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DRINKING MAGNETIZED WATER ALTERS BLOOD CONSTITUENTS, AND STRUCTURE OF SPLEEN AND KIDNEY IN RABBITS

Mohamed I. EL SABRY[1], Azza HASSAN[2], Tarek A. EBEID[3,4], Fatma ABOU-HASHIM[1]

[1]*Animal Production Department, Faculty of Agriculture, Cairo University, 6 El-Gamma Street, 12613, Giza, Egypt*
[2]*Department of Pathology, Faculty of Veterinary Medicine, Cairo University, 4 El-Gamma Street, 12613, Giza, Egypt*
[3]*Department of Animal Production and Breeding, College of Agriculture and Veterinary Medicine, Qassim University, Buraydah 51452, Saudi Arabia*
[4]*Department of Poultry Production, Faculty of Agriculture, Kafrelsheikh University, Kafr El-Sheikh 33516, Egypt*

Emails: m.elsabry@agr.cu.edu.eg; m.elsabry@daad-alumni.de

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ABSTRACT: This study aimed to investigate the effect of magnetic field on physicochemical properties of water and evaluating the effects of the magnetized water (MW) on the productive performance, liver enzymes, and histological structure of spleen and kidney of rabbits. Water samples were collected to determine pH and electrical conductivity (EC), and water structure was investigated by Transmission Electron Microscopy (TEM). Twenty-four weaned Rex rabbits, 21 d old, were allotted into two experimental groups: the first group was assigned as control, received regular tap water (TW), and the treated group, received MW, for 5 weeks. Productive traits were recorded weekly and at the end of the experiment, blood, spleen and kidney samples were collected for examinations. Results showed that pH and EC of MW were higher than those of TW. In addition, the arrangement MW cluster showed a unique alteration on the nano-scale. Growth performance indicators were similar in both experimental treatments, except FCR of the MW rabbits was better than that of the TW rabbits. The MW had no significant effect on plasma concentration of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), and creatinine in growing Rex rabbits received MW in comparison with those received regular TW. Spleen histological structure of rabbits of both groups was normal. However, the epithelial cells lining renal tubules of kidneys of rabbits in MW group appeared large with basophilic nuclei in comparison with TW group. Conclusively, magnetic field altered the physicochemical properties of water. The MW may consequently increase blood glucose level and spleen weight %, enhance kidney structure and improve FCR in growing rabbits. Moreover, more studies are still needed to know the benefits of providing the magnetized water to animals.

Keywords: Body weight; Liver; Magnetized water; Native water; Rabbits; Water quality.

INTRODUCTION

Many countries are facing critical challenges due to adverse impacts of climate change such water stress and the increase in temperature (El Sabry et al., 2023; Ebeid et al., 2023). Water is a vital nutrient in animal feeding. Thus, water quality, including the water content of minerals, could influence the performance of animals and the quality of their products (WHO, 1996; El Sabry et al., 2018; Aggrey et al., 2023).

Magnetized water (MW) application is exposing water to a magnetic field that alters the configuration of water clusters and the physical structure of water salts content (Alabi et al., 2015; Lingdinger 2021; El Sabry et al., 2022). In this context, Lingdinger (2021) suggested that the structured water (SW) cluster has a very structured and hexameric configuration, while the regular water cluster may have 3-dimensional shapes. Also, several studies reported that some physicochemical properties of water are affected by the magnetic field such as average size of water cluster, and induced changes in water pH, EC, and viscosity (Cheng and Weng, 2006; Cai et al. 2009; Alabi et al., 2015; El Sabry et al., 2018). However, the structuring mechanism is still poorly understood. Thus, MW/SW application has been suggested for improving water quality and mitigating salinity problems. It also has improved water quality and wellness, productivity, and reproductivity of animals (El-Hanoun et al., 2013; Zayed et al., 2018; El Sabry et al., 2021). It was reported that MW enhanced growth performance in growing rabbits (Helal et al., 2022), fertility in rabbit bucks (Attia et al., 2015), milk yield in dairy ewes (Shamsaldain and Al-Rawee, 2012), weight gain in geese (El-Hanoun et al., 2017) and egg production and hatchability in turkey (Ebrahim and Azab, 2017). Moreover, Zayed et al. (2018) found that MW had a protective effect on the kidney and maintained its structure in type-2 diabetic rats. On the other hand, other studies showed that MW did not affect the performance and livability of broiler chickens (Alhassani and Amin, 2012) and goats (Sargolzehi et al., 2009).

