

## Structured Water: effects on animals<sup>1</sup>

Michael I. Lindinger, PhD

President, The Nutraceutical Alliance Inc.

Burlington, ON, Canada L7N 2Z9

Email: [michael@nutraceuticalalliance.ca](mailto:michael@nutraceuticalalliance.ca)

Phone: +1 289 812 6176

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## **Abstract**

This review focuses on the effects of structured water on animals when it is consumed on a daily basis. Structured water 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 structured water with stability of at least 3.5 months. A variety of laboratory and farm animals have been studied over the past twenty years. Consistent (three or more studies) responses amongst animals consuming structured water for one month 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 structured waters. The mechanisms by which structured water affects biological systems are unknown and require investigation. Effects of structured water, when taken up by biological systems, are likely associated with altered water structuring around biological surfaces, such as proteins and membranes.

## **Keywords**

magnetized water, clustered water, production animals, antioxidant effects, reproduction, antioxidant status

## List of Abbreviations

ALP, alkaline phosphatase

AST, aspartate aminotransferase

ALT, alanine aminotransferase

DMI, dry matter intake

G, Gauss; The SI unit for magnetic field strength is Tesla; 1 Tesla = 10,000 G. Gauss is used in the manuscript because all of the cited literature reports units in Gauss.

HDL, high density lipoproteins

IgA, immunoglobulin A

IgG, immunoglobulin G

IgM, immunoglobulin M

LDL, low density lipoproteins

ppm, parts per million

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

## Introduction

Only in the past two 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. Structured water 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 two 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 us consider what is meant by “water gains structure”. As depicted in Figure 1, SW may be defined as liquid water that has gained structure compared to 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 two 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 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 amongst 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 structured water 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.

## **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 years 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; Pang and Deng, 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 short-lasting and, in all of the experiments of Pang and Deng (2008) represented by Figure 5, infrared absorbance had returned to baseline by 60 minutes 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 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, 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 two main 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 days. The degree of structuring is proportion 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 (Chibowski and Szcześ 2018; Pang, Deng, and Tang 2012). The second type of SW has long-term stability (months).

Nearly 20 years ago, a SW with months-long stability was developed by treating purified water with approximately 0.1% by weight of selected minerals that include potassium and silica, magnetic energy and light energy (Lorenzen, 1988; Lorenzen, 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 months after date of manufacture, this SW was found to retain significant structuring, even after boiling in a microwave for 5 minutes. 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<sup>®</sup> (Defiance Brands Inc., Nashville, TN, USA).

### **Effects of structured water on animal growth and development**

As detailed in the following paragraphs, there are more than two dozen studies that have reported beneficial effects of drinking or using SW on animals, including two 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 three 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 report in a trade magazine indicated increased milk yields in dairy cows (Lin, 1990). A 75-day study using Jersey cows drinking water treated with a static 32,4000 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 structured water 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) one week 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 days 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 (1000 G) compared to control, but this study also did not report water properties.

(Yacout et al., 2015) studied goat bucks and lactating does that consumed magnetized waters (1200 and 3600 G) for 60 days. Consuming the 3600 G water, compared to control and 1200 G water, resulted in significantly increased dry matter intake (DMI) – with water consumption matched to DMI and increased digestibility with both magnetic waters compared to 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 1000 G or 2000 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 2000 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 weeks. There was no effect of drinking 1000 G water on body weight, while body weights were significantly lower (~13%) for mice drinking the 2000 G water. There was no effect on feed consumption, although water consumption was increased by 40-50% in both groups drinking SW. The results of this study suggest a possible dose-response effect.

Lee and Kang (Lee and Kang, 2013) structured water by passing it through a magnetic field of 9,000 to 13,000 G and the water was consumed within 1 day. 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 two groups, one consuming control water and the other SW over a period of 8 weeks. There was no control group of rats that consumed SW. The diabetic

rats showed a greater than 60% reduction in weight gain compared to control rats despite a 15% increase in daily food intake and 2.5-fold greater daily water intake compared to controls. Diabetic rats drinking SW gained ~30% less weight than diabetic rats drinking control water, and daily food intake between the two 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-week period.

