

Journal of Animal Science, 2021, Vol. 99, No. 5, 1–11

doi:10.1093/jas/skab063 Advance Access publication February 26, 2021 Received: 4 December 2020 and Accepted: 23 February 2021 Animal Health and Well Being

ANIMAL HEALTH AND WELL BEING Structured water: effects on animals

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Abstract

This review focuses on the effects of structured water (SW) on animals when it is consumed on a daily basis. SW is liquid water that is given altered H-bonding structure by treatment with various forms of energy including magnetic fields and light. While most of the research has been conducted on 'magnetized' water, which has structure of short duration, recent research has examined effects of a SW with stability of at least 3.5 mo. A variety of laboratory and farm animals have been studied over the past 20 yr. Consistent (3 or more studies) responses among animals consuming SW for 1 mo or more include increased rate of growth, reduced markers of oxidative stress, improved glycemic and insulinemic responses in diabetics, improved blood lipid profile, improved semen and spermatozoa quality, and increased tissue conductivity as measured using bioelectrical impedance analysis. While it is known that fluids in and around cells and molecules are structured, it remains unknown if this endogenous water structuring is influenced by drinking SWs. The mechanisms by which SW affects biological systems are unknown and require investigation. Effects of SW, when taken up by biological systems, are likely associated with altered water structuring around biological surfaces, such as proteins and membranes.

Key words: antioxidant effects, antioxidant status, clustered water, magnetized water, production animals, reproduction

Introduction

Only in the past 2 decades has it been consistently shown that structured water (SW), when used as a nutrient source for animals, results in many positive responses including increased growth and productivity in agricultural settings (Ebrahim and Azab, 2017). Water is an essential nutrient and also the most abundant molecule in biological systems; when water is not in adequate supply or of adequate quality organisms do not thrive and may die. While the importance of water in biology may be taken for granted (Warner, 1970), it is universally recognized that water is needed for hydration and for optimizing health and performance (Bondy and Campbell, 2018). The importance of water in general, and SW in particular, remains underestimated and underappreciated in biology—the science of life. It is important that biologists are well trained in the fundamental principles underlying the physics and chemistry of water, whether it is in a beaker or in living systems. SW is very complex and poorly understood and, accordingly, the topic of SW and its effects on biological systems has been largely ignored.

In order to begin the story of SW 2 unrelated discoveries need to be mentioned. The first is that water treated with electromagnetic radiation gains structure (Del Giudice et al., 1988); see Pang 2014 for extensive review. The second is that the consumption of "magnetized" water was shown to have positive effects on animals, including those in agricultural production systems (Patterson and Chestnutt, 1994; Ebrahim and Azab, 2017). A theme of the SW research in agricultural settings continues to be improvement of water quality (Goldsworthy et al., 1999) while simultaneously improving animal wellness and productivity (Ebrahim and Azab 2017; Gilani et al. 2017).

Let is consider what is meant by "water gains structure". As depicted in Figure 1, SW may be defined as liquid water that

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Abbreviations

| ALP | alkaline phosphatase |
|--------|--------------------------------|
| ALT | alanine aminotransferase |
| AST | aspartate aminotransferase |
| DMI | dry matter intake |
| HDL | high-density lipoproteins |
| IgA | immunoglobulin A |
| IgG | immunoglobulin G |
| IgM | immunoglobulin M |
| LDL | low-density lipoproteins |
| RBC | red blood cells |
| SW | structured water |
| TBARS | thiobarbituric acid substances |
| TG | triglyceride |
| UV-vis | ultraviolet-visible |
| VLDL | very low density lipoproteins |
| WBC | while blood cells |

has gained structure compared with unstructured liquid water (often referred to as bulk water). Structure refers to increases in the numbers of aggregated hydrogen and oxygen atoms that form clusters comprising 2 to hundreds of water molecules (Del Giudice et al., 1988; Chaplin, 2000; Pang, 2014; Chibowski and Szcześ, 2018), as exemplified in Figure 2. The clusters are capable of getting very large and may have 3-dimensional shapes that are spherical (Pang 2014), helical (Lo et al., 2012; Ho, 2014; Elia et al., 2017), and planar (Hwang et al., 2018; Elton et al., 2020) as shown in Figure 3. It is important to point out that the structures are unlike those found in ice, which has a very structured, hexameric configuration (Figure 4). The mechanisms by which water becomes structured remain poorly understood (Chaplin, 2000; Pang, 2014; Ball, 2017; Chibowski and Szcześ, 2018; Elton et al., 2020). Yet, the present review reports that water structured in different ways are capable of having many and profound



Figure 1. A glass of water comprises billions of H_2O molecules (top) that are capable of accepting/releasing protons (H⁻) up to 1 million times/s (Edsall and Wyman, 1958; Harned and Owen, 1958). However, protons tend to not exist on their own in aqueous solutions and readily bind to H_2O to form H_3O^{-} (hydronium). In theory, 2 H_2O molecules can form 1 hydronium and 1 hydroxyl.

effects on the biological systems that come into direct contact with these waters.

Most of what we know about SW comes from research where water has been structured using energy from magnetic fields (Pang and Deng, 2008; Pang et al., 2012; Pang, 2014; Chibowski and Szcześ, 2018). It is these types of "magnetized" waters that have been studied for their effects on plants and animals, and particularly with an agricultural focus (Ebrahim and Azab, 2017). While some of the published literature in this area is scientifically weak, the consistency among the publications permits one to draw conclusions and formulate designs for future research. The main purpose of this review is to summarize what is currently known about the effects of SW on animals. In so doing, an aim is to highlight the types of research that need to be undertaken in order to understand how biological effects occur.



