

Institute for Christian Teaching
Education Department of Seventh-day Adventists

**INSULIN AND MITOCHONDRIAL FUNCTIONS:
IMPLICATIONS ON FASTING AND REFLECTIONS ON
SABBATH KEEPING**

by

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Prepared for the
38th International Faith and Learning Seminar
Held at
Loma Linda University
July 2008

Introduction

In retrospect, I would like to make it known that I am a professor of applied biochemistry and molecular biology and is employed in a state university but a Seventh Day Adventist convert way back in 1974. As a professor with background in human physiology and one who had been involved in lecturing and doing research in these two intertwined fields in science (biochemistry and molecular biology), I always try to find meanings on the things that I believe - from the subjects that I have learned and taught all through these years. Teaching several courses in cell/molecular biology, genetics, physiology, radiation biology, biochemistry, and microbiology have enabled me to integrate principles of life and reflecting or applying them to some fundamentals of Christian faith like the practice of fasting coupled with the observance of Sabbath and prayer. The presentation attempts to find some benefits of fasting and extending it to the principle of Sabbath keeping and relate it to the longevity of cells, wholesome lifestyle and possible lengthening of lifespan.

The framework which I want to present here is that there is sound scientific basis for fasting. In the succeeding discussions, I have presented an experiment on nematode worm about reduction of sugar or carbohydrate intake which resulted to 100% increase in lifespan. This result is so significant in our healthy lifestyle as Seventh-Day Adventists. When we fast, the cells are also resting (*Sabbath*), not in the absolute sense of stopping cellular activities, but rather on trying to slow down metabolic activities that would give time for the healing of the cell itself. Lines of evidence pointed to an **insulin receptor as the control mechanism for ageing and lifespan**. If we do the fasting on the Sabbath, it will be more meaningful and wholesome as it offers lots of cascading benefits as it is presented in the paper.

Declaration of Faith and Practice

Fasting in connection with prayer, penitence, and preparation for new ventures has been practiced from early times in many cultures and religions. The Bible recognizes it as regular in mourning for the dead (I Sam. 31:13), expression of penitence (Neh. 9:1), intercession (II Sam. 12:16) and prayer for God's aid (Judg. 20:26). Fasting was undertaken for personal reasons (Ps. 25:13), as a national act in the face of calamity (Joel 2:15), or as a periodic liturgical observance (Zech. 8:19); normally it involved abstinence from all food to show dependence on God and submission to his will. Jesus accepted fasting as a natural discipline, and he is described (Matt. 4:2) as deliberately fasting before his temptation and the start of his ministry, similar to the action of Moses (Exod. 24:28). **Isaiah 58:5-7**, outlined correct way of fasting i.e., that if this practice was to be of value, it must be accompanied by compassion and a concern for social justice (Metzger and Coogan, 1993).

As Adventists, we are supposed to *proclaim and live the truth*. This paper focuses on one significant fundamental doctrine among Seventh-Day Adventists, the Sabbath keeping which is declared in *Exodus 20: 8*- "Remember

the Sabbath day by keeping it holy" - and the other is a practice of fasting and prayer by Jesus Christ which is recorded in *Matthew 4: 1-2*. Also in *John 5:33 ... John's disciples often fast and pray...*".

The main purpose of the paper is to find insights and scientific learning that benefits man in keeping God's commandment. I am convinced that behind a command or a practice of faith is also a blessing of health and healing that keeps us more attuned to the voice of God as we worship Him in spirit and in truth.

Objectives:

For the purpose of this presentation, I would like to narrow it down to the following objectives:

- a) To present the basic endocrine function such as the release of insulin and glucagon in response to fluctuations of blood sugar levels in the body;
- b) To present the consequential activation of mitochondrial functions at all target and non-target cells to provide ATPs to keep the cell alive and functional;
- c) To show direct relations between intake of sugar (carbohydrates) and ageing (a revelation from the nematode worm, *Caenorhabditis elegans*);
- d) To show correlations of fasting as rest for the cells at the cellular level: A reflection on the physical benefits of Sabbath keeping;
- e) To show the right possible way of fasting.

A Review of Basic Endocrinology on Sugar or Carbohydrate regulation

Generally, carbohydrates that we take in our diet like rice, starch, and other forms of polysaccharides are first digested into monomer forms like glucose, fructose, and galactose. These monomer units are the only absorbable forms that can be used as the first line energy source that would serve as substrate for the production of ATP (energy). This energy currency inside the cell is very much needed by the cell for all the cellular functions (e.g., transport of molecules, secretions, growth and development). However, the body must have some kind of control mechanism to maintain homeostatic conditions at all times. When monomer sugar is not available, the body then utilizes stored carbohydrates like glycogen and other complex molecules that are stored primarily in the liver and muscles. The liver has at least four (4) hours of energy (ATP) supply, so when a person skips a meal or so, this reserve will be utilized. When the reserve carbohydrates are consumed, the body system shifts to utilizing fats/lipids that are stored in adipose tissues of the body converting them into glycerides and fatty acids which are later on converted into pyruvic acid and acetyl-CoA or the body will synthesize new sugars from non-carbohydrate sources in the process called *gluconeogenesis*. It can also utilize proteins in the body by breaking down complex proteins into amino acids and enter into the Krebs's Cycle. This is the reason why even if a person does not eat, the cells are still provided with the precursors for energy (ATP) production

because of the inter-conversions of metabolic moieties inside the cell. Now, take note that, at the molecular level, when a complex molecule is converted to monomer units, it is always coupled with **hydration**, in the process called hydrolysis performed by the specific enzyme that acts upon a substrate. Meaning, when one does not eat any complex food, **water** is always needed to break down stored complex molecules such as carbohydrates, fats, and proteins into simple monomer units like simple sugars, glycerides, fatty acids and amino acids, respectively. This is a striking revelation to us all. Therefore, the proper way of fasting is to provide the body with water so that the cell could still perform a basal metabolism for its minimal energy production just to be able to survive. (Fig. 4). Therefore, when we do fasting, it is important to drink plenty of water for the body to utilize the reserves efficiently.