Baliero Neto et al. (Balieiro Neto et al., 2014; Balieiro Neto et al., 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 days. Compared to control water, the magnetically treated water had an increased pH and reduced turbidity – other physical parameters were not assessed. Over the 45 days, 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 dry matter intake, 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. (El-Hanoun et al., 2013) provided tap water and well water either untreated or treated with a 4000 G magnetic field to rabbit does (n = 10 per group; aged 6 – 7 months) for 28 weeks, with 12 weeks prior to mating and 12 weeks post-gestation (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 ( $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{Cl}^-$ ,

$\text{CO}_3^{2-}$ ,  $\text{HCO}_3^-$ ). The growth performance of litters was followed from 6 to 12 weeks post-parturition and prior to weaning (n = 143 to 266 per group). Does drinking SW, compared to tap water or well water controls, for 12 weeks had significant 2-fold greater weight gains at mating and at 7 days after mating, accompanied by increased daily feed intake. Litter size and litter weight at birth and at 28 days 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 weeks post-parturition, 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 4000 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 4000 G and provided as drinking water to 7.5-month old rabbit bucks (n = 10 per group, 4 groups) for 28 weeks. 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 months were given normal tap water or magnetized tap water treated with 1200 G or 3600 G for 30 days (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 days 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 1200 G magnet resulted in the highest LBW at mating and kidding. Does drinking the 3600 G water had an intermediate LBW to that of control and 1200 G water. Litter weight gain was also significantly higher (by ~10%) with both magnetized waters. At weaning, the weight of the 1200 G kids was significantly greater at  $2807 \pm 104$  g compared to 2434 g (controls) and 2635 g (3600 G water). It was concluded that drinking water magnetized with 1200 G magnets conferred a number of important growth benefits without adverse effects, and that the results with 1200 G water were significantly better than those obtained using 3600 G water.

### **Poultry**

The first study using a 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 – 500 G each) at low speed and collected in graduated cylinders, with fresh SW provided at 12-hour intervals. There was no effect of SW on growth, water consumption, feed intake and feed conversion ratio during the 32 days after hatching. There was also no significant effect on carcass composition at 32 days, 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 three different speeds: slow (10 L / 15 minutes), medium and fast (10 L / 5 minutes). Effects of water treatment on physicochemical properties was not reported. Birds were studied from hatching till 42 days, with weekly measures. There was a weak dose-response effect between duration of magnetic water treatment and gain in live body weight, with no statistical different in body weight, weekly weight gain and feed conversion ratio between the 4 groups. Because of 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. (El-Katcha et al., 2017) studied the effects of drinking magnetized (details not provided except that exposure to magnets occurred every 6 hours) water on the growth of Pekin ducklings from age 1 day post-hatching to 12 weeks. 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, tissue weights) compared to non-treated water. Histological examination of the duodenal, jejunal ileum epithelium showed an increase in intestinal villi length, width and surface area with SW compared to 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. (Hassan et al., 2018) had growing chickens (hens) drink tap water treated with 2000 G, 3000 G, 4000 g or control water for 21 weeks. Body weight gain of hens was ~25% greater when drinking the 2000 g and 3000 G waters compared to control and 4000 G waters. This was associated with an increased daily feed intake only with 3000 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 1000 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 4000 G magnetic field strength resulted in effects that were less than optimal compared with waters treated with 2000 G and 3000 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 structured water on milk yield and composition**

The 28-day milk yield of rabbit does drinking SW was increased by 300 to 500 grams, compared to yields of 3808 g to 4200 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, more 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 days. 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 three studies (Sargolzei 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 1200 G and 3600 G magnets using a flow-through system. Animals consumed the water for 60 days. The water used by Sargolzei et al. (2009) was “low quality” and very hard (2168 ppm  $\text{CaCO}_3$ ) with concentrations of  $\text{SO}_4^{2-}$ ,  $\text{Na}^+$  and  $\text{Cl}^-$  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. (Yacout et al., 2015) also studied daily milk yield of lactating goats consuming control or 1200 G or 3600 G waters (n = 3 to 5 per group). Animals consuming 3600 g water had greater milk yield than both other groups. Milk from does drinking SW also has high total solids, solids not fat (SNF), fat, protein and lactose than those drinking control water. Shamsaldain and Al Rawee (Shamsaldain and Al Rawee, 2012) studied 3 groups of sheep that received control water or water magnetized at 500 G and 1000 G (n = 8 per group). The authors reported increased milk production, total solids, fat and protein from sheep consuming 1000 G water compared to 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 1200 G water; values with 3600 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 1000 and 3000 G, with field strength greater than 3000 G showing less than optimal outcomes, although still better than outcomes on control waters.