The abovementioned examples show a kind of contradiction about the effects of the MW application in farm animals' production. In addition, there is a lack of information about the influences of MW on the function and histological structure of organs. Therefore, this study aimed to study the effect of MW on the performance traits, liver and kidney functions, and the spleen and kidney structure in rabbits.

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MATERIALS AND METHODS

In the current study, twenty-four weaned Rex rabbits (three weeks old) of mixed sexes (1 male: 1 female) were used. In a semi-closed house, rabbits were kept individually in galvanized wire cages (40×50×35 cm^3) equipped with feeders and polyvinylchloride water lines. The ambient temperature ranged between 21°C and 26°C. The animals were randomly assigned into two experimental groups (12 rabbits per group): the control group, where rabbits received regular TW, and the treated group, where rabbits received MW. Before, starting the experiment, all rabbits were weighed individually to record the average of initial body weight of the rabbits of the control and treated groups, which were 370 ± 35 g and 368 ± 38 g, respectively. A standard growing diet was provided to rabbits *ad libitum* throughout the experimental period.

Strength and duration of water magnetization

The treated group (MW) received water was magnetized by exposing to 0.3 Tesla magnet apparatus (magnets obtained from K&J Magnetics, Inc., Pipersville, PA, USA) by fixing the magnet apparatus directly to the exterior surface of the polyvinylchloride water line through the experimental period to guarantee the exposure of fresh water to the magnetic field. The control group received regular tap water (Figure 1).

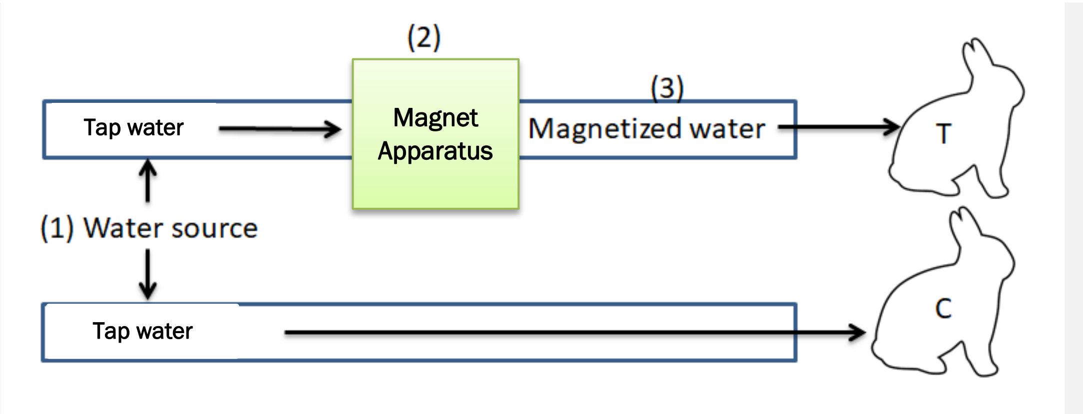

Figure 1 - The water supply for experimental animals showing magnetization process

Physicochemical properties of water

Three samples of water from both treatments were taken weekly to measure the key indicators of water structuring including pH using pH meter (805 MP, FISHER, Germany) and electric conductivity (EC) using a conductivity meter (mS m^{-1}) (WTW LF315 Conductivity Meter, UAS) at room temperature. After five weeks, water samples were also collected to determine the structure of clusters of water molecules using transmission electron microscope (JEOL JEM-1400, USA).

Blood biochemical parameters

Blood samples were collected from the jugular vein during slaughter bleeding into heparinized tubes for biochemical studies. Concentrations of blood plasma glucose (Glu), creatinine (CR), activities of alanine and aspartate aminotransferase (ALT, AST), and alkaline phosphatase (ALP) all were measured using colorimetric methods according to the instructions of the manufacturer.

Productive performance of rabbits

The feed intake and growth rate of rabbits were individually recorded weekly. By the end of the experiment at 56 day of age, rabbits were humanely sacrificed according to the Islamic method to determine the carcass composition. The relative organ weights as a percentage of the live body weight were also determined.

Spleen and kidney histological structure

For the histopathological investigation, five specimens from spleens and kidneys from both groups were taken and fixed in 10 % neutral formalin for 48 h. The tissues were dehydrated in graded concentrations of ethanol and cleared in xylene and then they were embedded in paraffin blocks. The tissues were cut into 5 µm thick sections and stained with hematoxylin and eosin. Five sections per group were examined using CX 31 light microscope (Olympus, Japan).