One of the endocrine glands involved in cell metabolism is the pancreas (see Figure 1a) particularly the alpha and the beta cells of the *Islet of Langerhans* (see Figure 1b). The alpha cells secrete *glucagon* when blood sugar level is low, a condition termed as **hypoglycemia**, while the beta-cells secrete *insulin* when blood sugar is beyond the normal levels or **hyperglycemia**. Both hormones are peptide in nature and because of that, their actual regulation into the cell requires some receptors that are found in the plasma membrane of any target cell or cells. The succeeding reactions are enzymatic in nature and are mediated first by the membrane proteins (mostly G-proteins) and the cascading reactions performed by different kinases. Example to this is the degradation of glycogen into simple sugar (monomer), usually glucose.

When blood sugar level is high (hyperglycemia), almost immediately, the *Islet of Langerhans* in pancreas will secrete *insulin* in order to maintain normal blood glucose levels in the body and prevent diabetes. These excess sugars are then converted into glycogen that are then stored into the liver and muscles (Fig. 2) The other action of insulin is to activate the pathway converting excess sugars into fats that are then stored in adipose tissues of the body. Now when the conditions are reversed, meaning a person becomes hypoglycemic (low blood sugar) the stored glycogen in the liver and muscles as well as fats are then converted back to the monomer sugar that the cell requires for ATP synthesis. This process is controlled by the peptide hormone glucagon that are secreted by the alpha cells of the Islets of Langerhans in pancreas (Fig. 3). These two opposing hormones play a very important homeostatic mechanism at the organ or systemic levels.

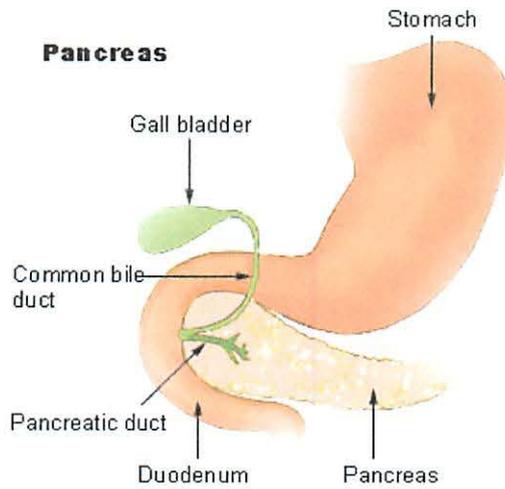
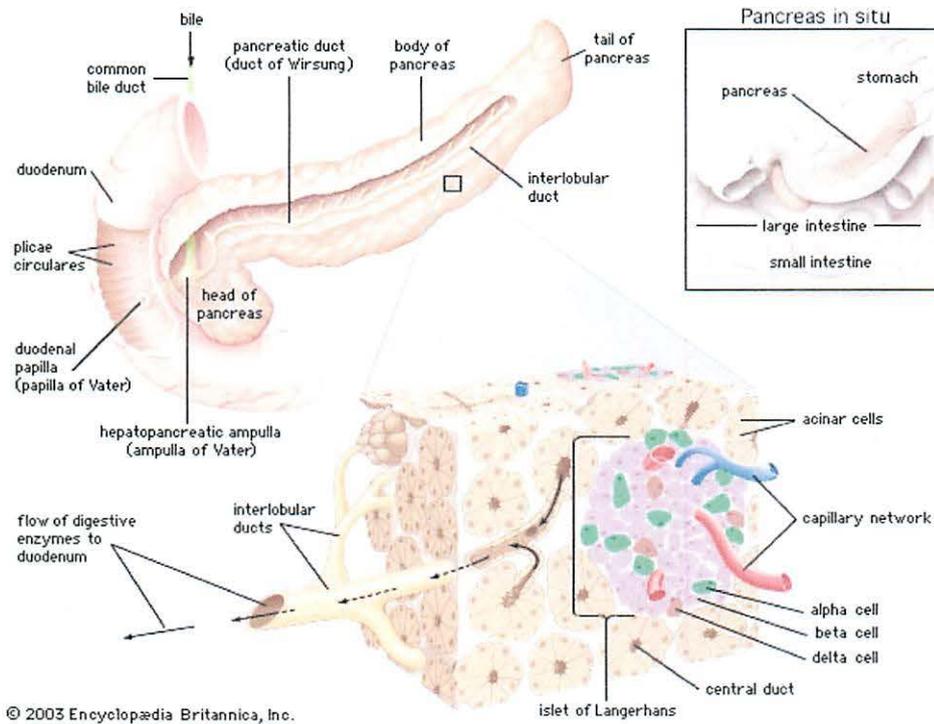
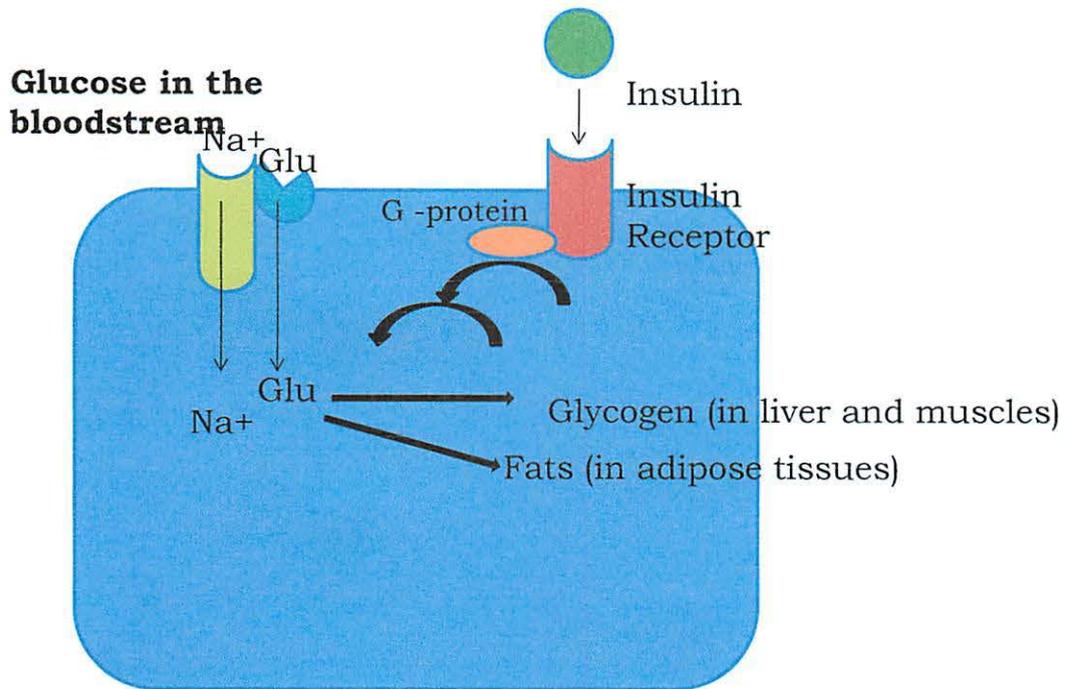