### **Effects of drinking structured water on blood hematology and biochemistry**

The 75-day study using cows (Balieiro Neto et al., 2013; Balieiro Neto et al., 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, PCO<sub>2</sub>. 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, 1200 G or 3600 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, 1200 G and 3600 G (Sargolzehi et al., 2009) reported no effects on serum biochemistry. Similar effects were reported when sheep consumed water treated with 1000 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 days (Khudiar and Ali, 2012).

Lindinger and Northrop (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 weeks.

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, progesterone). In contrast, mice given 1000 G and 2000 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 1200 G or 3600 G for a total of 60 days, 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:creatinine ratio indicative of improved 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 days, 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 1000 G magnets, which the birds drank for 60 days (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 days there were significant

increases in RBC count, hematocrit, hemoglobin concentration with both SWs. With 1000 G, but not 500 G, SW there were significant increases in WBC count, ALP and serum total protein.

In chicken hens consuming control 2000 G, 3000 g or 4000 G treated waters for 21 weeks 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 ration, with 2000 G and 3000 G waters. There were no effects on total protein, albumin, calcium, and calcium:phosphorous ratio.

#### **Effects of structured water on reproduction**

Forty female mice drank a pure SW (exposed to 4000 G field at 37.5°C for 4 hours), compared to 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 h later bred to males. After a further 54 hours, pregnant mice were killed and the reproductive system examined. The mean  $\pm$  SD number of corpus lutea in SW mice was significantly greater ( $9 \pm 4$ ) than in the control group ( $5 \pm 2$ ), 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 months were given normal tap water or magnetized tap water treated with 1200 G or 3600 G for 30 days. After 30 days of drinking SW does were then inseminated by a rabbit buck. Water magnetized

using the 1200 G magnet resulted in the highest first conception rate (50% compared to 40% control and 30% with 3600 G magnetization). There was no effect on gestation duration. The litter size per doe averaged ~10% higher for does receiving 1200 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 structured water on reproductive indices in rabbit bucks show consistent beneficial effects on semen and sperm quality. Mahmoud et al. (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 2000 G SW for 4 weeks. El-Ratel and Fouda (El-ratel and Fouda 2017) reported improved semen quality and sperm output when consuming SW (3600 G) for 90 days. 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 ( $p < 0.1$ ) towards increased egg production and, importantly there were significant increases in egg weight and egg mass/hen.day<sup>-1</sup> with all three SWs (2000, 3000 and 4000 G). The eggs from hens consuming 2000 g and 3000 g SW were characterized by significantly increased albumin and yolk weights compared to controls and 4000 G water, and shell thickness was increased with all three SWs.

#### **Effects of drinking structured water 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 days (Khudiar and Ali, 2012). Drinking the magnetically treated water

resulted in a significant ~40% increase in serum glutathione concentration by 30 days. 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 to the control tap and well water groups. El-Ratel and Fouda (El-Ratel and Fouda, 2017) a decrease in blood markers of oxidant stress (malonyl dialdehyde, TBARS, lysozyme content) while total antioxidant capacity and antibody titer were increased when rabbit bucks consumed structured water for 90 days.

Rats with induced type 2 diabetes consumed magnetized water for 4 weeks and, compared to 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 erythrocytes activities of catalase, glutathione peroxidase and superoxide dismutase. In the mouse study of Alhammer (Alhammer et al., 2013) a significant increase in adenosine deaminase when drinking 1000 G water, but not with 2000 G water, was attributed as an immune system response.

