A Literature Review on Intermittent Fasting

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Abstract

Intermittent fasting has been instructed in the Bible as an implied part of daily living. This paper has reviewed the physical benefits of intermittent fasting. In healthy test subjects, it has led to higher levels of autophagy, gut health, and lifespan. In non-healthy test subjects, it has proven beneficial in cancer treatment, as well as in protection against and/or attenuation of the effects of neurodegeneration, metabolic disorders, and cardiovascular diseases. The possible mechanisms underlying these benefits have been discussed in this review.

A Literature Review on Intermittent Fasting

Introduction

There are many instances in both the Old and New Testament where fasting is used as a means to seek a closer union with God. Paul and Barnabas fasted when seeking wise decision making (Acts 14:23, NASB), Ezra incited Israel to perform a fast to ask God for safe travels (Ezra 8:21-23), all of Nineveh took part in a fast as a sign of repentance (Jonah 3:5), and the prophetess Anna fasted as a sign of worship (Luke 2:37). In addition, Jesus' instructions in the gospel of Matthew implied that He believed fasting was an implicit practice (Matthew 6:16-18). With these instances and more showing a precedent for fasting throughout Scripture, those reading the Word of God can conclude that fasting is a spiritually beneficial act. As God is creator of not just our souls but also our earthly bodies, it would logically follow that He has in mind both of these components when he sets forth a precedent to partake in something. Indeed, modern research has evidenced many benefits of intermittent fasting for the health of the human body. The aim of this research has been to review both the advantages and disadvantages of intermittent fasting, and in what specific pathways God has designed both effects. The distinction has also been made between the effects of overall caloric restriction versus the effects of intermittent fasting.

Benefits in Healthy Persons

Intermittent fasting is undertaken in multiple different forms in today's world. Some participate in day-long fasts 1-2 times per week, while others may choose to restrict eating to a certain time frame every day. In healthy, non-diseased persons, either of these intermittent

fasting approaches may show various advantages, including increased autophagy, increased gut health, and possibly increased lifespan.

Autophagy

One way that intermittent fasting has proven beneficial is through the increased amount of autophagy occurring in the body (Kelekar, 2006). Autophagy is necessary to the human body, as it breaks down and recycles proteins and organelles. The purpose of this degradation is either to rid the cells of misfolded proteins and damaged organelles, or to use the components at another more vital location when there is low nutrient availability (Kelekar, 2006). However, this process naturally decreases with age, which causes protein aggregation that leads to degeneration of cells and many age-related diseases (Yang et al., 2019; Athanasios et al., 2018). In one study, mice participating on an alternate-day intermittent fasting regimen had retention of their autophagic processes, despite the effects of aging (Donati et al., 2008). One mechanism by which this may occur is through inhibition of the mTOR pathway, which is a cell regulator that promotes cell proliferation and inhibits autophagy during times of sufficient nutrient availability (Mattson et al., 2014). This pathway may be inhibited because the breakdown of molecules into their basic components is not necessary. However, in an environment lacking in nutrients, as it would be during the fasting period of intermittent fasting, the mTOR pathway is inhibited, and autophagy is therefore increased (Mattson et al., 2014). This observed increase in autophagy effectively acts to protect against the naturally occurring effects of aging that occur due to degeneration of cells from decreased autophagy.

Improved Gut Health

An additional effect that intermittent fasting has is improvement in gut health. The gut microbiota composition changes dramatically when intermittent fasting occurs (Wei et al., 2018). This change has been shown to be a positive one. For example, switch flies, who normally die from pathology relating to an abundance of gut bacteria, were found to have a significant decrease in harmful bacteria when on an intermittent fasting regimen (Catterson et al., 2018). Additionally, switch fly pathology normally includes tumor formation and weakening of the gut barrier, but both of these were reduced when flies were placed on an intermittent fasting diet (Catterson et al., 2018). In a human model, an increase in *A. municiphila* and *B. fragilis* has been shown. Both of these bacteria have been shown to decrease in abundance with metabolic disorders like diabetes and obesity, as well as with inflammatory disorders such as irritable bowel syndrome and ulcerative colitis, suggesting these species are related to a healthy gut (Ozukul et al., 2019; Remely et al., 2015). To date, studies showing the effects of intermittent fasting on the gut microbiome are lacking, so more research can be done in this area. However, these preliminary studies suggest that following an intermittent fasting diet can improve gut health.