Fig. 1a Relative Location of Pancreas in the body cavity



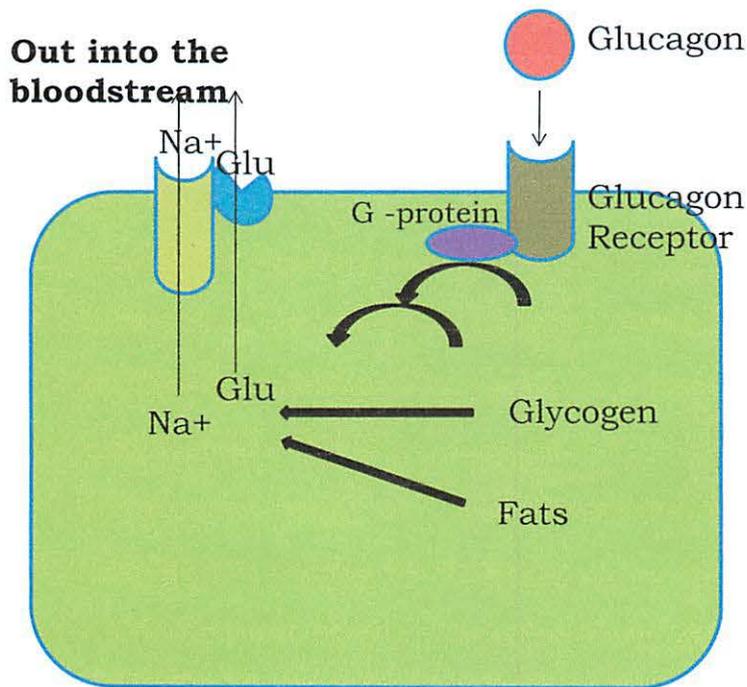
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Fig. 1b. The Islets of Langerhans in the Pancreas



Insulin Action when sugar is High (Hyperglycemia)

Figure 2. Action of Insulin in target cells



Glucagon Action when sugar is Low (Hypoglycemia)

Fig. 3. Action of Glucagon at target tissues/cells

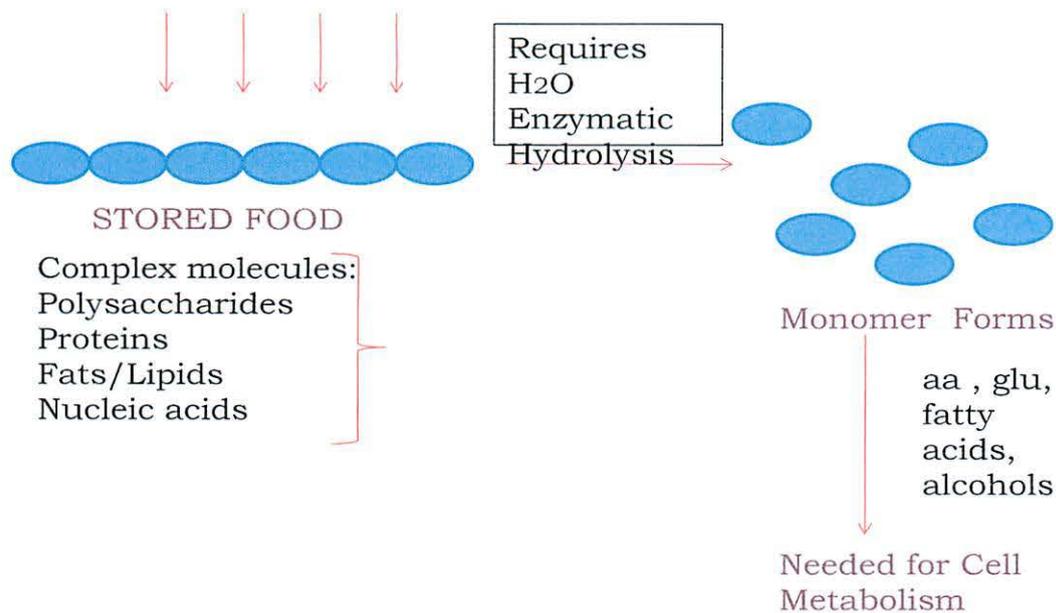


Fig. 4. Consequences when one is Fasting

So, therefore as we view the cell that way, the key role of insulin and glucagon are so important in maintaining a balance. On the other hand, after the cell has been provided with lots of monomer sugar, the mitochondria will now work in complex-coupled reactions, primarily to produce energy that the body needs.

A Review on the Functions of Mitochondria

The main function of the mitochondrion (Fig. 5) is the production of energy in the form of adenosine triphosphate (ATP). The cell uses this energy to perform the specific work necessary for cell survival and function. It is observed that a cell with more mitochondria indicates more metabolic activities are done on these sites (e.g. detoxifications, energy production, control of growth and metabolism).