Japanese quail drinking SE for 60 days had significantly increased serum glutathione concentrations, with the increase positively correlated with intensity of water treatment (500 G and 1000 G magnets) (Al-Hilali, 2018). In rabbit bucks (Attia et al., 2015), 28 weeks of drinking magnetized tap water or magnetized well water significantly increased the serum concentration of glutathione, glutathione peroxidase, glutathione S-transferase, IgA and antibody titer. These were associated with significantly reduced concentration of the lipid peroxidation marker malonyl dialdehyde and TBARS, with no effect on IgG, IgM, lysozyme concentration, superoxide dismutase activity and total antioxidant capacity.

### **Effects of drinking structured water on serum lipid profile**

In the study of male rabbits by Khudiar and Ali (Khudiar and Ali, 2012) the authors reported that drinking magnetically treated water resulted in significant reductions in serum triacylglycerol and VLDL concentrations, and significantly increase HDL, at 60 days, compared to controls. A 28-week study on 7.5 month 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 to diabetic rats drinking control water, with no effect on total cholesterol, HDL and 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 triglyceride 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 1000 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 1000 G or 2000 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 2000 G, and was without effect on conductivity, dissolved oxygen, pH and salinity. Mice that consumed 1000 G or 2000 G water (duration not stated) had a ~30% and ~40% decrease, respectively, in blood glucose with no effect on aspartate aminotransferase or alkaline phosphatase activities. Japanese quail that drank 500 G and 1000 G SW for 60 days also showed significant large (12-15%) decreases in serum glucose with the magnitude of decrease positively correlated with intensity of water treatment (500 G and 1000 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 weeks; the authors reported increased pancreatic  $\beta$ -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; Zhou 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 weeks; 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 greater than 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 II diabetes.

#### **Other biological effects of drinking structured water**

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, 1000, 1500 G every day for 30 days, 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 G and 1000 G waters. Examination of spleen also showed hyperplasia of the white pulp with 250 G water, and lymphoid hyperplasia with 750 G and 1000 G waters, with lesions progressing to areas of necrosis with 1500 G water.

In the study by Lee and Kang (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 to control rats drinking control water. However, in diabetic rats consuming SW there was a significant ~70% reduction in DNA damage compared to 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 days. The magnitude of increase in mitotic index was positively correlated with the intensity (500 g and 1000 G) of the tap water (probability of DNA damage: control 0.055, 500 G water 0.063 and 1000 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 days there was an increase in bone mineral content, bone mineral density and increased breaking resistance by 45 days (Balieiro Neto et al., 2017).



The study performed on Thoroughbred racehorses in active training showed that compared to control water, horses drinking 10 L per day of SW for 4 weeks 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. (Chen et al., 2005) provided by gavage 0.5 ml of 0, 33.3% or 100% of a stabilized SW product to mice (n = 14 treatment and 14 controls) for 30 days. Mice normally drink 4 – 6 ml per day, and a 25 g mouse has a maximum stomach volume of 0.5 ml. Before provided SW, and after 30 days, 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  $16 \pm 8$  min to  $24 \pm 10$  min (33.3% SW;  $p < 0.05$ ) and to  $38 \pm 30$  minutes (100% SW). The authors correlated increased swim time with increased pre-exercise liver glycogen stores (g/100 g; control:  $41 \pm 12$  vs 33.3 % SW:  $65 \pm 20$  ( $p < 0.002$  compared to control) and 100% SW:  $67 \pm 16$  ( $p < 0.0002$  compared to 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 (Gupta and Bhat, 2011) examined the effects of water magnetized for 24 or 72 hours on the ability to inhibit oral *Streptococcus mutans*. The details of the water treatment are not provided, other than 72 but not 24 hours 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 minutes. The authors reported significant reductions in *S. mutans* with 1 minute (72 h treated water) and 3 minutes (24 h treated water) of rinsing with SW, and that results obtained with 1 minute of rinsing with 72 h-treated water were similar to those obtained with chlorhexidine. Goyal et al. (Goyal et al., 2017) also magnetized pure, still water for 72 hours. When children used 10 ml of this water as a mouth rinse, twice daily for 2 weeks, 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.