Longer Lifespan

Another effect that can occur as a result of intermittent fasting is a longer lifespan, though there have been arguable results on this front. It has been shown that a regimen of fasting intermittently increases the lifespan of *C. elegans* by 66.5% (Honjoh et al., 2009). Another supportive finding has been the increase in lifespan for bacteria, yeast, fruit flies, nematode worms, and mice (Catterson et al., 2018). One possible explanation for an increase in lifespan is

a decreased level of pathogenic bacteria in the gut (Catterson et al., 2018). However, it has also been shown that simple caloric restriction increases lifespan (Colman et al., 2014; Weindruch & Sohal, 1997). It does not seem as though there is much research to date that examines the effect of intermittent fasting on lifespan without an overall caloric deficit, so this topic may be explored more in the future. There is a shortage of research on the relationship between lifespan and intermittent fasting in humans, which would likely be due to the length and difficulty with compliance of such a study.

Benefits in Non-Healthy Persons

Intermittent fasting has demonstrated benefits in healthy persons. In addition to these, there were also advantages observed for non-healthy persons. These included an anti-inflammatory effect, improved prognosis with cancer treatment, protection against neurodegeneration, improvement in metabolic disorders, and increased cardiovascular health.

Anti-Inflammatory Effect

One significant benefit found with intermittent fasting is the anti-inflammatory effect it has on the body. For example, inflammation is a major cause of pulmonary dysfunction, specifically asthma. Lung function is quantified using many measurements, one of which is the rate that air can be exhaled, known as peak expiratory flow. Losing weight on its own has been shown to increase peak expiratory flow, and therefore lung function, in asthma patients, but only when there was a weight reduction of 13% or greater. However, in intermittent fasting patients, an increase in peak expiratory flow was shown with only a weight reduction of 4% (Johnson et al., 2007). These results might indicate that the improvement in lung function was because of something other than the weight loss itself; it is possible that the method of weight loss played a

role. The anti-inflammatory effect on the digestive system has previously been discussed. To be discussed in detail further on in this review is the anti-inflammatory effect on multiple other body systems, specifically the responses in both the cardiovascular and nervous systems. These findings give evidence for a body-wide anti-inflammatory effect in response to intermittent fasting, and one possible mechanism for how this occurs is through the reduction of oxidative stress.

It has previously been established that long-term oxidative stress causes activation of inflammatory pathways (Reuter et al., 2010). It has been shown that there are decreases in oxidative stress markers in patients that participate in an intermittent fasting regimen as compared to patients who consistently undergo caloric restriction (Harvie et al., 2011). These patients had an overall similar caloric restriction, so the decrease in oxidative markers was not due to weight loss. It is possible that a reason for the anti-inflammatory effect demonstrated in intermittent fasting models is due to a reduction of oxidative stress. This may be related to the fact that cells are already put under some amount of stress during periods of intermittent fasting, and they react to this stress by gearing up to protect against more stress. For example, corticosterone is a glucocorticoid that has damaging effects on the entire body system when at high levels for prolonged periods of time due to stress. Corticosterone is also secreted in higher levels during intermittent fasting. However, when it is secreted during the latter condition, it actually leads to downregulation of glucocorticoid receptors (Lee et al., 2000; Figure 1). When these receptors are downregulated, cells are better able to protect themselves against stress, since they are less sensitive to the presence of glucocorticoids. It has also been shown that intermittent fasting causes an increased production of antioxidant enzymes and protein chaperones (Martin et

al., 2006). The antioxidant enzymes act to change reactive oxygen species in the body to non-toxic forms, reducing oxidative stress, so a higher number of such enzymes in the body would protect against cellular damage. Protein chaperones work to promote proper folding of proteins, while also aiding in the degradation of impaired proteins, so a higher number of these would help to regulate bodily processes.

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Figure 1. Glucocorticoid receptor type II mRNA levels, type I (MR) and type II (GR), are measured in different parts of the brain in ad libitum (AL) mice versus every other day intermittent fasting (DR) mice. DR decreased levels of GR mRNA in all regions of the brain, while DR decreased levels of MR mRNA in some regions of the brain and increased levels of MR mRNA in other regions of the brain. Retrieved from "Dietary restriction selectively decreases glucocorticoid receptor expression in the hippocampus and cerebral cortex of rate," by J. Lee, 2000, *Experimental Neurology, 166.* Copyright [2000] by Academic Press.