Statistical analysis

Data were analyzed using the SAS program (Version 9.4, 2013). A T-test was performed, where the magnetic field is the main effect on water pH and EC. Also, data of biochemical parameters and growth performance traits were analyzed using one-way analysis of variance. The following model was used:

$Y_{ij} = \mu + L_i + e_{ij}$

Where: Yij: The j[th] observation within the i[th] line. μ: The overall mean. Li: The effect of the i[th] type of water. Eij: Random error. Standard errors: SE. Means values were separated, when significance existed, using Duncan's multiple range test (Duncan's, 1955). Significance level was set at 5%.

RESULTS

Physicochemical properties of water

The pH value of the MW was higher than (7.20) the pH value of regular TW (6.82) ($P<0.01$). The mean of electrical conductivity (EC) of the MW420.3 mS.m-1 was greater than the EC of regular water 393.5 mS.m-1 ($P<0.01$). The transmission electron microscope of magnetized and regular water drops revealed the differences in the structure of water clusters on the nano-scale, which is suggested to be used as proof of the effect of the magnetic field on the water properties (Figures 2 and 3). The molecules of TW did not exhibit a distinct arrangement (Figure 2a). After passing water through the magnetic field, the molecules of the water droplet had a clear spherical shape border and the overview of the arrangement of the molecules appeared like a tree shape (Figure 2b). In Figure (3) edges of water clusters of magnetized and regular (un-magnetized) water clusters were different, being the edge of the MW cluster was well organized as well as the size of water molecules ranged from 9 to 12.7 nm (Figure 3b). While, the periphery of the TW cluster showed a lack of clear organization, with larger molecules measuring between 29.6 and 58.1 nm present (Figure 3a) (Figure 3a).

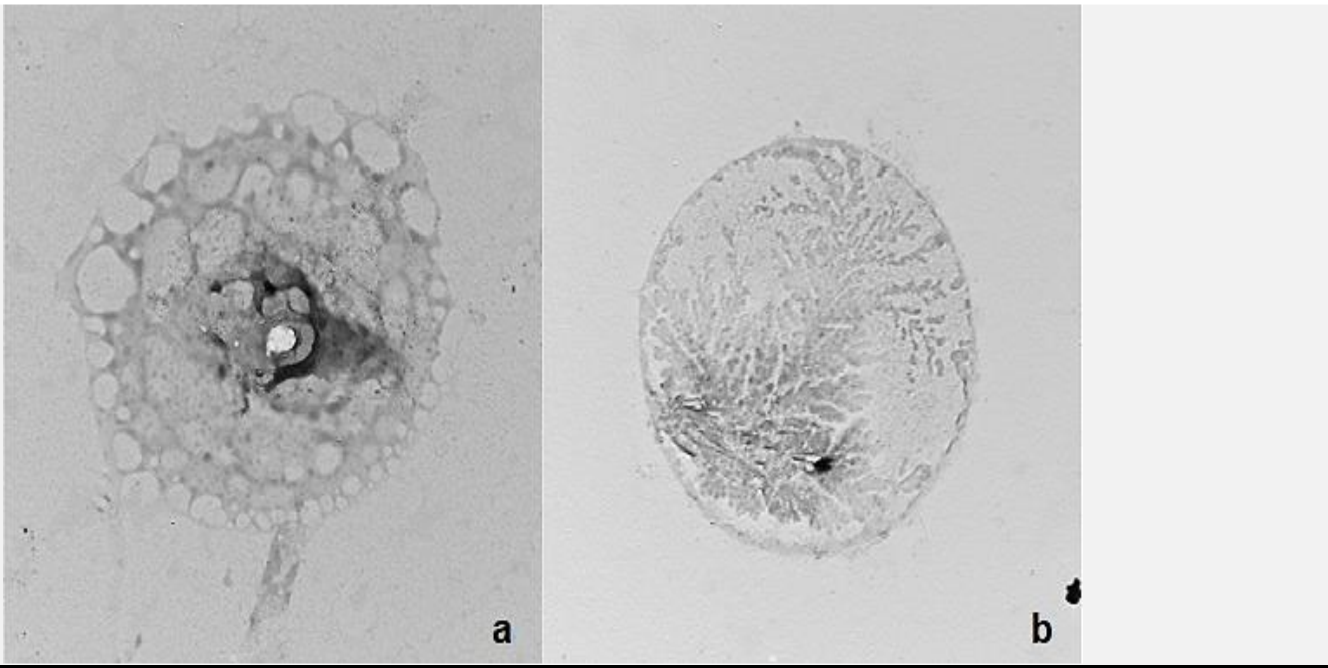

Figure 2 - Arrangement of salts molecules in a) regular water; b) magnetized water molecules arrangement appeared like a tree shape, using transmission electron microscope (20000X).