### **Perspectives**

Research conducted during the past two decades provide scientific evidence that the consumption of structured water, compared to 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 1000 G and 4000 G for brief periods, i.e. flow-through systems, provided a number of physiological benefits compared to control waters. Some studies, however, indicate that field strengths of 3000 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 well-designed research to examine effects of structured water 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 a 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 G to 32,400 G, with duration of treatment ranging from seconds (magnetic flow-through systems using several high field strength magnets) to 72 hours 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 1000 to 3000 G is required to generate water capable of exerting beneficial effects, while waters treated with field strengths greater than 5000 G may result in detrimental effects. Future studies need to provide detailed methodology and some key physico-chemical 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 particular, 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 structured waters are safe to consume and use over the long term.

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

Parameter	Untreated tap water	500 G-treated	1000 G-treated
Conductivity (mS/cm)	650 ± 8.1	655.0 ± 8.6	710.0 ± 8.9*
pH	7.60 ± 0.07	7.62 ± 0.05	7.85 ± 0.02*
Density (mN/ml)	50.1 ± 2.25	40.0 ± 2.01*	40.0 ± 2.12*
Surface tension (dyn/cm <sup>2</sup> )	60.5 ± 2.8	52.4 ± 2.9*	50.4 ± 2.9*

Values are mean ± standard error.

\* indicates significantly different from untreated tap water. Data from Al-Halili (2018).

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## Figure Legends

Figure 1. A glass of water comprises billions of  $\text{H}_2\text{O}$  molecules (top) that are capable of accepting / releasing protons ( $\text{H}^+$ ) up to one million times per second (Edsall and Wyman, 1958; Harned and Owen, 1958). However, protons tend to not exist on their own in aqueous solutions and readily bind to  $\text{H}_2\text{O}$  to form  $\text{H}_3\text{O}^+$  (hydronium). In theory, two  $\text{H}_2\text{O}$  molecules can form one hydronium and one hydroxyl.

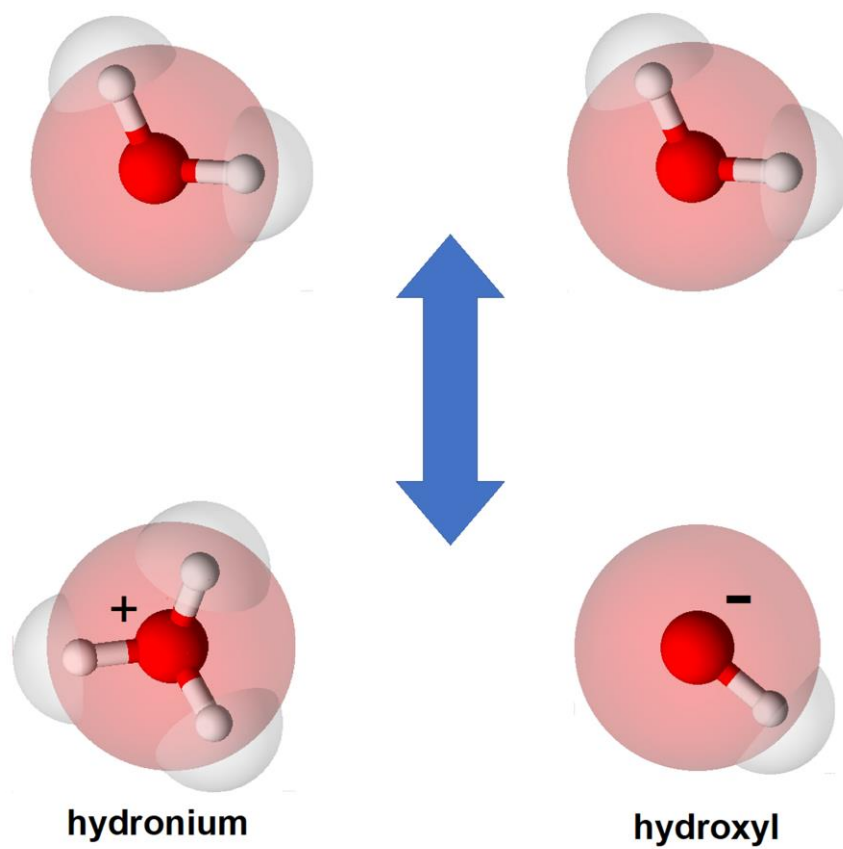
Figure 2. Examples of how individual  $\text{H}_2\text{O}$  may combine into linear or non-linear clusters. Water molecules are connected by hydrogen bonds as depicted by black dashes. Adapted from Pang (2014).