The raw materials used to generate ATP are the foods that we eat. The tissues within the body utilize these complex food precursors (carbohydrates, fats/lipids, proteins and nucleic acids) are broken down into simpler and absorbable forms (monomer units) in a process called *catabolism*. These molecules are then transferred into the mitochondria, where further processing occurs. Reactions within the mitochondria produce specific molecules that can have their electrical charges separated within the inner mitochondrial membrane. These charged molecules are processed within the five electron transport chain complexes to finally combine with oxygen to make ATP. The

process of the charged substances combining with oxygen is called *oxidation*, while the chemical reaction making ATP is called *phosphorylation*. The overall process is called *oxidative phosphorylation*. The product produced by this process is ATP.

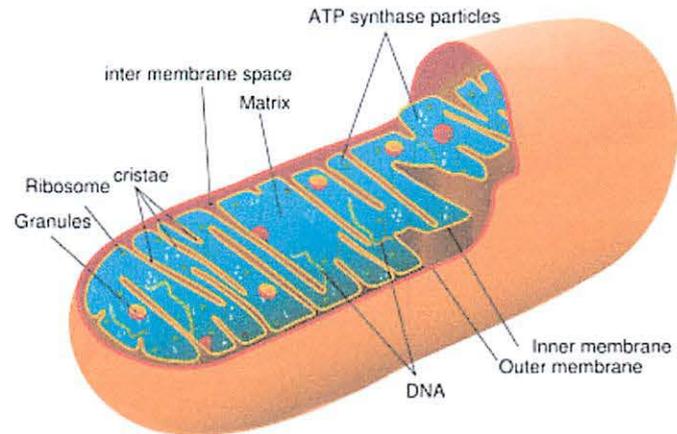


Fig. 5. The structure of a mitochondrion (with its parts)

Mitochondria (singular **mitochondrion**) are the source of energy production within a cell. They are semiautonomous and self producing organelles, residing in the cytoplasm of eukaryotic cells. Converting cellular energy metabolites in the Krebs Cycle, through the process of oxidative phosphorylation they produce adenosine triphosphate (ATP) which is used to power other processes in the cell (Fig. 6 and 7).