Figure 2. Examples of how individual H₂O may combine into linear or nonlinear clusters. Water molecules are connected by hydrogen bonds as depicted by black dashes. Adapted from Pang (2014).



Figure 3. Networked hydrogen bonding (black dashes) of water molecules into an hexameric-shaped water cluster. Adapted from Pang (2014).



https://commons.wikimedia.org/wiki/File:Ice_XI_View_along_c_axis.png

Figure 4. Ordered, hexameric structure of ice.

Structuring Water Alters the Physico-Chemical Properties of the Water

Chibowski and Szcześ (2018) concluded from their detailed review of the literature on magnetized water that the results of experiments performed over the past 20 yr do not provide a consistent mechanism that accounts for effects of magnet treatments on altering the physico-chemical properties of water, including water clustering and water-ion interactions. This was echoed in a recent review by Elton et al. (2020) on planar forms of structured, mostly interfacial, water. Chibowski and Szcześ (2018) stated that research conducted in the past decade strongly implicates changes in the structure of water via hydrogen bonding in intraclusters and between interclusters. According to one theory, upon exposure to a magnetic field clusters can be transformed in size, such that water intercluster bonds are weakened while intraclusters bonds are strengthened, with a slight increase in the amount of the bonding (Chang and Weng, 2006).

That the structuring of water has been altered is known from scans made by measuring the absorption of different visible and ultraviolet light wavelengths by water, changes in the absorption of the infrared spectrum, changes in the absorption of Raman spectra as well as other approaches (Pang and Deng, 2008, 2009; Slavchev et al., 2015; Tsenkova et al., 2018). The increase in structuring, as evidenced from the increase in infrared absorption, is proportional to both the magnetic field intensity and the duration of treatment (Figure 5). Upon removal of the magnetic field, the effects are shortlasting and, in all of the experiments of Pang and Deng (2008) represented by Figure 5, infrared absorbance had returned to baseline by 60 min after removal of the water from the magnetic field.

Prior to the work of Pang and colleagues, it was known that magnetically treating water altered the physical properties of the water and that the altered physical properties were due to "changed dimensions of water clusters" (Baranov et al., 1995; Ibrahim, 2006). Table 1 highlights some of the changes in



Figure 5. Representation of the effects of treating water with different magnetic field intensities for different durations. G, Gauss; 1 T, 10,000 G. Adapted from Pang and Deng (2008).

physical properties that can occur when treating water with magnetic fields. Several researchers have reported increases in conductivity and pH while there are decreases in density and surface tension. In addition to these, there are reports of increased dielectric constant (Ibrahim, 2006; Pang and Deng, 2008), an increase in vaporization enthalpy (Toledo et al., 2008).

While it is important to understand the changes in physical properties that occur with pure water, from an applications point of view it is also important to understand what occurs when tap water or ground (well) water is used. Tap water, ground water, lake water, and reservoir water are universally used in plant and animal agricultural. These types of water vary with respect to mineral and organic material contents and exposure to effects (light, heat, and mechanical disturbances). Each of these influences appears to affect the magnitude, type of structuring, and stability of structuring that may occur (Chibowski and Szcześ, 2018). Presently, in the peer-reviewed scientific literature, there is evidence for 2 main

| Table | 1. | Some | of | the | physical | properties | of | untreated | and |
|---|----|------|----|-----|----------|------------|----|-----------|-----|
| magnetically treated tap water ¹ | | | | | | | | | |

| Parameter | Untreated tap water | 500 G-treated | 1000 G-treated |
|---|------------------------|-------------------|---------------------|
| Conductivity, mS/cm | 650 ± 8.1 | 655.0 ± 8.6 | 710.0 ± 8.9* |
| рН | 7.60 ± 0.07 | 7.62 ± 0.05 | $7.85 \pm 0.02^{*}$ |
| Density, mN/mL | 50.1 ± 2.25 | $40.0 \pm 2.01^*$ | $40.0 \pm 2.12^{*}$ |
| Surface tension, dyn/cm ² | 60.5 ± 2.8 | 52.4 ± 2.9* | 50.4 ± 2.9* |

¹Values are mean ± SEM.

*Significantly different from untreated tap water. Data from Al-Hilali (2018).

types of SW based on stability. Water that is intentionally structured by treating with magnets, either statically or with flow-through systems, is stable for relatively short periods of time, typically hours and not more than 3 d. The degree of structuring is proportional to the intensity and duration of the imposed energy, whether it be magnetic or light and the decay of structuring is approximately exponential in time course (Pang, et al., 2012; Chibowski and Szcześ 2018). The second type of SW has long-term stability (months).