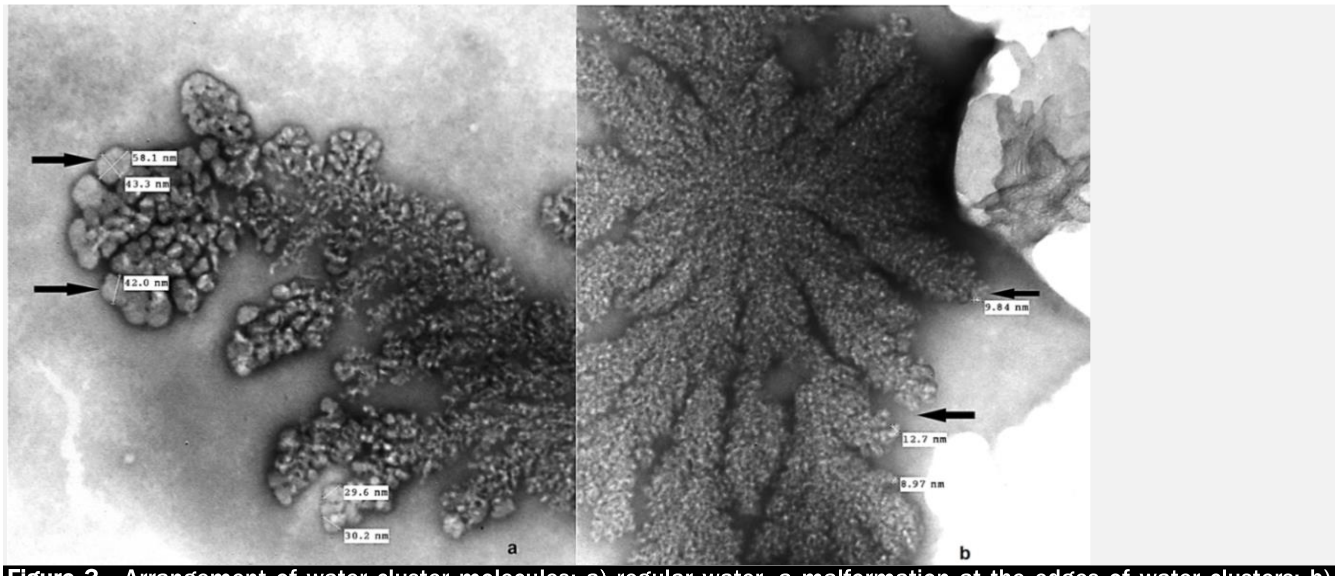

Figure 3 - Arrangement of water cluster molecules: a) regular water, a malformation at the edges of water clusters; b) magnetized water, molecules of water cluster are well structured (Transmission electron microscope, 120000X).

Blood biochemical parameters of growing Rex rabbits

Table 1 shows the biochemical response of growing Rex rabbits. No significant changes were found in plasma content of ALT, AST, ALP, and creatinine with a numerical increase in the group that received MW than those that received regular TW. Only glucose levels were significantly increased in groups that received MW ($P<0.05$).

Productive traits, carcass and organs relative weight

Throughout the experimental period, the growth performance traits of rabbits of MW or TW groups showed insignificant differences ($P>0.05$). At slaughter age (56 d of age), final body weight, feed intake, and fur weight of rabbits received either magnetized or regular drinking water were similar (Table 2). But, feed conversion ratio of the MW rabbits was better than that of the TW rabbits ($P=0.052$) .The relative weight of liver, spleen, and kidney of rabbits received either MW or regular TW were similar and within normal ranges (Table 3).

Table 1 - Biochemical parameters of rabbits received either regular (control) or magnetized water at 56 d of age

Parameter	Control	MW	P-Value
Glucose concentration (mg/dl)	84.25[a] ± 2.61	94.44[b] ± 2.92	<0.05
ALT (U/L)	2.50 ± 0.17	3.00 ± 0.18	NS
AST (U/L)	1.75 ± 0.20	1.38 ± 0.22	NS
Alkaline phosphatase (U/L)	68.25 ± 5.02	66.81 ± 5.61	NS
Creatinine (mg/dl)	0.60 ± 0.04	0.68 ± 0.04	NS

*No significant differences between treatments were observed; ALT: Alanine aminotransferase, AST: Aspartate aminotransferase

Table 2 - Productive traits and relative weight of carcass and organs of rabbits received either regular (control) or magnetized drinking water (MW) at 56 d of age.