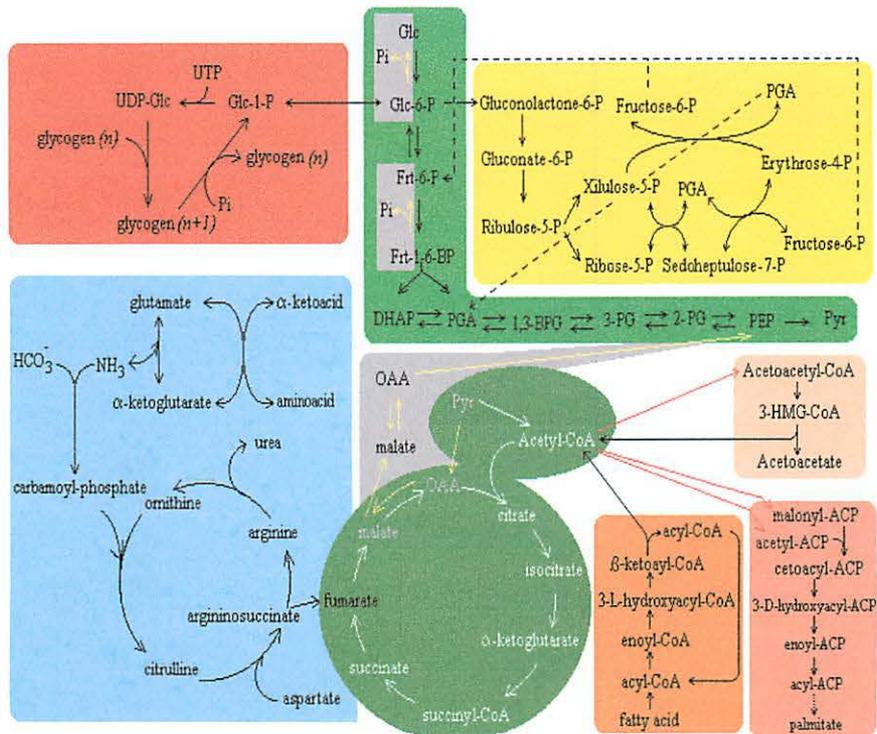


Fig. 6. Metabolic pathways showing Glycolysis to Krebs's Cycle

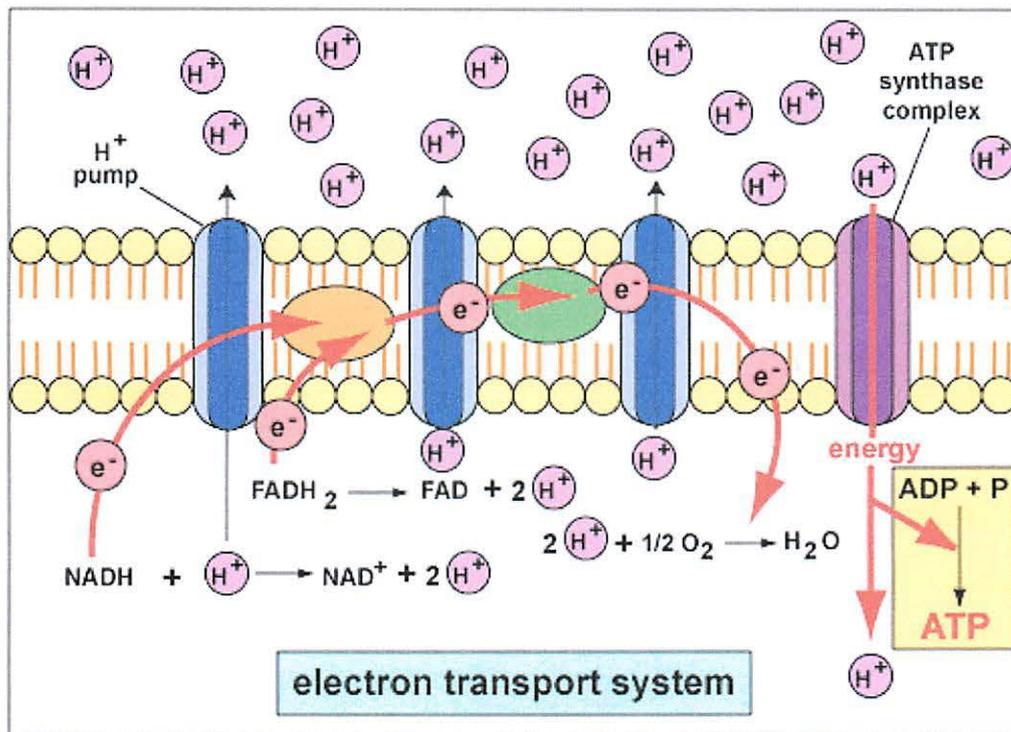


Fig. 7: ATP Production during Aerobic Respiration by Oxidative Phosphorylation involving an Electron Transport System and Chemiosmosis

Normally, ATP production is coupled to oxygen consumption. The mitochondria become partially “uncoupled” and produce highly reactive oxygen species called free radicals. When the production of free radicals overwhelms the mitochondria’s ability to “detoxify” them, the excess free radicals damage mitochondrial function by changing the mitochondrial DNA, proteins, and membranes. As this process continues, it can induce the cell to undergo apoptosis. Abnormal cell death due to mitochondrial dysfunction can interfere with organ function. Cell death can occur either by injury due to toxic exposure, by mechanical damage, or by an orderly process called *programmed cell death* or *apoptosis*.

At this point this is something that we must take note, with increasing sugar intake, mitochondria are then busy doing its catabolic functions so much so that it may also cause its rapid deterioration and hence led to apoptosis and death to the cell. Thus, rapid mitochondrial functions led to shorter life span and rapid aging.

During fasting, the cells are then rested because there are no new sugars that are synthesized or are provided from the digestion of carbohydrates. The cell then undergoes rejuvenation, repair, and prevents generation of free radicals which are causing death of mitochondria and the cell itself as a whole. The practice of fasting results to longer lifespan for mitochondria, to the cell and the whole body itself.

Other functions of mitochondria are related to the cell type in which they are found. Mitochondria are involved in building, breaking down, and recycling products needed for proper cell functioning. For example, some of the building blocks of DNA and RNA occur within the mitochondria. Mitochondria are also involved in making parts of blood and hormones such as estrogen and testosterone. They are required for cholesterol metabolism, neurotransmitter metabolism, and detoxification of ammonia in the urea cycle. Thus, if mitochondria do not function properly, not only energy production but also cell-specific products needed for normal cell functioning will be affected.

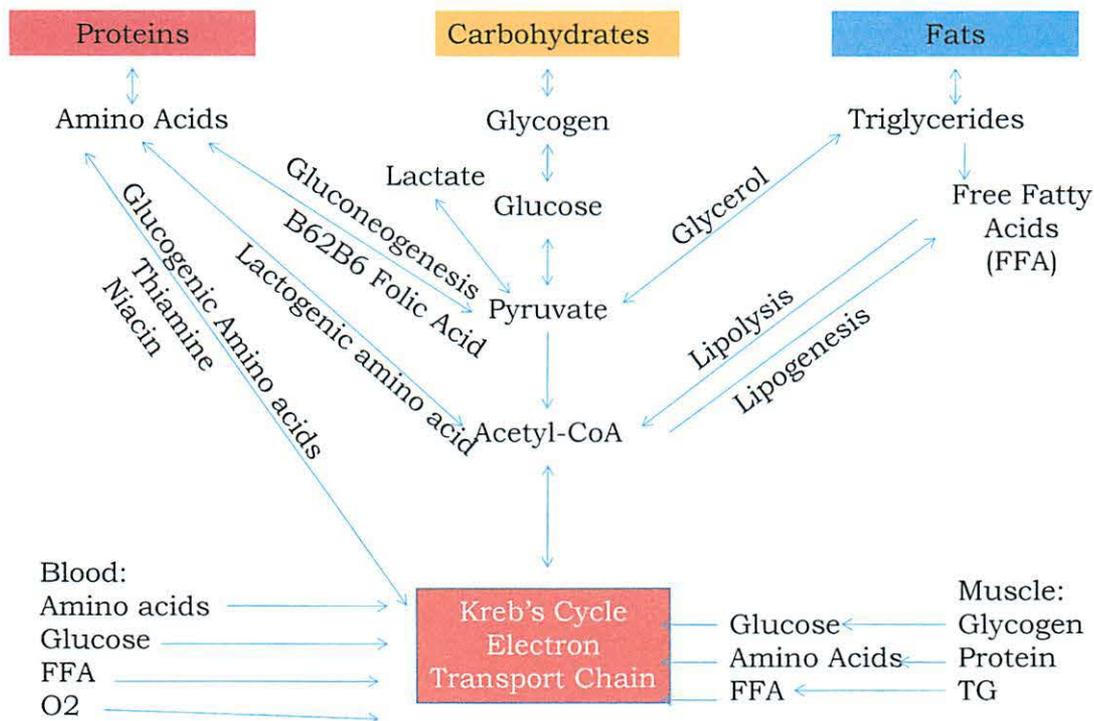


Fig. 8 Inter-conversions of major bio-molecule substrates in the body

Fig. 8 depicts that carbohydrate is not the sole source of ATP for the body. When there is no sugar available, the cell shifts to utilizing other substrates like fats and proteins. Triglycerides from degradation of fats can either be converted into pyruvate or the fatty acids through lipolysis can be converted into acetyl CoA and eventually enters into the Kreb's Cycle which generates NADH and FADH. These two precursors are fed into the Electron Transport Chain and in the presence of oxygen, produce the bulk of energy (ATP) for the cell. Proteins on the hand, has three possible pathways by which they can indirectly give of ATP for the cell, Amino acids from proteins can then become pyruvate through gluconeogenesis, or through ketogenic reactions, amino acids can become acetyl-CoA and down the line through the Kreb's Cycle. Or some amino acids could directly enter into Kreb's Cycle and finally reach the electron transport Chain. Expanding this concept onto the implications of fasting, reserved complex molecules in muscles, liver and other

cells, are then utilized for basal metabolism and build-up of its own complex molecules. This is normal process in the body of an organism as part of the homeostatic functions.