Nearly 20 yr ago, a SW with months-long stability was developed by treating purified water with ~0.1% by weight of selected minerals that include potassium and silica, magnetic energy and light energy (Lorenzen, 1988, 2000). This water has been tested in biological systems, including humans (Ling et al., 2004; Wang et al., 2004; Chen et al., 2005). More recently, this water was used in a clinical field trial of Thoroughbred race horses in training (Lindinger and Northrop, 2020). This highly stable SW has been analyzed using the aquaphotomics approach developed by Dr. Roumiana Tsenkova at Kobe University in Japan (Slavchev et al., 2015; Tsenkova et al., 2018; Kraats et al., 2019). When analyzed up to 3.5 mo after date of manufacture, this SW was found to retain significant structuring, even after boiling in a microwave for 5 min. Infrared spectroscopy and aquaphotomics analysis determined structures in this liquid SW to include protonated water clusters, hydrated water, and water dimers (Lindinger and Northrop, 2020). This SW is available commercially as Defiance Fuel (Defiance Brands Inc., Nashville, TN).

Effects of SW on Animal Growth and Development

As detailed in the following paragraphs, there are more than 2 dozen studies that have reported beneficial effects of drinking or using SW on animals, including 2 studies that examined effects on oral health in children. The other species include horses, cattle, fish, sheep, goats, mice, rats, rabbits, Japanese quail, ducks, and chickens. A few studies have reported adverse effects (see below), and these appear to occur with waters that have been treated for a prolonged duration with magnets or with too great a magnetic field. A few studies have examined the effects of duration of "magnetization" or of magnetic field strength on biological effects and is evident from these that there is a "dose-response" effect. Relatively, few studies have reported key indicators of water structuring such as electrical conductivity, pH, infrared spectrum, UV-vis spectrum, and surface tension-this is a limitation of most studies to date.

Cattle

Patterson and Chestnutt (1994) cited 3 animal research reports in which SW was used—these studies are no longer readily available and commentary is taken from Patterson and Chestnutt as well as others (El-Hanoun et al., 2013; Balieiro Neto et al., 2017). Increased growth has been claimed in calves and sheep, with a reduction in fat in carcasses (Lin and Yotvat, 1988). A 75-d study using Jersey cows drinking water treated with a static 324,000 G magnetic field reported a significant increase in subcutaneous fat thickness as measured using ultrasound (Balieiro Neto et al., 2013).

Sheep and Goats

The first reasonably well-designed experimental research study investigating effects of SW on animals appears to be that of Patterson and Chestnutt (1994). The authors magnetized local tap water or ground water which was "hard," i.e., 236 and 332 mg/L total solids (in Hillsborough, Northern Ireland, UK) and provided the control water or SW to groups of lambs (n = 10/group) 1 wk after weaning. The intensity of, and duration of exposure to, the magnetic field was not provided. The effects of water treatment on water physical or chemical properties was not determined. Lambs continued with the treatment until they reached a live weight of 54 kg (~40 to 80 d after weaning). The data indicate that the study was underpowered: there were tendencies (P > 0.05 and < 0.10) for water intake to be lower with SW and for an increased feed conversion ratio. There were no effects on growth performance or carcass composition. The authors concluded that the more intense the water treatment, the greater the tendency for adverse effects on lamb performance. In contrast, Shamsaldain and Al Rawee (2012) reported significantly increased weight of lambs and ewes when ewes consumed magnetized water (1,000 G) compared with control, but this study also did not report water properties.

Yacout et al. (2015) studied goat bucks and lactating does that consumed magnetized waters (1,200 and 3,600 G) for 60 d. Consuming the 3,600 G water, compared with control and 1,200 G water, resulted in significantly increased dry matter intake (DMI)—with water consumption matched to DMI and increased digestibility with both magnetic waters compared with control. This was associated with increased rumen microbial population, reduced ruminal ammonia production, with markedly increased volatile fatty acid concentrations and decreased methane production.

Rodents

Adult mice were given tap water treated with 1,000 G or 2,000 G magnets or consumed normal tap water (controls; Alhammer et al., 2013). The magnetic treatment reduced the water density (~10%), increased total dissolved solids at 2,000 G and was without effect on electrical conductivity, dissolved oxygen, pH, and salinity. The duration of the experiment was not provided but appears to have been 4 wk. There was no effect of drinking 1,000 G water on body weight, while body weights were significantly lower (~13%) for mice drinking the 2,000 G water. There was no effect on feed consumption, although water consumption was increased by 40% to 50% in both groups drinking SW. The results of this study suggest a possible dose–response effect.

In the study by Lee and Kang (2013), SW by passing it through a magnetic field of 9,000 to 13,000 G and the water was

consumed within 1 d. The authors studied a group of control rats and a group of rats with streptozotocin-induced type 2 diabetes, with the diabetic rats further split into 2 groups, 1 consuming control water and the other SW over a period of 8 wk. There was no control group of rats that consumed SW. The diabetic rats showed a >60% reduction in weight gain compared with control rats despite a 15% increase in daily food intake and 2.5-fold greater daily water intake compared with controls. Diabetic rats drinking SW gained ~30% less weight than diabetic rats drinking control water, and daily food intake between the 2 diabetic groups was similar. These results imply that consuming SW may result in an increased metabolic rate and energy expenditure. The results of this study also demonstrated a negative effect when using water treated with the relatively high magnetic field when consumed over an 8-wk period.

Balieiro Neto et al. (2014, 2017) conditioned the drinking water of rats by using a magnetic monopole field of 32,000 G and studied a variety of parameters at 15, 30, and 45 d. Compared to control water, the magnetically treated water had an increased pH and reduced turbidity—other physical parameters were not assessed. Over the 45 d, the authors reported a significant 25% reduction in daily weight gain which was associated with increased mass-specific dietary nitrogen retention. There were no effects on DMI, water intake, urine output, fecal nitrogen, and urine nitrogen. This study, similar to that of (Lee and Kang, 2013), also suggested that 32,000 G field strength was excessive.