Roles for Intermittent Fasting in Cancer

There is also some evidence that another benefit of intermittent fasting is the effect it produces in patients undergoing cancer treatment. In one experiment, intermittent fasting conditions were simulated in *Saccharomyces cerevisiae* for 24 hours for both wild-type *S. cerevisiae* cells as well as those that had an oncogene mutation in Ras, $RAS2^{val19}$. When exposed to solutions that would create oxidative stress, such as hydrogen peroxide, the wild-type "fasted" cells were able to better survive against oxidative stress as compared to those that had not been in fasting conditions, while the $RAS2^{val19}$ mutated cells had a lower survival rate than the wild-type cells (Lee et al., 2013). Chemotherapy drugs are also oxidatively stressful to the body, and this research shows that intermittent fasting conditions in *S. cerevisiae* caused normal cells to become "protected" from oxidative stress, while cancerous-like cells were less protected and more prone to death. Another study supported this finding, showing that normal *S. cerevisiae*

cells were protected against oxidative stress in the form of chemotherapy, up to one thousand times that of cancerous cells when in an environment simulating short-term fasting conditions (Raffaghello et al., 2008). This provides a possible model for enhanced treatment of cancer in combination with chemotherapy drugs.

In a mammalian mouse model, it was found that proliferation levels of cancerous cells including breast cancer, neuroblastoma, glioma, and ovarian cancer were decreased on an intermittent fasting regimen when treated with chemotherapeutic drugs. The growth of these cells was preferentially slowed over wild-type cell proliferation, more so than it was with mice on an ad libitum diet (Lee et al., 2013). This finding is supported by other evidence that shows the same anti-proliferative effect of intermittent fasting on abnormal cell growth, even at different levels of fat intake during feeding periods, which might make this diet easier for humans to adhere to (Varady et al., 2009). During intermittent fasting, ovarian and blastoma tumors show a slower growth rate, but this effect is discontinued after the fasting period has finished (Lee et al., 2013). This finding may represent the inability of intermittent fasting on its own to provide lasting effects, but the results of intermittent fasting in conjunction with chemotherapeutic drugs seem to be the most effective. For example, there is evidence of a higher susceptibility to and rate of death in cancer cells when on an intermittent fasting regimen with treatment of the chemotherapeutic drugs chlorambucil-prednisone and doxorubicin (Lee et al., 2013). As it pertains to tumor size, it has been shown that one 48-hour round of fasting in melanoma and glioma mice reduces tumor size as much as two rounds of chlorambucil prednisone alone; however, it is again noted that the best effect was achieved with a combination of both fasting and the drug mixture (Figure 2). A particularly notable finding was the

combination of intermittent fasting with doxorubicin in breast cancer mice, which caused a slowed tumor growth rate as seen in other cancer types, but after the fast and drug administration was over, the tumor stopped growing altogether. Additionally, it has been shown that mice are able to handle a larger dose of the doxorubicin while fasting, up to a quantity that would normally cause death (Lee et al., 2013). These results may demonstrate a role for fasting while patients are on chemotherapy to help stave off tumor growth, while also allowing the patient to undergo chemotherapy in larger doses. This may lead to a better outcome, but may also cause toxicity in the patient.

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Figure 2. Effect of fasting on tumor progression as a percent of initial tumor size in murine breast cancer (4T1) tumors on an ad libitum diet (Control), intermittent fasting diet (Fasted), chemotherapy administration (CP), and a combination of an intermittent fasting diet and chemotherapy administration (Fasted/CP). Retrieved from "Fasting cycles retard growth of tumors and sensitize a range of cancer cell types to chemotherapy," by C. Lee, 2013, *Science Translational Medicine 4*(124), 124-127. Copyright [2012] by the American Association for the Advancement of Science.

One possible reason for the protection against tumor growth that is exhibited by patients undergoing an intermittent f g regimen is the ketogenic effect. The ketogenic effect is a bodily process observed where cells turn to using ketones as a source of fuel, which are created from fatty acids. This state is called ketosis, and it is normally caused by an increase of bodily ketones as a result of low carbohydrate availability and high fatty acid availability, as fatty acids are converted to ketones and then used to create ATP for energy. One study showed that intermittent fasting increased levels of fat metabolism as a source of energy (Shin et al., 2018). This is supported by the fact that during intermittent fasting, there is an increase of the ketone β-hydroxybutyrate (Anson et al., 2003). It has also been shown that cancerous cells mainly use

glucose as a source of energy (Seyfried et al., 2014). Because there is an increase in bodily ketones with intermittent fasting, this may deprive cancerous cells of their preferred form of energy, and therefore inhibit tumor growth.