Rabbit productive traits	Control	MW	P-Value
Initial body weight (g)	370 ± 35	368 ± 38	0.70
Final body weight (g)	1584 ± 37	1677 ± 38	0.20
Total feed intake (g)	3551 ± 83	3507 ± 89	0.72
Feed Conversion Ratio	2.92 ± 0.05	2.67 ± 0.05	0.052

Means ± SE, within row, with different superscripts are significantly different. (P<0.05)

Table 3 - Carcass and organs weight % of rabbits received regular (control) or magnetized water at 56 d of age

Water source / Traits	Control	Magnetized	P-value
Fasting weight (g)	1472	1629	NS
Empty carcass (g)	974	1004	NS
Carcass yield, %	60.7 ± 0.9	61.6 ± 0.8	NS
Spleen weight, g	1.04	1.07	NS
Heart weight, g	5.04	5.65	NS
Kidney weight, g	12.02	12.07	NS
liver weight, g	51.27	49.07	NS
Fur weight, g	148 ± 5	146 ± 5	NS
Relative weight of organs, %			
Liver	3.19 ± 0.16	3.08 ± 0.15	NS
Spleen	0.06 ± 0.004	0.06 ± 0.004	NS
Kidney	0.74 ± 0.026	0.73 ± 0.024	NS

*Means ± SE, within row, with different superscripts are significantly different. (P<0.05)

Alteration in spleen and kidney histological structure

The spleen histological structure of rabbits received TW showed normal histological structure with normal red and white pulp (Figure 3a,b). Similarly, spleen of rabbits received MW appeared normal in most examined sections with normal red and white pulp. But, in examined sections, the white pulp was slightly expanded with the increased number of lymphocytes and reticular cells. Kidneys of rabbits received TW showed normal renal tubules with normal epithelial lining and glomeruli (Figure 4a). On the other hand, kidney of rabbits received MW showed normal glomeruli and renal tubular epithelium, however, the epithelial cells lining some renal tubules appeared large with basophilic nuclei (Figure 4b).