The important implications then are: (1) reserve glycogen in the liver are utilized and as a consequence, new batch of reserved are synthesized once the body resumes normal eating regimes; (2) fat deposits are also utilized therefore makes the body more toned and less stressed; (3) it may also prevent liver cirrhosis.

Reflecting on the previous discussions, the framework here is that, there is rapid metabolic activity when there is rapid insulin secretion, which we know is a result of high blood glucose levels. But this condition is readily attained by directly taking carbohydrates. Take note that, sugars can also be produced from non-carbohydrate sources as described, however, such pathways are only used when the cell does not have ready monomers of sugar in order to survive at basal condition (Fig. 4).

A review on *Caenorhabditis elegans* and the experiment on Dietary Restrictions related to prolonged lifespan

(Selman *et al.*, 2007; van Heemst *et al.*, 2008; Walker *et al.*, 2005)

Caenorhabditis elegans (Fig. 9a and 9b) is a small, free-living, round worm found in nutrient- and microorganism-rich habitats such as in compost, mushroom beds and garden soil where it feeds on bacteria and probably other microorganisms.



Fig. 9a.

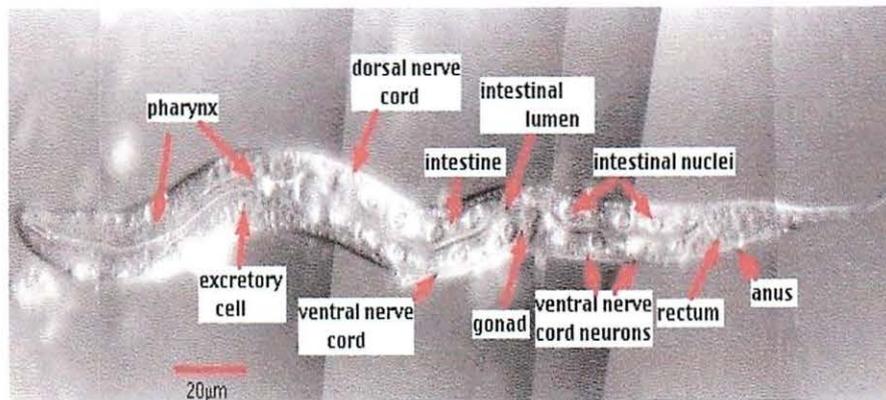


Fig. 9b

Fig. 9. *Caenorhabditis elegans* and its parts

Several attractive features of *C. elegans* biology have contributed its choice as a model organism for the study of genetics, developmental biology, neurobiology, cell biology and behavior. It is easy to maintain in the laboratory, growing quickly on a bacterial lawn grown on an agar plate.

C. elegans has a short generation time (about 14 days or 2 weeks), going from the single-cell egg to an adult in 3.5 days. The embryo hatches and goes through four larval stages before becoming a mature adult. Each larval stage looks similar to the adult, only smaller. *C. elegans* can adapt an alternative life form, call a dauer larva, in response to overcrowding or the absence of adequate food supply. This dauer larva can remain viable for as long as three months while it roams around in search of food. There is so much implications on fasting here. If food is restricted (fasting), the lifespan is extended to as long as three months.

The genome is small compared to humans (about 30 times smaller), yet it encodes over 22,000 proteins, only slightly fewer than humans. About 35% of *C. elegans* genes are closely related to human genes. The mammalian *Ku heterodimer* has important roles in DNA double strand break repair, telomere maintenance, cell cycle checkpoint-arrest, tumor suppression, and cellular stress resistance. To investigate the evolutionarily conserved functions of Ku, we knocked down expression by RNA interference (RNAi) of Ku genes in *C. elegans*. We found that *C. elegans* Ku70 (CKU-70) is required for resistance to genotoxic stress, regulates cytotoxic stress responses, and influences aging. The latter effects are dependent on an IGF-1/insulin-like signaling pathway previously shown to affect life span. Reduction of CKU-70 activity amplifies the aging phenotype of long-lived insulin receptor *daf-2* mutations in a *daf-16*-dependent manner. These observations support the view that organismal stress resistance determines life span and Ku70 modulates these effects.