Neuronal Protection

There have been several previously outlined effects of intermittent fasting on the protection of neurons. Neuronal damage can occur from age-related illnesses such as Alzheimer's disease and dementia, and can also result from Down syndrome, multiple sclerosis, or ischemic stroke. In the former conditions, this damage can be due to demyelination, or breakdown of the myelin sheath surrounding nerve cells, which eventually leads to loss of neuronal function. It can also be due to inflammation in the brain caused by pro-inflammatory cytokines, which are present at higher levels in individuals with the beforementioned conditions and have degenerative effects on cognitive function (Shin et al., 2018; Vasconcelos et al., 2014). The protective effect of intermittent fasting against the degeneration described has been demonstrated in many studies. For example, kainic acid is an excitotoxin, a chemical that has an excitatory effect on neurons and eventually leads to their damage. When rats on an alternate-day fast were injected with kainic acid, there was an increased rate of neuronal survival as compared to mice on regular diets as well as mice on a caloric restriction injected with the same chemical (Anson et al., 2003; Figure 3). The fasting rats had an overall similar caloric intake as compared to ad libitum mice, so this protective effect on neurons was not due to an overall lowered consumption of calories. Another study showed that Alzheimer's-induced mice on an intermittent fasting diet had better cognitive function as compared to Alzheimer's-induced mice on an ad libitum diet, specifically improving their short-term and spatial memory (Shin et al.,

2018). Yet another study showed that the cognitive effects of a bacterial infection, which causes cognitive dysfunction, were overcome through an intermittent fasting regimen (Vasconcelos et al., 2014; Figure 4). The neurologic benefits of intermittent fasting as a protective measure against various illnesses are manifold, and there are many proposed mechanisms by which these effects may be carried out.

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Figure 3. Mice on three different diets for 20 weeks were injected with the excitotoxin kainic acid into two separate regions of the brain (CA1 and CA3) and were also injected with PBS on the contralateral side as a buffer. Here is shown a comparison between mice on an ad libitum (AL) diet, an intermittent fasting (IF) diet, and a limited daily feeding (LDF) diet. Pair-fed (PF) mice were used as a control against the IF group. Evidenced is the higher percentage of undamaged neurons in the IF group as compared to all others injected with kainic acid. Retrieved from "Intermittent fasting dissociates beneficial effects of dietary restriction on glucose metabolism and neuronal resistance to injury from calorie intake," by M.R. Anson, 2003, *Proceedings of the National Academy of Sciences of the United States of America 100*(10), 6216-6200. Copyright [2003] by the National Academy of Sciences.

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Figure 4. Comparison of mice with various treatments: control which was fed ad libitum and injected with saline solution; LPS which was fed ad libitum and injected with LPS, a compound that induces bacterial infection; IF which was fed every other day and injected with saline; and IF+LPS which was fed every other day and injected with LPS. This comparison shows the rate at which each group made their way through a maze over five trial days. The findings demonstrate that IF+LPS had a faster time each day compared to LPS ($p < 0.001$), which showed higher cognitive capabilities for IF+LPS than LPS. IF also showed a faster time each day as compared to the control ($p < 0.01$). Retrieved from "Intermittent fasting attenuates lipopolysaccharide-induced neuroinflammation and memory impairment," by A. R. Vasconcelos, 2014, *Journal of Neuroinflammation 11*(85). Copyright [2014] by Springer Nature.

Anti-Inflammation. One suggested mechanism for why neuronal protection occurs is related to the previously mentioned anti-inflammatory effects of intermittent fasting. In the brain, this anti-inflammatory effect leads to the inhibition of pro-inflammatory cytokines. In a study investigating ischemic stroke, which results in neuron cell death, it was found that intermittent

fasting decreases inflammasome activity (Fann et al., 2014). Inflammasomes are complexes of proteins that are involved in inflammation, and two specific types, NLRP1 and NLRP3, are active during ischemic stroke and promote neuronal death. These two complexes in particular had lowered activity in mice that participated in intermittent fasting (Fann et al., 2014). This suggests that a possible mechanism through which neurons are protected is through the decreased activity of these inflammasome complexes.