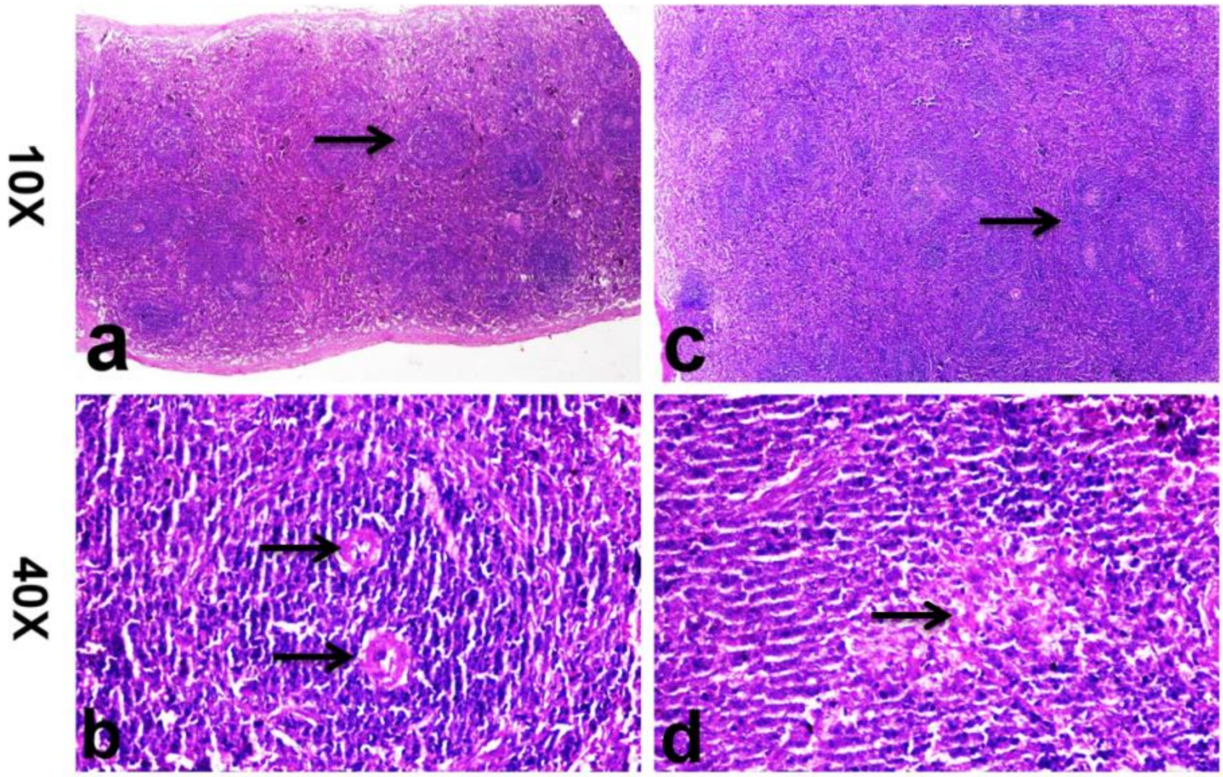

Figure 4 - Spleen (a and b) of rabbits received regular drinking water showing normal histological structure with normal red and white pulps (H&E,10X for a and H&E,40X for b). 4 c and d: spleen of rabbits received magnetized drinking water showing slightly expanded white pulp (H&E,10X for c) with increased number of reticular cells (H&E, 40X for d).

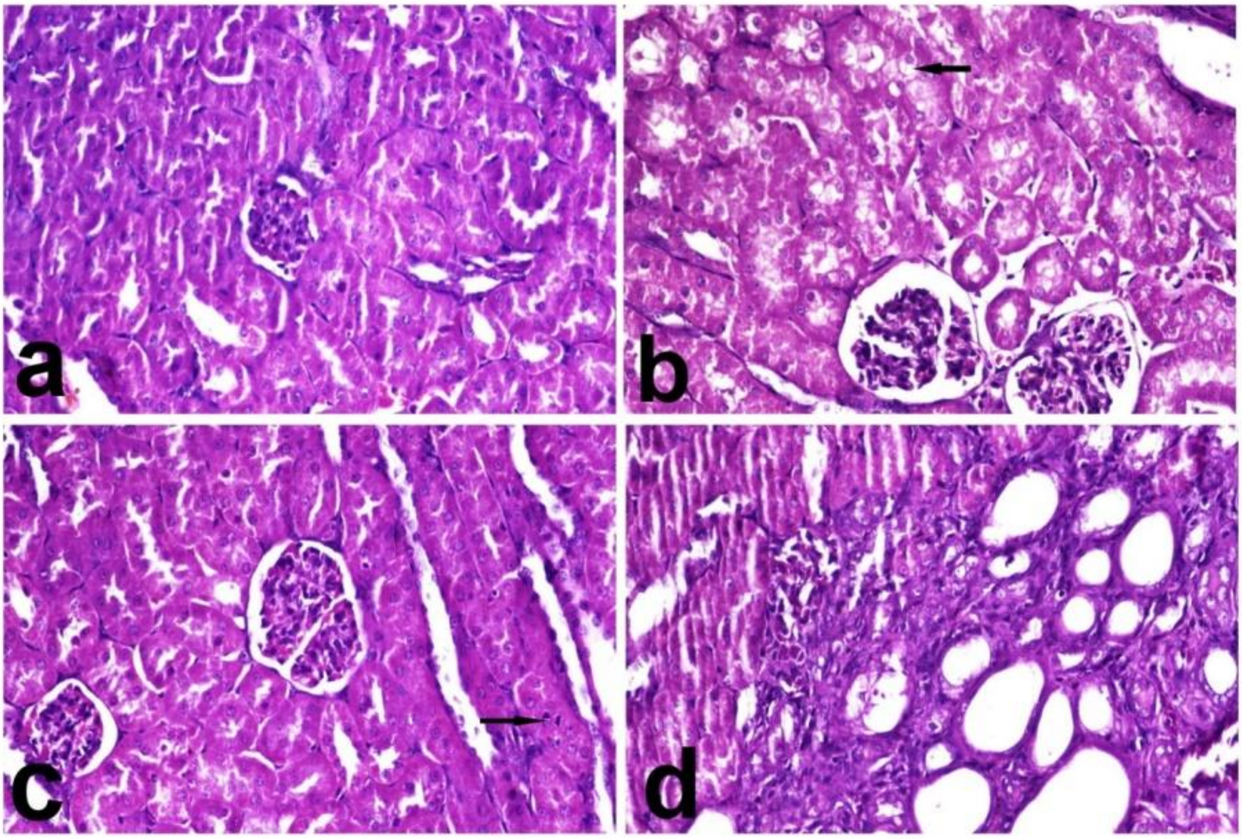

Figure 5 - Kidneys of (a) rabbits received normal drinking water showing normal renal tubules and glomeruli, (b, c & d) rabbits received MW showing mild degeneration of individual epithelial cells lining some renal tubules (arrows) (b), mitotic figure (arrow) (c) and presence of regenerative renal tubules characterized by basophilic cytoplasm and large vesicular nuclei (d), (H&E, 40X).