Rabbits

El-Hanoun et al. (2013) provided tap water and well water either untreated or treated with a 4,000 G magnetic field to rabbit does (n = 10 per group; aged 6 to 7 mo) for 28 wk, with 12 wk prior to mating and 12 wk postgestation (time of weaning). Magnetic treatment of both tap and well water resulted in increased pH, salinity, electrical conductivity, reduced organic matter, and reduced hardness of well water. There were no effects on dissolved oxygen and individual ion concentrations (Na⁺, K⁺, Ca²⁺, Mg²⁺, Cl⁻, CO₂²⁻, and HCO₂⁻). The growth performance of litters was followed from 6 to 12 wk postparturition and prior to weaning (n = 143 to 266 per group). Does drinking SW, compared to tap water or well water controls, for 12 wk had significant 2-fold greater weight gains at mating and at 7 d after mating, accompanied by increased daily feed intake. Litter size and litter weight at birth and at 28 d were significantly increased for does that consumed SW. The body weight gain of offspring of does drinking SW from tap water, from 6 to 12 wk postparturition, was 9.5% greater than that offspring of does drinking control tap water, and this was 3-fold greater in magnitude than the gains seen with offspring of does drinking SW from well water. Increased weight gain was accompanied by significant decreases in feed intake, feed conversion ratio (g feed/g weight gain) and mortality. This study demonstrated that water treated with a magnetic field of 4,000 G was beneficial, and no adverse effects were reported.

Attia et al. (2015) provided tap water and well water treated at a magnetic field strength of 4,000 G and provided as drinking water to 7.5-mo-old-rabbit bucks (n = 10 per group, 4 groups) for 28 wk. The magnetic treatment increased pH, conductivity, salinity, dissolved oxygen of both tap, and well water and reduced the hardness of well water. Compared to control tap or control well water, both SWs resulted in significantly increased body weight gain (23% for tap water; 84% for well water) which was associated with increased daily feed intake. The larger relative effect for the well water was because untreated well water had negative effects on growth (weight gain) that were substantially mitigated when magnetically treated water was used. Thus, magnetic treatment of the well water completely reversed the negative effects of the hard well water on growth and performance.

Rabbit does aged 7 to 8 mo were given normal tap water or magnetized tap water treated with 1,200 G or 3,600 G for 30 d (Ragab and Mahmoud, 2015). Magnetically treating the water increased pH, increased electrical conductivity, salinity, oxygen content and reduced surface tension, evaporating temperature, and chloride concentration with no effect on viscosity or bacterial count. After 30 d of drinking SW, the does were then inseminated by a rabbit buck, and live body weight (LBW) was measured at mating and kidding. Water magnetized using the 1,200 G magnet resulted in the highest LBW at mating and kidding. Does drinking the 3,600 G water had an intermediate LBW to that of control and 1,200 G water. Litter weight gain was also significantly higher (by ~10%) with both magnetized waters. At weaning, the weight of the 1,200 G kids was significantly greater at 2,807 ± 104 g compared with 2,434 g (controls) and 2,635 g (3,600 G water). It was concluded that drinking water magnetized with 1,200 G magnets conferred a number of important growth benefits without adverse effects, and that the results with 1,200 G water were significantly better than those obtained using 3,600 G water.

Poultry

The first study using an SW for the drinking water supply in poultry was performed using growing chickens (Al-Mufarrej et al., 2005). Tap water was passed through a magnetic funnel (7 circular magnets of 450 to 500 G each) at low speed and collected in graduated cylinders, with fresh SW provided at 12-hr intervals. There was no effect of SW on growth, water consumption, feed intake, and feed conversion ratio during the 32 d after hatching. There was also no significant effect on carcass composition at 32 d, nor on antibody response to sheep red blood cells. The authors concluded that drinking the SW had no influence on measured parameters. It appears that the degree of water structuring was too low to produce any effects.

Alhassani and Amin (2012) provided chickens with water treated by a 500-G magnetic device, with tap water (control) flowing past the magnet at 3 different speeds: slow (10 L/15 min), medium, and fast (10 L/5 min). Effects of water treatment on physicochemical properties was not reported. Birds were studied from hatching till 42 d, with weekly measures. There was a weak dose–response effect between duration of magnetic water treatment and gain in LBW, with no statistical difference in body weight, weekly weight gain, and feed conversion ratio between the 4 groups. Because the tendency for a dose– response effect was present, this raises the possibility that the intensity/duration of magnetic treatment of the water was not adequate to elicit the types of biological responses reported in most studies.

El-Katcha et al. (2017) studied the effects of drinking magnetized (details not provided except that exposure to magnets occurred every 6 hr) water on the growth of Pekin ducklings from age 1 d posthatching to 12 wk. The characteristics of the water were not reported. There was no effect of the water treatment on body weight gain, nor on any measured parameter (hematology, serum biochemistry, liver function, renal function, serum lipids, immune parameters, and tissue weights) compared with nontreated water. Histological examination of the duodenal, jejunal ileum epithelium showed an increase in intestinal villi length, width, and surface area with SW compared with controls. The authors postulated that these effects of magnetically treated water would be associated with increased nutrient absorption. The paucity of effects in this very detailed study suggests that the intensity of water treatment was inadequate compared to that performed by most other studies.