In other studies, it has been shown that specific anti-inflammatory molecules are upregulated during intermittent fasting. A cytokine called IL-10 hinders the creation of inflammatory cytokines and is found in greater abundance after a period of intermittent fasting (Vasconcelos et al., 2014; Faris et al., 2019; Figure 5). This same cytokine was also present in decreased amounts in those with prion disease, a condition in which a misfolded protein called a prion causes others to misfold, leading eventually to chronic neurodegeneration (Thackray et al., 2004). It would therefore seem that IL-10 also plays a role in impeding neurodegenerative diseases and may be one of the ways that intermittent fasting causes a protective effect on the brain.

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Figure 5. Comparison of mice with various treatments: control which was fed ad libitum and injected with saline solution; LPS which was fed ad libitum and injected with LPS, a compound that induces bacterial infection; IF which was fed every other day and injected with saline; and IF+LPS which was fed every other day and injected with LPS. This comparison shows the differing levels of IL-10 in the hippocampi of rats after thirty days of each treatment. The IF group showed a greater amount of IL-10 as compared to the control group ($p < 0.01$). Retrieved from “Intermittent fasting attenuates lipopolysaccharide-induced neuroinflammation and memory impairment,” by A. R. Vasconcelos, 2014, *Journal of Neuroinflammation 11*(85). Copyright [2014] by Springer Nature.

Another possible way that inflammation is decreased during intermittent fasting is through the reduction in oxidative stress. In the peripheral nervous system, aging is related to an increase in production of reactive oxygen species, which can lead to degeneration by impacting DNA and other important cellular components. These age-related symptoms can lead to peripheral neuropathy, where neurons appear microscopically to have lost significant amounts of their myelin sheaths as a result of this oxidative stress (Lee & Notterpek, 2012). As already described, intermittent fasting has been shown to increase the production of antioxidant enzymes, so this effect may also contribute to the protective effect of intermittent fasting through a reduction in oxidative stress in neurons.

Neurotrophic factors. As another possible mechanism, it has been shown that intermittent fasting increases the amount of neurotrophic factor in the brain, which is a molecule that supports neuronal growth (Lee et al., 2002). This is supported by a finding in another study where intermittent fasting was found to increase the amount of neurotrophic factor, which promoted neurogenesis as well as development and enhancement of neuronal synapses (Mattson et al., 2014). Yet another study showed that in the presence of bacterial infection, which normally lowers neurotrophins, intermittent fasting counteracted this decrease and maintained neurotrophin levels (Vasconcelos et al., 2014). A particularly significant neurotrophic factor is BDNF, or brain-derived neurotrophic factor, which has been shown to counteract the effects of damage due to excitotoxin administration and is also shown in higher quantities in the brain during intermittent fasting (Duan et al., 2008). This may be another cause for the protective effect of this diet.

Prevention of defective protein aggregation. As previously discussed, demyelination of nerve cells occurs as a result of old age, and intermittent fasting acts to prevent this myelin breakdown in three main ways. Damaged or misfolded proteins tend to accumulate around the myelin sheath with old age, and this causes disruption in signaling needed to initiate myelin repair that normally occurs often in neurons. If this maintenance is inhibited for a prolonged period of time, demyelination occurs. As a result of intermittent fasting, the defective protein aggregates are broken down through the ubiquitin-proteasome system, as well as through the beforementioned upregulation in protein chaperones (Lee & Notterpeck, 2012; Martin et al., 2006). In addition, the increase in autophagy associated with intermittent fasting, as described above, may also be responsible for this protective effect on neurons, as the defective proteins can also be cleared through autophagy (Bergamini & Gori, 1995). In these ways, regular maintenance of myelin can continue in older aged persons at a better rate when on an intermittent fasting regimen.

Ketogenic effect. As outlined above, there have been heightened levels of ketones observed in intermittent fasting models (Anson et al., 2013). In addition to being beneficial for cancer patients, this may also prove beneficial for people experiencing ischemic stroke. During this event, neurons are deprived of oxygen, and they are subsequently unable to undergo cellular respiration and ATP production. However, studies have shown that if ketones are present in abundance, there is an increase in mitochondrial activity during ischemia (Tai et al., 2008). This means that the neurons can still perform their needed activities for a longer amount of time, due to the increase in energy reserves caused by the higher presence of ketones. This may act to protect neurons from damage during ischemia.