DISCUSSION

Physicochemical changes in water properties

Diamagnetic materials such as water and organic materials have a weak response to a magnetic field, which can be altered due to the magnetic field and the period of exposure to it (Alabi et al., 2015). When water is subjected to the magnetic field, the frequency of collision between ions was increased, and ions' configuration could be changed (Cai et al., 2009; Alabi et al., 2015). Moreover, Cho and Lee (2005), Xiao and Miwa (2017) mentioned that the magnetic treatment of water decreased water tension, altered magnetic spin H+, and weakened H-bonds. Subsequently, these alterations in chemical and physical properties of water are followed by more changes in the pH, electrical conductivity, and molecular energy of magnetically treated water (Parsons et al., 1997; Cai et al., 2009; Wang et al., 2018; El Sabry et al., 2018). Ebrahim and Azab (2017) suggested that treating water with a magnetic field improved the water molecule structure, which enhances the overall reactions of the MW. Although, the pH and EC values of MW were higher than those of TW, values were laid within the accepted pH value (6.5-8) and very satisfactory limits of EC (1.5- 5 ds.m-1) for poultry species (Ayers and Westcot, 1994; WHO, 1996). Also, Mahmoud, et al. (2015) elucidated that magnetic treatment induced greater effect on magnetic treated water in terms of pH, conductivity, salinity, and dissolved oxygen.

The transmission electron microscope was used to show if there are changes in the physicochemical properties of water due to exposure to the magnetic field. The current finding indicated changes in the physical feature of MW cultures at the nano-scale, which consequently might differently influence the digestive and absorptive processes in animals.

Changes in water properties might be attributed to one of the following explanations: 1) Cai et al. (2009) suggested that proton transfer in the closed hydrogen-bonded chains causes the magnetization of water, and 2) Xiao and Miwa (2017) mentioned that the magnetic treatment of water decreased water tension, altered magnetic spin H+, and possess weaker H-bonds. Therefore, it might be indicated that there is a potential to integrate magnet technology in manipulating some water properties, especially where water resources are limited. However, further studies are still needed for a better understanding the mode of action of magnetization on water.

Blood biochemical parameters

Water stress, high temperature, high feeding costs, and lack of proper management are the main reasons for the decline in the productivity of poultry and livestock (Szendrő et al., 2012; El Sabry et al., 2021; Abbas et al., 2022; El Sabry and Almasri, 2023). Under these circumstances, rabbits can be a potential source of animal protein due to their limited water requirement compared to livestock, ability to adapt to a variety of feed, and capacity to regulate their body temperature over a wide range of temperatures (Nistor et al., 2013; El Sabry et al., 2021).

As shown in Table 3, there is a numerical increase in plasma content of ALT, AST, ALP, and creatinine due to drinking of MW in comparison with TP. A mild increase in AST and ALT in the blood of rabbits received MW may show a positive effect on liver function. On the other hand, Helal et al. (2022) elucidated that MW reduced plasma concentrations of ALP, AST, ALT gamma-glutamyltransferase (GGT), total bilirubin, blood urea nitrogen and creatinine in growing rabbits.

The improvement in plasma glucose concentration might be attributed to enhancing the metabolism and nutrient transfer to various body cells. Rabbits that drank MW showed a significant increase in glucose concentration compared to those of the regular water group (Table 3). Similarly, Helal et al. (2022) postulated that MW increased plasma glucose concentration in growing rabbits. Also, Mahmoud et al. (2015) pointed out that administration of MA increased serum glucose concentration in growing black Balady rabbits. This improvement might be attributed to high conductivity MW, which may increase blood circulation and increase glucose uptake by the cells (Bonhomme-Faive et al., 1998; High et al., 2000; Alhammer et al., 2013).

Productive performance

Since water is the most important nutrient for farm animals, it is plausible that offering them MW may influence the productivity of the animal and/or its organ's characteristics. The current study's results showed no significant effect on the productive traits; final body weight, feed intake, fur weight, and carcass % through the experimental time. Nevertheless, it appears that all findings favored the MW group. These results could be due to providing the MW for a short period or the power of the magnet should be altered to achieve the desired effect on the studied traits. These results are in correspondence with Mahmoud et al. (2015) who demonstrated that drinking MW had no significant effect on feed intake, final body weight and weight gain in growing rabbits. In broiler chicks, similar findings were reported by Alhassani and Amin (2012), who found that there is no significant difference in the growth performance of broilers received MW or regular water.