Hassan et al. (2018) had growing chickens (hens) drink tap water treated with 2,000, 3,000, and 4,000 G or control water for 21 wk. Body weight gain of hens was ~25% greater when drinking the 2,000 and 3,000 G waters compared with control and 4,000 G waters. This was associated with an increased daily feed intake only with 3,000 G water, and feed conversion ratio (g feed/g egg) and daily water intake were significantly reduced with all SWs.

The poultry studies indicate that waters treated with magnetic field strengths of less than 1,000 G do not result in any differences in measured parameters related to growth and performance. The study by Hassan et al. (2018) indicates that water treated with a 4,000 G magnetic field strength resulted in effects that were less than optimal compared with waters treated with 2,000 and 3,000 G.

Fish

In one of the first reported studies on animals, Zhang and Wu (1987) demonstrated that fish living in magnetized water had a reduction in renal calcium crystal and in tissue calcium content. No additional details are readily available.

Effects of Drinking SW on Milk Yield and Composition

The 28-d milk yield of rabbit does drinking SW was increased by 300 to 500 g, compared with yields of 3,808 to 4,200 g in tap and well water control groups, respectively (El-Hanoun et al., 2013). The milk from does drinking SW had significantly greater fat, lactose, and total energy, with no effect on milk protein and total solids, compared to milk from does drinking control waters.

Balieiro Neto et al. (2014) studied the effects of drinking a magnetized water (static magnetic field of 32,300 G applied to water troughs) on cows for a period of 75 d. There were significant increases in milk protein, urea, and casein, with no effect on daily milk yield, milk fat, and milk lactose.

Milk yield and milk composition of lactating goats has been reported in 3 studies (Sargolzehi et al., 2009; Ragab and Mahmoud, 2015; Yacout et al., 2015). All studies used tap water as a control, as well as waters conditioned using 1,200 and 3,600 G magnets using a flow-through system. Animals consumed the water for 60 d. The water used by Sargolzehi et al. (2009) was "low quality" and very hard (2,168 ppm CaCO₃) with concentrations of SO42-, Na+, and Cl- that greatly exceeded the recommended upper level for livestock (Ayers and Westcot, 1985). The study was also greatly underpowered as there were only 4 animals per treatment. The authors reported no effect of consuming magnetized, low-quality water on milk composition and serum biochemistry. No adverse effects were reported. Yacout et al. (2015) also studied daily milk yield of lactating goats consuming control or 1,200 or 3,600 G waters (n = 3 to 5 per group). Animals consuming 3,600 G water had greater milk yield than both other groups. Milk from does drinking SW also has high total solids, solids not fat, fat, protein, and lactose than those drinking control water. Shamsaldain and Al Rawee (2012)

studied 3 groups of sheep that received control water or water magnetized at 500 and 1,000 G (n = 8 per group). The authors reported increased milk production, total solids, fat, and protein from sheep consuming 1,000 G water compared with control. In the study by Ragab and Mahmoud (2015), daily milk yield was also highest (10% to 40% higher than controls), with significantly greater fat, protein, lactose, and total solids when consuming the 1,200 G water; values with 3,600 G water were intermediate.

The results of these milk studies indicate consistent improvements in milk production and quality when animals drink SW treated with magnets at field strengths between 1,000 and 3,000 G, with field strength greater than 3,000 G showing less than optimal outcomes, although still better than outcomes on control waters.

Effects of Drinking SW on Blood Hematology and Biochemistry

The 75-d study using cows (Balieiro Neto et al., 2013, 2014) also reported on arterial and venous blood acid–base and ion status when drinking water treated with a 32,400 G magnetic field in a water trough. While all parameters remained within normal reference ranges, there were statistically significant decreases in base excess, bicarbonate concentration, osmolality, and PCO₂. There were significant increases in arterial pH, venous oxygen saturation, and blood urea, and no effects on serum glucose and ion concentrations.

The study performed using goats by Yacout et al. (2015) reported a positive dose–response effect of consuming 0, 1,200, or 3,600 G magnetized water on the concentrations of erythrocytes, hemoglobin, and white blood cells. Similar dose–response increases were reported for serum glucose, total protein, albumin, and globulin, while cholesterol was reduced. A study on goats (n = 4 per group), also receiving "low-quality" hard water treated at 0, 1,200, and 3,600 G (Sargolzehi et al., 2009) reported no effects on serum biochemistry. Similar effects were reported when sheep consumed water treated with 1,000 G magnet (Shamsaldain and Al Rawee, 2012) and plasma protein concentration was also significantly increased when rabbits consumed magnetically treated water for 30 or 60 d (Khudiar and Ali, 2012).

Lindinger and Northrop (2020) also reported an absence of effect on all full-panel indices of hematology serum biochemistry when Thoroughbred racehorses drank 10 L per day of a stable SW product for 4 wk.