Counteractive Effect of Obesity and other Metabolic Disorders

In patients with metabolic disorders such as obesity and diabetes, there are associated increases in variants such as blood pressure, insulin resistance, glucose serum levels, cholesterol levels, and triacylglycerol levels. Intermittent fasting showed improvement in each of these indicators of disease (Mattson et al., 2014). However, it has also been shown that weight loss has the same effects completely independent of intermittent fasting, so the question remains as to whether this effect can be attributed to the time between meals or the weight loss that is brought on by caloric deficit. There is some evidence that the frequency of meals itself is the cause. For example, mice with induced type 2 diabetes showed lower glucose serum levels and prevention of β cell loss when on an intermittent fasting diet. Those with induced type 1 diabetes showed an increase in the amount of β cells present, and both of these subjects had a similar weight to ad libitum mice throughout the 8-week experiment (Wei et al., 2018). Another experiment showed that mice held on an intermittent fasting diet with no significant change in weight had increased levels of both blood glucose and insulin (Anson et al., 2003; Figure 6). The decrease in these biomarkers was even more pronounced in intermittently fasted mice who had sustained body weight as compared to mice on a 40% caloric restriction who lost body weight. These results seem to suggest that intermittent fasting works independently of caloric restriction to provide improvement to various biomarkers for metabolic disorders.

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Figure 6. A comparison of glucose concentration (left) and blood insulin levels (right) in three groups: mice fed ad libitum (AL) for 20 weeks, mice held on an intermittent fasting (IF) diet eating every other day for 20 weeks, and mice held on a 40% caloric restriction (LDF) for 20 weeks. Retrieved from “Intermittent fasting dissociates beneficial effects of dietary restriction on glucose metabolism and neuronal resistance to injury from calorie intake,” by M.R. Anson, 2003,

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As to weight loss itself on an intermittent fasting diet, there is also the question of whether the loss is due to caloric restriction or to the intermittent fasting regimen. Some evidence shows that the answer is the latter. For example, one study demonstrated that a regimen of restricted feeding for only eight hours a day in mice protected them against obesity (Hatori et al., 2012). In a human model, women on a six-month intermittent fasting regimen with overall 25% calorie reduction lost an average of 6.5 kg, while women on a 25% caloric restriction consistently for six months lost an average of 5.7 kg (Harvie et al., 2011). Overall, it seems that intermittent fasting itself may provide a slight edge in weight loss and/or preventing weight gain. The weight loss caused by intermittent fasting, in combination with the positive effects on obesity indicators caused by fasting without overall weight loss, may work together to mitigate the effects of metabolic disorders.

Beige fat cell activation leads to weight loss. One mechanism as to how intermittent fasting helps attenuate the effect of metabolic disorders is through the activation of beige fat. Brown adipose tissue is known to use more energy than white adipose tissue and is able to convert this energy to heat. This tissue is largely inactive in human adults and mostly occurs in youth. However, there is a type of tissue called inducible thermogenic adipose tissue, or beige fat, which is a modified form of white fat tissue that mimics the energy expenditure of brown adipose tissue (Wu et al., 2013). There have been ongoing studies to investigate the ways that this modification to beige fat can occur, and one of them may be through intermittent fasting. The main cause of heat production and increased energy expenditure in brown and beige fat is the activity of uncoupling protein 1 (UCP1). One study showed that an alternate-day fasting

regimen in mice resulted in the formation of beige fat from white adipose tissue (Li et al., 2017). This was evidenced by an increase in core body temperature as compared to ad libitum mice, showing that thermogenesis was more active in intermittent fasting mice. The formation of beige fat was also evidenced by an increase in activity of UCP1 in the white adipose tissue of mice after their fasting regimen (Figure 7). As previously discussed, the composition of the gut microbiome is changed on an intermittent fasting regimen. When the microbiota from mice with an intermittent fasting regimen were transplanted into mice with depleted gut microbiota, UCP1 was present in higher abundance as compared to transplantation of microbiota from ad libitum mice. However, placing microbiota-depleted mice on an intermittent fasting regimen did not increase beiging as much, suggesting that the gut microbiome shift incurred during intermittent fasting was responsible for the beiging effect found during intermittent fasting (Li et al., 2017). With this possible mechanism of transforming white adipose tissue into beige cells, those with obesity could participate in intermittent fasting and use more energy than they intake, causing weight loss independent of caloric restriction.

