±0.265 ^a
Litter size	4.6±0.213 ^a	8.0±0.214 ^b	7.5±0.236 ^b	6.0±0.251 ^a
Fetal losses	0.135±0.011	0.046±0.211	0.500±0.199	0.062±0.210
Litter weight at birth (g)	184.0±1.203 ^a	320.4±1.433 ^b	300.0±1.521 ^b	243.0±1.635 ^{ab}
Kits BW at weaning (g)	300.9±2.210 ^a	560.4±2.255 ^b	490.5±2.213 ^b	414.0±1.985 ^c
Kits BWG (g)	116.9±1.804 ^a	240.0±2.125 ^b	190.5±2.108 ^b	171.0±1.801 ^c

*Means which superscript with different small letters (a, b, c...) within the same row differ significantly at (P<0.05).

Results presented in table 4, showed a significant (P<0.05) differences in core temperature (°C) and respiratory rate were higher in the control group in comparison with treated groups. The results are in agreement with that recorded by **Marai et al. (2001)** and **Marai et al. (2007)** who mentioned the highly significant increase in respiratory rate and temperatures of ear, rectum and skin may be attributed to the exposure of the rabbits to severe heat stress which led to the failure of the physiological mechanism (**Marai et al., 2001**). Also, respiration becomes the main pathway for the latent heat loss,

since most of the sweat glands in rabbits are not functional well and perspiration is not great. So, the panting process increased the respiratory rate and evaporative water loss in rabbits reared under high ambient temperature to enable the animals to dissipate heat by vaporizing high moisture through the respiratory air, which accounts to about 30% of total heat dissipation (**Richards, 1976**). Among the treated groups, honey bee treated group was much better group in improving rectal temperature and respiratory rate followed by FR and blue light color. These findings are coincided with **Marai et al. (1999)**.

Table 4. Hematological parameters, rectal temperature and respiratory rate of New Zealand rabbits White under Egypt summer conditions.

Treatments	C	HB	FR	BL
RBCs (x10 ⁶ /μL)	4.033±0.2404 ^a	7.633±0.6438 ^b	6.267±0.2603 ^b	7.133±0.2333 ^b
Hb (g/dl)	7.933±0.2028 ^a	13.23±0.2728 ^b	10.17±0.2028 ^c	713.40±0.5508 ^b
PCV (%)	28.00±1.528 ^a	38.67±0.8819 ^b	35.33±1.856 ^b	40.33±0.8819 ^b
WBCs (μL)	3821±0.125 ^a	8513±0.319 ^b	7251±0.123 ^c	9652±0.326 ^b
Neutrophils (%)	34.52±0.215 ^a	45.51±0.368 ^b	49.58±0.854 ^{ab}	56.48±0.513 ^b
Lymphocytes (%)	57.64±0.524 ^a	41.63±0.265 ^{ab}	43.12±0.321 ^{ab}	32.124±0.251 ^b
Basophils (%)	4.23±0.312 ^a	4.32±0.521 ^a	4.20±0.241 ^a	4.25±0.125 ^a
Respiratory Rate (cycle/min)	179.6±0.162 ^d	153.4±0.210 ^b	149.3±0.365 ^a	168.2±0.412 ^c
Rectal temperature (oC)	40.5±0.012 ^a	39.8±0.041 ^b	39.6±0.014 ^b	40.1±0.023 ^a
Pulse Rate (beats/min)	122 ±1.012 ^a	148 ±2.31 ^b	140±2.06 ^c	138±2.07 ^c

*Means which superscript with different small letters (a, b, c...) within the same row differ significantly at (P<0.05).

Conclusion

From the obtained results, it could be concluded that the harm effect of heat stress on reared rabbits during summer season in Egypt can be mitigated by the addition of honey bee in the drinking water or using feed restriction, as they have a positive effect on rabbit's performance and reproductive traits. But, blue light had a little role in the mitigation of heat stress in rabbits.

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