The study by El-Hanoun et al. (2013) on rabbit does reported significantly lower serum concentrations of liver enzymes (aspartate aminotransferase, AST; alanine transaminase, ALT) and significantly greater serum concentrations of ovarian hormones (estrogen and progesterone). In contrast, mice given 1,000 and 2,000 G water to drink reported no effect on blood concentrations of the liver enzymes AST and alkaline phosphatase (ALP; Alhammer et al., 2013). In the study by Ragab and Mahmoud (2015) rabbit does were given normal tap water or magnetized tap water treated with 1,200 or 3,600 G for a total of 60 d, before and during gestation and during lactation. There were minor differences in serum biochemistry between waters, with all parameters within normal reference ranges, and no adverse effects. The study by Attia et al. (2015) reported significantly increased serum albumin without change in globulin or albumin: globulin ratio, a reduction in ALT, and significant decreases in serum urea and urea:creatine ratio indicative of improve renal function. There were no effects on RBC count, WBC count nor on white cell differentials. Mahmoud et al. (2019) using rabbit bucks consuming SW for 90 d, reported increases in RBC count, hematocrit and hemoglobin without effect on WBC count and platelet count.

A study on Japanese quail used tap water treated with 500 or 1,000 G magnets, which the birds drank for 60 d (Al-Hilali, 2018). Magnetic treatment of water resulted in significantly increased electrical conductivity and pH, with significant reductions in density, dissolved oxygen, surface tension, and Cl⁻ concentration. At 60 d, there were significant increases in RBC count, hematocrit, and hemoglobin concentration with both SWs. With 1,000 G, but not 500 G, SW, there were significant increases in WBC count, ALP, and serum total protein.

In chicken hens consuming control 2,000, 3,000, or 4,000 G treated waters for 21 wk, the authors reported increased RBC count and hemoglobin without change in hematocrit, increased serum pH, glucose, globulin, phosphorous and triiodothyronine concentrations and a reduced albumin:globulin ratio, with 2,000 and 3,000 G waters. There were no effects on total protein, albumin, calcium, and calcium:phosphorous ratio.

Effects of SW on Reproduction

Forty female mice drank a pure SW (exposed to 4,000 G field at 37.5 °C for 4 hr), compared with a control group (n = 40) that drank normal tap water (Hafizi et al., 2014). Two weeks after starting to drink the water, mice were stimulated to ovulate and 48 hr later bred to males. After a further 54 hr, pregnant mice were killed and the reproductive system examined. The mean \pm SD number of corpus lutea in SW mice was significantly greater (9 ± 4) than in the control group (5 ± 2), with a ~10% increase in height of fallopian tube epithelial cells and a ~5% increase (P = 0.052) in height of uterine epithelial cells. The authors postulated that drinking SW had positive effects on cell growth, mediated by unknown mechanisms. The increased number of corpus lutea, and increased reproductive tract epithelial cell height, may translate to improved implantation and litter size.

In the study by Ragab and Mahmoud (2015), rabbit does aged 7 to 8 mo were given normal tap water or magnetized tap water treated with 1,200 or 3,600 G for 30 d. After 30 d of drinking SW does were then inseminated by a rabbit buck. Water magnetized using the 1,200 G magnet resulted in the highest first conception rate (50% compared with 40% control and 30% with 3,600 G magnetization). There was no effect on gestation duration. The litter size per doe averaged ~10% higher for does receiving 1,200 G water and mortality rate was similarly reduced by ~10%. Litter weight gain was also significantly higher (by ~10%) with both magnetized waters.

Effects of drinking SW on reproductive indices in rabbit bucks show consistent beneficial effects on semen and sperm quality. Mahmoud et al. (2019) reported increased libido, improved semen volume and quality, increased spermatozoa count and motility, and reduced numbers of abnormal and dead spermatozoa when drinking 2,000 G SW for 4 wk. El-ratel and Fouda (2017) reported improved semen quality and sperm output when consuming SW (3,600 G) for 90 d. Both these studies confirmed previously reported effects (Attia et al., 2015), and these authors additionally reported significantly increased testosterone concentration.

In the study on chicken hens (Hassan et al., 2018), the authors reported a tendency (P < 0.1) toward increased egg production and, importantly there were significant increases in egg weight

and egg mass/hen day⁻¹ with all 3 SWs (2,000, 3,000, and 4,000 G). The eggs from hens consuming 2,000 and 3,000 G SW were characterized by significantly increased albumin and yolk weights compared with controls and 4,000 G water, and shell thickness was increased with all 3 SWs.

Effects of Drinking SW on Blood Antioxidant/Immune Status

Adult male rabbits (n = 10 per group) were provided with magnetically treated tap water (resulted in elevated pH and oxygen content, with reduced surface tension and chloride) or control water for 60 d (Khudiar and Ali, 2012). Drinking the magnetically treated water resulted in a significant ~40% increase in serum glutathione concentration by 30 d. The study on rabbit does by El-Hanoun et al. (2013) reported significant increases in serum total antioxidant capacity with reduced TBARS in both groups drinking SW compared with the control tap and well water groups. El-Ratel and Fouda (2017) a decrease in blood markers of oxidant stress (malonyl dialdehyde, TBARS, and lysozyme content) while total antioxidant capacity and antibody titer were increased when rabbit bucks consumed SW for 90 d.

Rats with induced type 2 diabetes consumed magnetized water for 4 wk and, compared with controls had decreased activities of glutathione and superoxide dismutase 2 that the authors associated with a reduced level of oxidative stress (Saleh et al., 2019). In the earlier study using rats with induced diabetes (Lee and Kang, 2013) found no effect of drinking SW on erythrocyte activities of catalase, glutathione peroxidase, and superoxide dismutase. In the mouse study of Alhammer et al. (2013), a significant increase in adenosine deaminase when drinking 1,000 G water, but not with 2,000 G water, was attributed as an immune system response.