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

Figure 4. Ordered, hexameric structure of ice.

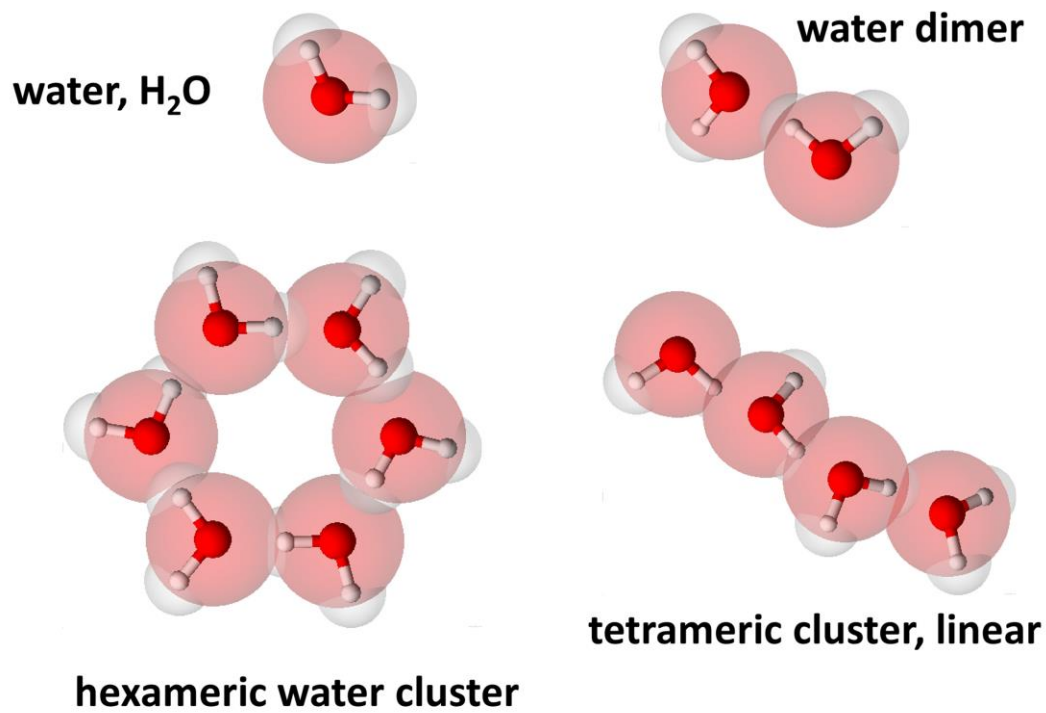
Figure 5. Representation of the effects of treating water with different magnetic field intensities for different durations. G – Gauss; 1 Tesla – 10,000 G. Adapted from Pang and Deng, 2008).