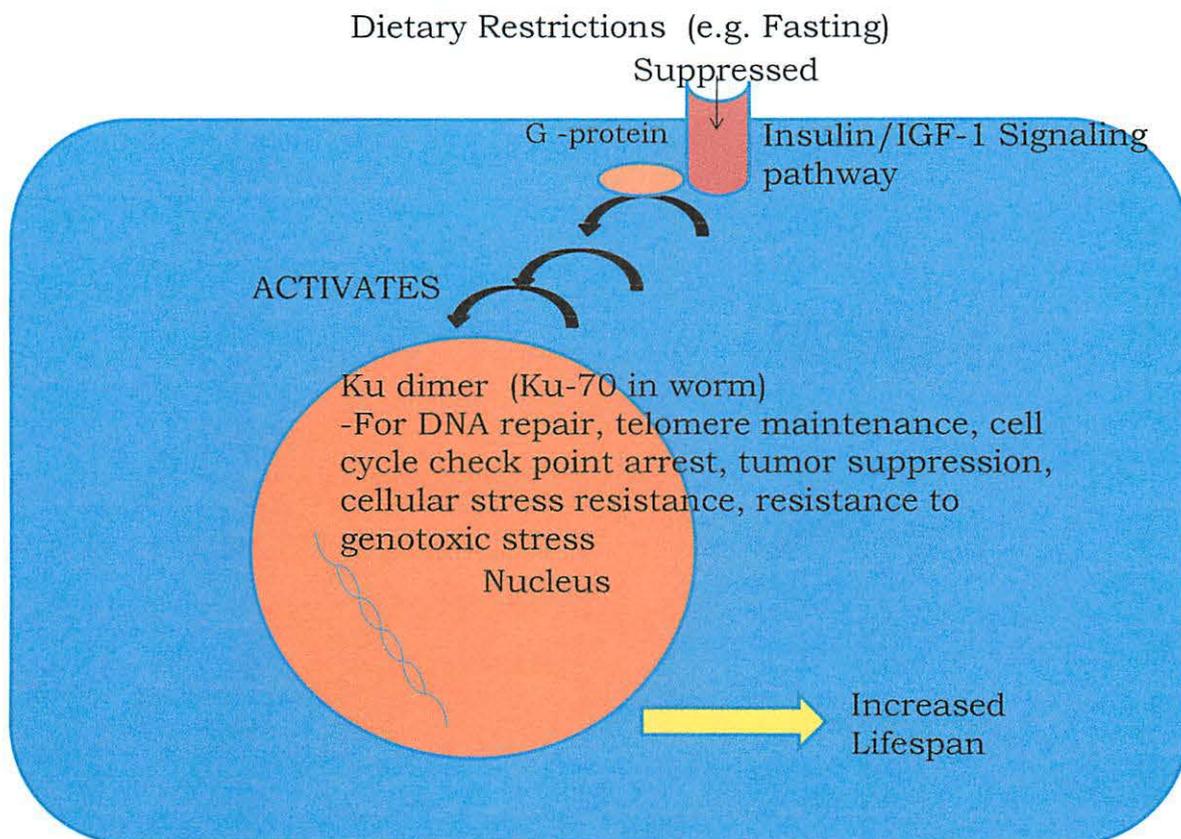


Fig. 10 Cascading Effects when under Dietary Restriction

The nematode *Caenorhabditis elegans* has been subjected to dietary restriction (DR) by a number of means, with varying results in terms of fecundity and lifespan. Two possible mechanisms by which DR increases lifespan are reduction of metabolic rate and reduction of insulin/IGF-1 signaling. However, interaction studies suggest that DR and insulin/IGF-1 signaling may act in parallel on common regulated processes. In this review, we discuss recent developments in *C. elegans* DR research, including new discoveries about the biology of nutrient uptake in the gut, and the importance of invasion by the bacterial food source as a determinant of lifespan.

Mammalian Experiment on Fasting

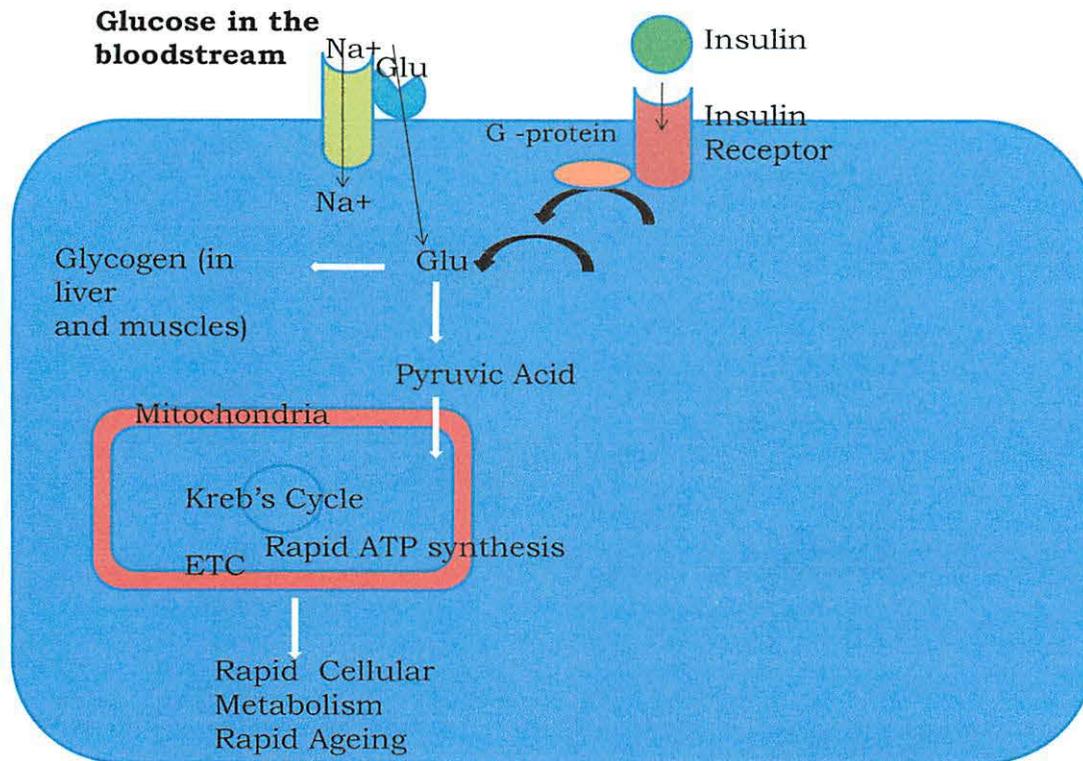
A major focus of current work is the understanding of genes and biochemical processes by which reduced insulin/IGF-1 signaling and dietary restriction increased lifespan. Recent evidence suggests that alterations in insulin/insulin-like growth factor 1 (IGF1) signaling (IIS) can increase mammalian life span. For example, in several mouse mutants, impairment of the growth hormone (GH)/IGF1 axis increases life span and also insulin sensitivity.