Japanese quail drinking SW for 60 d had significantly increased serum glutathione concentrations, with the increase positively correlated with intensity of water treatment (500 G and 1,000 G magnets; Al-Hilali, 2018). In rabbit bucks (Attia et al., 2015), 28 wk of drinking magnetized tap water or magnetized well water significantly increased the serum concentrations of glutathione, glutathione peroxidase, glutathione S-transferase, IgA, and antibody titer. These were associated with significantly reduced concentrations of the lipid peroxidation marker malonyl dialdehyde and TBARS, with no effect on IgG, IgM, lysozyme concentrations, superoxide dismutase activity, and total antioxidant capacity.

Effects of Drinking SW on Serum Lipid Profile

In the study of male rabbits by Khudiar and Ali (2012), the authors reported that drinking magnetically treated water resulted in significant reductions in serum triacylglycerol and very low density lipoproteins (VLDL) concentrations, and significantly increase high-density lipoproteins (HDL), at 60 d, compared with controls. A 28-wk study on 7.5-mo-old rabbit bucks reported a significant (~20%) increase in serum total lipid concentration with no effect on total cholesterol concentration (Attia et al., 2015). In rats with induced type 2 diabetes, drinking SW prevented a 50% increase in plasma triglycerides (TGs) compared with diabetic rats drinking control water, with no effect on total cholesterol, HDL, and low-density lipoproteins (LDL; Lee and Kang, 2013). In the study on in Japanese quail (Al-Hilali, 2018), both SWs had similar effects on lowering by ~20% total serum cholesterol and TG concentrations and increasing HDL. The magnitude of decrease in LDL and VLDL was positively correlated with the intensity of magnetic treatment of the water (550 G and 1,000 G). These are considered to be positive, beneficial effects on serum lipid profile.

Glycemic Responses and Type 2 Diabetes

Adult mice were given tap water treated with 1,000 G or 2,000 G magnets or consumed normal tap water (controls; Alhammer et al., 2013). The magnetic treatment reduced the water density (~10%), increased total dissolved solids at 2,000 G, and was without effect on conductivity, dissolved oxygen, pH, and salinity. Mice that consumed 1,000 G or 2,000 G water (duration not stated) had a ~30% and ~40% decrease, respectively, in blood glucose with no effect on AST or ALP activities. Japanese quail that drank 500 G and 1,000 G SW for 60 d also showed significant large (12% to 15%) decreases in serum glucose with the magnitude of decrease positively correlated with intensity of water treatment (500 and 1,000 G magnets; Al-Hilali, 2018).

Rats with induced type 2 diabetes consumed water treated by passing through a magnetic field of 9,000 to 13,000 G for 4 weeks. Compared to diabetic rats drinking control water, diabetic rats drinking the SW showed decreased blood glucose and glycated hemoglobin concentrations, with reductions in blood and liver DNA damage, but there was no difference in the results of the intra-peritoneal glucose tolerance test or plasma insulin (Lee and Kang, 2013). In another study of induced type 2 diabetes in rats, water was passed through a 600 G magnet, and the water consumed for 4 wk; the authors reported increased pancreatic β -cell mass and insulin expression (Saleh et al., 2019).

In an abstract and study report, the results of a clinical study performed using diabetic patients are presented (Wang et al., 2004). In this multicenter, clinical trial subjects with type II diabetes (n = 164) were provided 250 mL of SW (control group received distilled water; n = 162) twice daily for 4 wk; this represents about 20% of daily water intake. In subjects with blood glucose lower than 8 mmol/L, there were no changes in cellular hydration, and no adverse effects. In subjects with blood glucose >8 mmol/L, there were significant increases in cellular hydration and health as determined using bioelectrical impedance analysis. It was concluded that SW has beneficial effects on cell health and metabolism in subjects with moderate-to-severe type 2 diabetes.

Other Biological Effects of Drinking SW

The consumption of magnetized water by children infected with the parasitic condition ascariasis resulted in resolution of the condition in "most cases" with "no side effects" (Wu 1989). Further details on both these studies are not readily available.

Adult rats (n = 5 per group) were given drinking water from the tap (controls) or magnetized water at intensities of 250, 750, 1,000, 1,500 G every day for 30 d, after which the heart, lung, and spleen were examined (Al-Saffar et al., 2013). There were no gross or histological effects reported for heart. Histological examination of lung tissues showed lymphoid hyperplasia when rats consumed the 750 and 1,000 G waters. Examination of spleen also showed hyperplasia of the white pulp with 250 G water, and lymphoid hyperplasia with 750 and 1,000 G waters, with lesions progressing to areas of necrosis with 1,500 G water.

In the study by Lee and Kang (2013) using diabetic rats, the authors also tested the lymphocyte and hepatocyte populations for evidence of DNA damage using comet assays. DNA damage was significantly greater in diabetic rats consuming control water or SW compared with control rats drinking control water. However, in diabetic rats consuming SW, there was a significant ~70% reduction in DNA damage compared with diabetic rats drinking control water. Al-Hilali (2018) used the mitotic index of bone marrow cells (a measure of the rate of cell division) to assess "genetic" damage after quail drank SW for 60 d. The magnitude of increase in mitotic index was positively correlated with the intensity (500 and 1,000 G) of the tap water (probability of DNA damage: control 0.055, 500 G water 0.063, and 1,000 G water 0.085) all of which are well below the probability (0.20) that begins to be associated with genetic damage (Pedersen et al., 2016). Thus, the increase in mitotic index indicates only small increases in mitotic cells division; one could even consider this to be a beneficial effect.