Figure 1



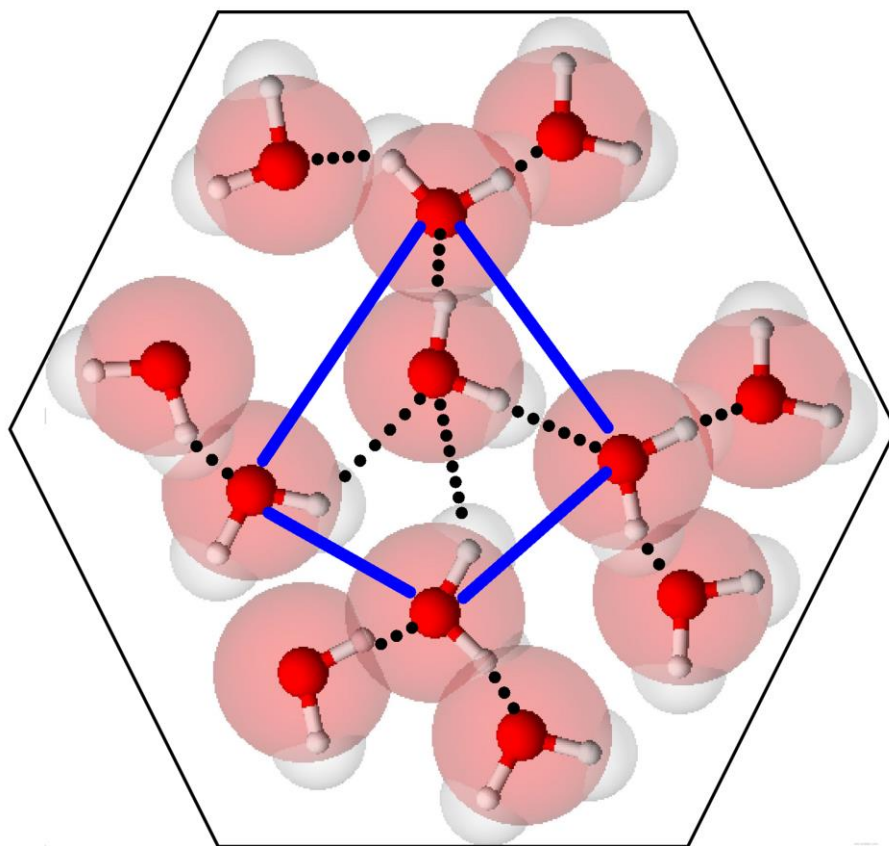
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Figure 2



Accepted

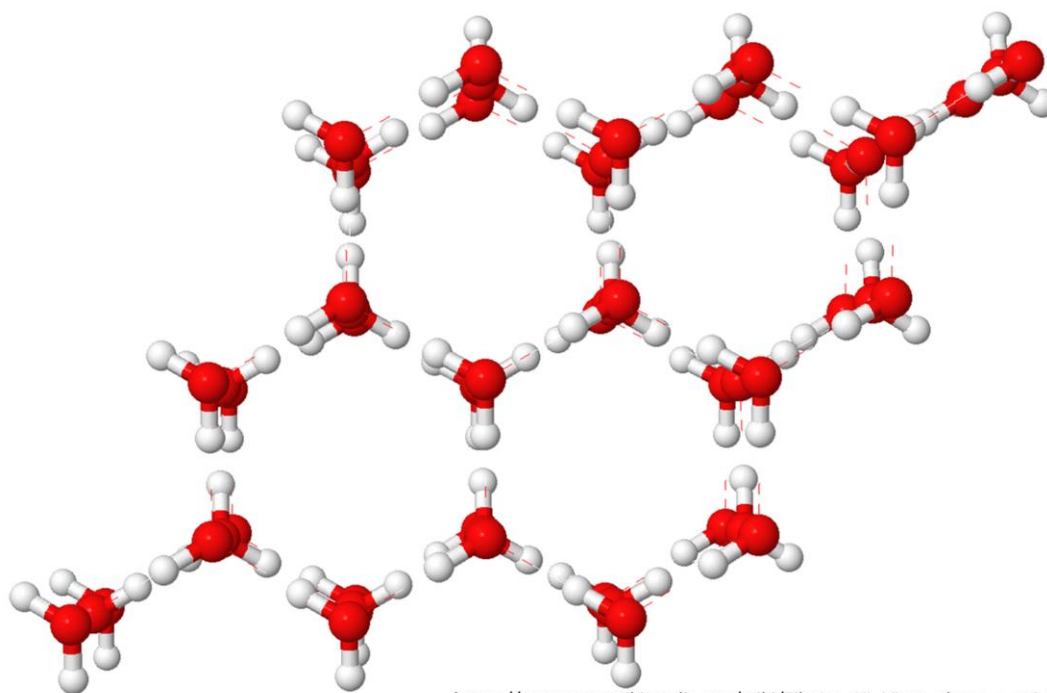
Figure 3



Accepted



Figure 4



[https://commons.wikimedia.org/wiki/File:Ice\\_XI\\_View\\_along\\_c\\_axis.png](https://commons.wikimedia.org/wiki/File:Ice_XI_View_along_c_axis.png)

Accepted

Figure 5