In this study, better feed conversion ratio of MW group could be improved due to the new physiochemical properties of water that might enhance the availability of nutrients and the absorption process in rabbits. In this context, Helal et al. (2022) reported that average daily gain, growth rate and feed conversion ratio were elevated; however, feed intake was reduced due to drinking MW in comparison with non-magnetized water in growing rabbits. Also, El-Hanoun et al. (2013) found that the growth rate of offspring born to does that consumed MW during pregnancy had a greater growth rate than those born to does that consumed regular tap water. This discrepancy in the results of growth performance might be due to the variation in physicochemical characteristics of MW and age and strain of rabbits.

Organs relative weight

Oliveira et al. (2013) and El Sabry et al. (2015) reported changes in organs' weight due to the exposure of the animal to improper environment or stress. For example, lighter spleen weight % was recorded due to exposing birds to uneven lighting programs (El Sabry et al., 2015). Since, rabbits' relative weight of both groups was within normal range, it could be speculated that the MW did not present a kind of stress on rabbits.

Alteration in spleen and kidney histological structures

The spleen is one of the main lymphoid organs that comprise two main compartments, which are white and red pulps. Functionally, white pulp plays an essential role in the immune response, while red pulp filters the blood from the old and damaged red blood cells (Mebius and Kraal, 2005; El Sabry et al., 2015). Therefore, it might be suggested that MW had no detrimental effect on the spleen histological structure. In this context, the slight expansion in white pulp might refer to an improvement in the immune status of rabbits that received MW. According to our knowledge, no studies in rabbits were conducted on the effect of MW on spleen histological structure. In rats, Al Saffar et al. (2013) indicated that rats treated with 250 gauss intensity of MW exhibited hyperplasia of the white pulp in their spleen tissue, whereas rats treated with 750 gauss intensity of MA displayed a notable hyperplasia of the lymphoid tissue in the periarterial sheath.

There are contradictions in the available information about the influence of MW on kidneys' histology due to the limited number of investigations on farm and laboratory animals. In addition, Khater and Ibraheim (2016) reported that water subjected to extremely high-power magnets (1.8 Tesla) could negatively affect the histological structure of the kidney in rats. Whereas, Zayed et al. (2018) found that the provision of MW (600 Gauss) protected the kidneys of type 2 diabetic rats from nephrotoxic damage by reducing hyperlipidemia, oxidative stress, uremia, and renal dysfunction. Ebrahim and Azab (2017) mentioned that MW could be a factor in breaking up kidney stones in humans. Sargolzehi et al. (2009) reported that MW did not influence urea and Na+, K+, Mg++, and P- in the blood of goats.

As shown in Fig (4a), kidneys of rabbits that received TW showed normal renal tubules with normal epithelial lining and glomeruli. These results are in harmony with Zayed et al. (2018) who noted that MW treatment prevented type 2 diabetic rat kidneys from nephrotoxic damage by reducing hyperlipidemia, uremia, oxidative stress, and renal dysfunction in rats. In the present study, mild hyperplasia of the renal tubular epithelium of some renal tubules in the kidney of rabbits that received MW may be due to the increase in the blood glucose level. Also, Thomson et al. (2001), Henegar et al. (2001) and Tobar et al. (2013) showed that high blood glucose level could induce glomerular enlargement is associated with hypertrophy of tubules. Moreover, enlargement of renal tubules is not necessarily a bad sign (Seely et al., 2018), and a few basophilic tubules are a normal feature in young rats (Frazier et al., 2012).

CONCLUSION

Water exposure to magnetic field alters physicochemical properties of water. The MW improves the FCR increases the relative weight of spleen. The positive changes in the liver and kidney structures of MW rabbits may indicate a positive influence on the animal's health. Moreover, more studies are still needed to determine the benefits of providing MW to animals.

DECLARATIONS

Corresponding author

Correspondence and requests for materials should be addressed to Mohamed I. El Sabry; E-mails: m.elsabry@agr.cu.edu.eg; m.elsabry@daad-alumni.de; ORCID: https://orcid.org/0000-0001-6855-0111

Ethical Approval

This study was approved by the Institutional Animal Care and Use Committee of Cairo University, protocol No. (CU-II-F-37-17).

Author contribution

M.I. El Sabry, Fatma Abou-Hashim: paticpated in experimental design, collecting data, data analysis, and writing of the original manuscript. M.I. El Sabry, Tarek A. Ebeid: editing the final version. Azza Hassan: participated in histological investigation.

Data availability

Upon request.

Consent to participate

Consent was obtained from all authors.

Consent for publication

All participants have consented to the submission of the review article to the journal.

Competing interests

The authors declare no competing interests in this research and publication.

Funding declaration

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