Measurement was done on the life span of mice lacking either insulin receptor substrate (IRS) 1 or 2, the major intracellular effectors of the IIS receptors. Provisional results indicate that female *Irs1*^{-/-} mice are long-lived. Furthermore, they displayed resistance to a range of age-sensitive markers of aging including skin, bone, immune, and motor dysfunction. These improvements in health were seen despite mild, lifelong insulin resistance. The results therefore suggested that IRS1 signaling is an evolutionarily conserved pathway regulating mammalian life span and may be a point of intervention for therapies with the potential to delay age-related processes.

Evidence is accumulating that aging is hormonally regulated by an evolutionarily conserved insulin/IGF-1 signalling (IIS) pathway. Most long-lived IIS mutants also show increased resistance to oxidative stress. Results that increased lifespan is significantly associated with lower body height and improved old age survival.

Several converging lines of evidence obtained over the last years in a wide variety of experimental model organisms suggest that the ageing process is regulated by genes that encode proteins from the somatotroph axis: longevity genes like *daf-2*, which were identified using mutant *Caenorhabditis elegans* strains, turned out to be orthologues of the mammalian genes encoding **insulin-like signaling cascade proteins**. Results suggest that in mammals, lifespan can be increased by continuous, long-term downregulation of IGF signalling.

Mutation analyses in the nematode, *Caenorhabditis elegans*, and mice have identified genes that increase their life-span via hormonal signal transduction, i.e. the insulin/insulin-like growth factor-1 (IGF-1) pathway in nematodes. Figure 11 below describes the cascading effects of dietary restrictions and the activation of the Ku-heterodimer which is responsible for the extended lifespan through hormonal pathway in the absence of insulin.



Consequence of Rapid Insulin Release

Fig. 11

Figure 11 clearly illustrates the consequence of rapid insulin release and the reverse is true which results to slow aging and longer lifespan. Therefore, considering the above discussions it is very clear that there are profound healthful effects of fasting. Fasting is equated with **cell resting** from its metabolic activities which results to lengthening of lifespan and delayed ageing processes as evidenced in the experiment on mice and the worm *Caenorhabditis elegans*.

Integration of Faith and Learning:

1. Our fundamental belief on Sabbath keeping emphasizes physical rest from all our earthly labors and we spend our time acknowledging the ownership of God over us. Jesus' custom was to go the synagogue on Sabbath to study the word and be filled with the Holy Spirit. When we practice fasting – **we are also allowing the resting (sabbath) for the cells in our body which results to longer lifespan and delayed ageing.** But far more important than these are the implications on the life of Jesus when he fasted. In Matthew 1:1-4 it says.... "Then Jesus was led by the Spirit into desert to be tempted by the devil. After fasting forty days and nights, he became hungry and then the tempter came to him and said, "If you are the Son of God, tell these stones to become

*bread. Jesus answered, **it is written: Man shall not live by bread alone, but on every word that comes from the mouth of God***

As we can see in this passage, Jesus gained spiritual strength after fasting. It seemed ironical to think that in physical weakness, He gained spiritual strength. Jesus fasted before facing his ultimate sacrifice on the cross. Had he not done this preparation, he could have yielded to the devil.

And if we practice fasting and prayer on the **Sabbath**, just like Jesus – we will be more receptive to God’s word as he manifested in his answer to the devil... *That man shall live by **bread** alone but by every word that proceedeth from the mouth of God. We can also be ready to do greater service in spreading the good news of salvation.*

2. As we take a physical rest from our daily routines, fasting and prayer could be one of our recommended practices because the power of spiritual renewal is more efficacious in the midst of our physical weakness. Jesus demonstrated it when he was about to face his ultimate sacrifice on the cross.
3. **In Isaiah 58**, fasting -- becomes very meaningful if it is accompanied with social justice. Let’s say we fast one meal.... and put money value in it. Instead of using that money to buy that one meal, we do the fasting – and we offer that money to the church. We can use this fund to feed the hungry, clothe the naked, take care of the orphans and widows and we can even help build more churches. **This is the very essence of having relationship with God – the very core of Sabbath Keeping.**
4. Isn’t it true that in **Romans 12:1**, *it says...“Therefore, I urge you, brothers and sisters, in view of God’s mercy, to offer your bodies as living sacrifices, holy and pleasing to God – this is your spiritual act of worship.* Isn’t it true that our Creator designed our bodies to have longer but wholesome life? Practice fasting or not? The choice is ours to ponder.
5. *Finally, I am exhorting everyone to practice fasting and prayer on the Sabbath because it does not only allow a rest for our cells but it develops in us spiritual strength..... such simple sacrifice, ultimately becomes holy and pleasing to God.*

Main References:

Alberts, B., Johnson, A., Lewis, J., Raff, M., and Roberts, K. 2008. The Molecular Biology of the Cell. Garland Science, U.S.A.

Metzger, Bruce M and Michael D. Coogan eds. 1993. The Oxford Companion to the Bible. Oxford University Press. New York. p.225.

Selman C, Lingard S, Choudhury AI, Batterham RL, Claret M, Clements M, Ramadani F, Okkenhaug K, Schuster E, Blanc E, Piper MD, Al-Qassab H, Speakman JR, Carmignac D, Robinson IC, Thornton JM, Gems D, Partridge L, Withers DJ. 2007. Evidence for lifespan extension and delayed age-related biomarkers in insulin receptor substrate 1 null mice. Centre for Diabetes and Endocrinology, Department of Medicine, Rayne Institute, University College London, University St., London, UK.

Van Heemst D, Beekman M, Mooijaart SP, Heijmans BT, Brandt BW, Zwaan BJ, Slagboom PE, Westendorp RG. 2008. Reduced insulin/IGF-1 signalling and human longevity. Section of Gerontology and Geriatrics, Department of General Internal Medicine, Leiden University Medical Centre, PO Box 9600, 2300 RC Leiden, The Netherlands. **FASEB J. 22(3):807-818.**

Walker, G. Houthoofd, K., Vanfleteren, J.R., Gems, D. 2005. Dietary restrictions in *C. elegans*: From rate of living effects to nutrient sensing pathways. **Mechanism of Ageing and Development 126: 929- 937.**