Figure 7. UCP1 immunohistochemical staining of white adipose tissue of mice on an ad libitum diet after 15 days (left) versus staining of white adipose tissue of mice on an alternate-day diet after 15 days (right). Retrieved from "Intermittent fasting promotes white adipose browning and decreases obesity by shaping gut microbiota," by G. Li, 2017, *Cell Metabolism 26*, 672-685. Copyright [2017] by Elsevier Inc.

Visceral fat. One notable research finding was the selective decrease in visceral fat that has been observed on an intermittent fasting diet (Shin et al, 2018). An increase in visceral fat occurs with aging and has been associated with increased insulin resistance and inflammation, and is also a risk factor for developing metabolic disorders (Xiuquan et al., 2015). This decrease

in abdominal fat due to intermittent fasting may be another way that this diet regimen lessens the effects of metabolic disorders.

Muscle retention. Intermittent fasting may also be a way to lose fat while simultaneously retaining muscle mass, which is a difficult thing to do on a basic caloric-restriction diet, where weight loss normally comes from both muscle and fat (Winston et al., 2018). Interestingly, it has been shown that mice on high fat and sugar diets that participate in only intermittent fasting gain muscle mass, though only slightly, while at the same time losing fat mass. In comparison, mice on this same diet that did not partake in intermittent fasting, but did participate in high intensity interval training (HIIT) exercises, gained both fat mass and lean muscle mass (Wilson et al., 2018; Figure 8). While losing weight is a difficult undertaking and almost always leads to loss of both fat and muscle, intermittent fasting may provide a possible way to retain that muscle while simultaneously losing fat mass.

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Figure 8. Comparison of body composition of mice from four different groups, each fed high fat and sugar-water diets of similar caloric intake levels: CON which was fed this diet only; HIIT which was fed this diet and participated in HIIT exercises; IF which was fed this diet and fasted one day per week; and IF+HIIT which was fed this diet, fasted one day per week, and participated in HIIT exercises. Retrieved from "Intermittent fasting with or without exercise prevents weight gain and improves lipids in diet-induced obese mice," by R. A. Wilson et al., 2018, *Nutrients 10*(3): 346. Copyright [2018] by the authors.

Cardiovascular Benefits

Intermittent fasting has shown a host of benefits for the cardiovascular system. For example, there has been lower observed levels of overall cholesterol, LDL cholesterol, triacylglycerols, blood pressure, and heart rate with intermittent fasting, all of which are risk factors for multiple cardiovascular diseases (Klempel et al., 2013; Wan et al., 2013). Another

example is the protection against myocardial infarction. This event occurs when there is a blood clot that inhibits flow to the heart, causing lack of oxygen, or ischemia, to the tissue. If this occurs, the tissue becomes damaged and heart function is stunted, which may result in mortality. It has previously been shown that rats on an intermittent fasting diet for three months exhibited less heart tissue damage after a myocardial infarction was induced as compared to rats on an ad libitum diet (Ahmet, 2005). One possible mechanism explaining this is through the observed increase in adiponectin levels, which is a protein that has been shown to protect against myocardial damage (Wan et al., 2010). In addition to the cardiovascular benefits noted when intermittent fasting is performed prior to an ischemic event, there is evidence that intermittent fasting can also be beneficial when started following an ischemic event. Vascular endothelial growth factor, or VEGF, is a biomolecule that promotes blood vessel growth and increases survival chances after ischemia. It has been shown that after intermittent fasting following myocardial infarction, there was a higher expression of VEGF over ad libitum mice (Katare et al., 2009). With increased VEGF there is an increase in angiogenesis and therefore a decrease in mortality after ischemia, so these results may show benefits of intermittent fasting both before and after an ischemic event. Another benefit to note is protection against hypertrophy of the myocardium, a normal result of myocardial ischemia (Katare et al., 2009; Figure 9).