When rats drank SW for up to 45 d, there was an increase in bone mineral content, bone mineral density and increased breaking resistance by 45 d (Balieiro Neto et al., 2017).

The study performed on Thoroughbred racehorses in active training showed that compared with control water, horses drinking 10 L per day of SW for 4 wk showed an increase in whole body and extracellular hydration (Lindinger and Northrop, 2020). The horses also had improved upper airway health (less mucous, swelling, and indications of inflammation) when examined endoscopically after workout gallops, and an increased heart rate variability when resting quietly in their stalls. The increase in resting heart rate variability is indicative of a more restful autonomic state.

In an abstract, Chen et al. (2005) provided by gavage 0.5 mL of 0, 33.3% or 100% of a stabilized SW product to mice (n = 14treatment and 14 controls) for 30 d. Mice normally drink 4 to 6 mL/d, and a 25-g mouse has a maximum stomach volume of 0.5 mL. Before provided SW, and after 30 d, the mice performed a swim endurance test. Compared to baseline and a control group that did not receive SW, mice that were given SW increased swim duration from 16 \pm 8 to 24 \pm 10 min (33.3% SW; P <0.05) and to $38 \pm 30 \text{ min}$ (100% SW). The authors correlated increased swim time with increased pre-exercise liver glycogen stores g/100 g; control: 41 ± 12 vs. 33.3 % SW: 65 ± 20 (P < 0.002 compared with control) and 100% SW: 67±16 (P < 0.0002 compared with control) in a separate group of mice treated the same way except for no exercise. It was concluded that providing 3% to 10% of the daily water intake in the form of SW was able to increase liver glycogen content and increase swim duration.

While not a drinking study, Gupta and Bhat (2011) examined the effects of water magnetized for 24 or 72 hr on the ability to inhibit oral Streptococcus mutans. The details of the water treatment are not provided, other than 72 but not 24 hr of treatment results in increased pH and a 55% reduction in electrical conductivity. Children were provided with 10 mL of the SWs or 10 mL of a 0.2% chlorhexidine solution and instructed to rinse the mouth for 1 or 3 min. The authors reported significant reductions in S. mutans with 1 min (72 hr treated water) and 3 min (24 hr treated water) of rinsing with SW, and that results obtained with 1 min of rinsing with 72 hr-treated water were similar to those obtained with chlorhexidine. Goyal et al. (2017) also magnetized pure, still water for 72 hr. When children used 10 mL of this water as a mouth rinse, twice daily for 2 wk, there was a significant reduction in S. mutans count in samples of dental plaque and saliva. Therefore, water structured in this way appears to exhibit anti-microbial effects.

Research conducted during the past 2 decades provide scientific evidence that the consumption of SW, compared with unstructured liquid water, confers a wide range of benefits to all of the animals studied to date so long as intensity and duration of water treatment are not excessive. The few studies that examined "dose-response" effects consistently showed that water exposed to magnetic fields of between 1,000 and 4,000 G for brief periods, i.e., flow-through systems, provided a number of physiological benefits compared with control waters. Some studies, however, indicate that field strengths of 3,000 G and higher may generate undesirable effects. There is evidence of increased growth, egg mass, milk yield, carcass mass, improved reproductive indices, improved blood lipid and glycemic profiles and improved blood/systemic antioxidant and inflammation profiles. It is both interesting and disappointing that none of these studies have examined putative mechanisms for these effects, so we are left with descriptive studies. These descriptive studies, however, can be used to guide innovative and welldesigned research to examine effects of SW on biological systems. Based on these descriptive studies, the applications are broad and include all aspects of agriculture, as well as plant and animal health. The potential for applications in animal and human medicine was also exemplified in studies that focused on diabetes, and other animal studies indicated improvements in blood lipid profile. Based on what is known about how water is organized around cells, it is likely that perfusing the cells with an SW product will change cellular functions, protein functions, and molecular interactions in numerous and various ways. Future studies need to be aimed at elucidating main effects using cellular and organ physiology techniques.

Conclusions

Magnetic field strengths used to treat water vary from 500 to 32,400 G, with duration of treatment ranging from seconds (magnetic flow-through systems using several high field strength magnets) to 72 hr using small volumes with static, low field strength magnets. Inadequate water treatment results in no or minimal biological effect, whereas excessive treatment may be associated with adverse effects (see below). Based on the results of the studies presented below, a magnetic field strength of 1,000 to 3,000 G is required to generate water capable of exerting beneficial effects, while waters treated with field strengths greater than 5,000 G may result in detrimental effects. Future studies need to provide detailed methodology and some key physicochemical properties that are changed by the structuring process, and the duration of structural stability. Future research needs to determine the water treatment conditions that optimize for specific biological outcomes and the researchers must measure and report several key indicators that demonstrate water structuring. The latter, in particularly, would be most helpful in performing comparison between studies. The animal research conducted to date consistently demonstrated beneficial effects of SW consumption. Additional research is needed to demonstrate how these effects occur, and if these types of SWs are safe to consume and use over the long term.

Acknowledgment

The research for this review was conducted as part of a larger paid research contract to The Nutraceutical Alliance Inc. by Defiance Brands, Inc., Nashville, TN, USA. The authors declare no real or perceived conflicts of interest.

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