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Figure 9. This graph shows the cross-sectional area of myocytes resulting from hypertrophy of four groups: sham operated and normally fed (SO-NF), sham operated and undergoing intermittent fasting (SO-IF), myocardial ischemia and normally fed (MI-NF), and myocardial ischemia and undergoing intermittent fasting (MI-IF). Demonstrated is the smaller cross-sectional area, and therefore smaller amount of hypertrophy, seen in MI-IF mice as compared to MI-NF mice. Retrieved from "Chronic intermittent fasting improved the survival following large myocardial ischemia by activation of BDNF/VEGF/PI3K signaling pathway," R.G. Katare et al., *Journal of Molecular and Cellular Cardiology,*

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There is also some damage caused by an increase in production of reactive oxygen species during myocardial ischemia, subsequently leading to inflammation and further tissue damage (Kurian et al., 2016). Cardiomyocytes in those mice who underwent intermittent fasting following a myocardial infarction showed lower levels of oxidative stress than ad libitum mice (Katare et al., 2009). This is yet another example of the antioxidant and anti-inflammatory effects of intermittent fasting.

Disadvantages

With all of these possible benefits of intermittent fasting, studies have shown some potentially harmful side effects. For example, the restriction of intermittent fasting may cause infertility in young rats, decreasing reproductive abilities. It has been shown that female mice fasted intermittently have disturbed reproductive cycles, and male mice fasted intermittently have lower levels of testosterone (Kumar & Kuar, 2013). However, this was shown when there was reduction in overall energy intake. There has actually been other evidence to show that it can improve fertility in women with polycystic ovary syndrome, which is related to high glucose and insulin levels and may be improved by the reduction of these markers with intermittent fasting (Chiofalo et al., 2017). However, there have not been studies relating to infertility and intermittent fasting in otherwise healthy test subjects that had no overall caloric deficit with intermittent fasting. Future studies in this area would be necessary to determine if there is risk for infertility solely from intermittent fasting without a change in overall caloric intake in healthy test subjects.

There is also some evidence that intermittent fasting leads to bone density loss, which is a major concern for the elderly, as bone density is naturally lost with age and can lead to osteoporosis and increased risk of fractures. In one study, mice on a four-day fast lost bone density, and though some was gained back, they did not return to the same bone density as their ad libitum counterparts through the end of the 14-day study (Hisatomi & Kugino, 2019). However, this experiment did show that there was an overall decrease in caloric intake over the period of the study in the experimental mice as compared to control mice, so it is unclear as to whether this bone density loss is from the intermittent fasting regimen itself or from the weight loss that occurred as a result of a caloric deficit. While this may pose a true risk for those who participate in intermittent fasting with an overall caloric deficit, future studies would be necessary to determine the risk for bone density loss during an intermittent fast without an overall caloric deficit. One study on humans during Ramadan, a daily 15-hour fast that occurs for 28 days, showed an increase in bone health by measuring various markers of bone metabolism, which were shown to be elevated (Bahijri et al., 2015). This study did not directly measure bone density though, so there is still further research that could be done in this area.

Another possible disadvantage to intermittent fasting is the fatigue associated with periods of fasting. This eating regimen can be undertaken without a caloric deficit, but when there is lower overall energy intake, there can be adverse performance effects, particularly for athletes. One study found that the Ramadan intermittent fast demonstrated a decrease in athletic performance for regular athletes, but no significant difference for experienced athletes, who were able to continue consuming a sufficient amount of calories necessary for maintaining their activities (Chaouachi et al., 2009). For those who are desiring to perform an intermittent fasting

regimen, there may be a need for caution if there is a caloric restriction when performing physical activity, or even any daily activity, just as there is a need for caution in this area with a regular calorie-restricting diet.

To note is also the possible difficulty of adherence to such a diet. Some may find this regimen easier to undertake, as there is room to eat high-fat food while in the feeding stage and still receive some of the aforementioned benefits (Varady et al., 2009). However, some may find this more challenging, as it requires much control to refrain from eating for a period of time.

Conclusion

In conclusion, this paper provided an overview of the benefits and disadvantages of intermittent fasting. The numerous advantages evidenced seem to be beneficial for a body-wide effect, including protection to the nervous, cardiovascular, respiratory, and digestive systems. While there may be possible disadvantages, more research is needed to determine what effect solely intermittent fasting has on both bone mineral density and the reproductive system. It is also necessary to proceed carefully with this diet regimen, as it is necessary to do with all others. It would seem from this review that, while fasting intermittently was first introduced and instructed as a spiritual practice, there are many underlying benefits that occur within the human body during a period of fasting.

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