Inaugural Lecture

by

PROF EMMANUEL MUKWEVHO

Topic:
Chromatin Remodeling: The Epigenetics of NRF-1 in Diabetes & Obesity Therapeutics

Date:
Wednesday, 15 August 2018

Time: 18h00 for 18h15

Venue:
North-West University, Mafikeng,
Lecture Room A1-G45

FACULTY OF NATURAL AND AGRICULTURAL SCIENCES
PROFILE:

Academic Profile:

Prof Mukwevho holds BSc (Univen), BSc Hons (UL) MSc (UCT), PhD (UCT) and Certificate in Financial Management (UCT) and Certificate in Project Management (UCT). He is currently in a process of submitting his MBA thesis having already completed all its course work at NWU. He graduated for his PhD in June 2010 and employed as Lecturer that very year at the University of Johannesburg (Kingsway Campus) in the department of Biochemistry. He was then promoted to Senior Lecturer at University of Johannesburg in November 2012, where he also served in the Science Faculty board for 5 years and also a Biochemistry Academic Advisor committee member to the Food Technology at UJ. He was then head hunted to NWU in 2014 as an Associate Professor and joined NWU, 1st October 2014. He was promoted to a Full Professor of Biochemistry in 1st January 2017. He has also received an NRF rating of Y2 in 1st January 2017 (Rating recognition given to young scientist under the age of 40). He also serves as a Subject Chair of Biochemistry in Mafikeng since 2016. He has been involved in various activities with the department of Science & Technology of SA.

Research Focus:

His research focus is on Obesity, Diabetes and Metabolic Syndrome where he focuses on Molecular pathways and signaling molecules involved. In this regard he studies these events within the cell by the use of Mice, Rats, Human Beings and Cell cultures models from the chromatin to the protein level. Both Lipids and Carbohydrate metabolism are the focus, as they are the major events in the etiology and pathogenesis of the metabolic disorder. Furthermore, he studies these pathways with the view of finding novel therapeutics to manage and cure Diabetes, Obesity and Metabolic Syndrome. Exercise, plants extracts and synthesized compounds such TZDs are explored in quest of finding a cure and better management of these disorders. In the three years Prof Mukwevho has been in NWU, has produced postgraduate students at all levels.

In Research funding, Prof Mukwevho has been funded under Thuthuka for 6 years by National Research Foundation, and now as a rated researcher funded through the ‘Incentive for Rated Researchers’ and the ‘Competitive Incentive for Rated Researchers’.

Postgraduate students Production:

Since joining NWU, 3 years ago, he has produced already 7 MSc students (Molepo, Masilo, Matumbo, Isaiah, Munansangi, Masinye & Fashii) and 3 PhDs (2 in NWU - [Dr Ayeleso Betty & Dr John Owonobi] & one PhD produced at WITS [Nyangunda] through the existing collaboration with Prof Kennedy Erwiggen). He has also trained 4 Post-Docs [Dr Ayeleso, Dr Fukai, Dr Amal & Dr Tella] since joining North West University 3 years ago, two of which are in tenure positions and are Senior Lecturers, one in SA and another in Nigeria.

He has also produced about 15 BSc Hons graduates all from North West University only.

Involvement in Committees worldwide & Collaborations:

He serves in many International journals and reviewed for groups such Elsevier, Wiley, and Springer. In research funding, he has reviewed national for the National Research Foundation (NRF) and internationally, the American Heart Association (AHA). Collaborations that exist with Prof Mukwevho are with Melbourne University (Prof Mark Hargreaves -DVC Research & Postgraduate) in Australia, Cambridge University-(Prof Nick Gay) in the UK, Texas Southern University with Prof Mazayaani, Ashraf in USA and many more. Nationally, he collaborate with UCT, Wits, UJ, UKZN and CSIR. He has received several funding instruments from the National research Foundation and MRC.

He has just been appointed to serve in the Council of South African Society for Biochemistry & Molecular Biology (SASBMB) which serves to promote the field of Biochemistry & Molecular Biology in the country from 2018-2021. He has also been appointed at CSIR to serve at the institution External Ethics committee from 2018-2021. He is a
member of ASBMB (American Society of Biochemistry & Molecular Biology) and also APS (American Physiological Society). He has been serving also at CHE (Council for Higher Education) since 2012 as a reviewer for the entity for Quality control.

TEACHING & LEARNING

Prof Mukwevho has taught Biochemistry both at undergraduate and Postgraduate Levels both University of Johannesburg and NWU. At UCT he has served as guest Lecturer for Physiology and also in teaching MBCHB 2nd & 3rd students between 2003-2006 under the department of Human Biology. He teaches mostly Introduction to Biochemistry, Metabolism, Research and also Analytical Biochemistry. Passionate in delivering quality in Teaching & Learning.

COMMUNITY SERVICE

Prof Mukwevho has been involved in various projects that imparts communities within South Africa, having served as guest on SABC2, DSTV as Science & Technology experts. In Radio he has also provided many talks as guest experts on issues involving Science and Technology and Lifestyle diseases awareness on SABC Radios. He has been also involved around Mafikeng in Lifestyles Disease awareness campaign.
Chromatin Remodeling:
The Epigenetics of NRF-I in Diabetes & Obesity Therapeutics

You're Fat. Don't try and sugar coat it because you'll eat that too.
CONTENT OUTLINE

• THE PROBLEM
  ✓ Diabetes- Carbohydrates
  ✓ Obesity- Lipids
  ✓ Metabolic Syndrome

• INTERVENTIONS
  ✓ Exercise
  ✓ Phytotherapies/Drugs
  ✓ Solving the Problem

• Models: Human, Rats & Cell Culture
  ✓ NRF-1 ...Link for parallel pathways for Lipids & Glucose
  ✓ CaMKII in Lipid and Glucose metabolism

TERMINOLOGIES

1) DIABETES - is a disease that affects your body's ability to produce or use insulin. Insulin is a hormone. When your body turns the food you eat into energy (also called sugar or glucose).

2) OBESITY - is a medical condition in which excess body fat has accumulated to the extent that it may have a negative effect on health. People are generally considered obese when their body mass index (BMI), a measurement obtained by dividing a person's weight by the square of the person's height, is over 30 kg/m², with the range 25–30 kg/m² defined as overweight.

3) EPIGENETICS - the study of changes in organisms caused by modification of gene expression rather than alteration of the genetic code itself.

4) THERAPEUTICS-the branch of medicine concerned with the treatment of disease and the action of remedial agents

5) CHROMATIN REMODELING-Chromatin remodeling is the dynamic modification of chromatin architecture to allow access of condensed genomic DNA to the regulatory transcription machinery proteins, and thereby control gene expression.

6) INSULIN RESISTANCE- Insulin resistance (IR) is a condition in which the body's cells become resistant to the effects of insulin

7) NRF-1- Its a protein that homodimerizes and functions as a transcription factor which activates the expression of some key metabolic genes regulating cellular growth and nuclear genes required for respiration, heme biosynthesis, and mitochondrial DNA
THE COMPLEX, ever busy WORLD

- In the ever changing, fast & busy world, Man has forgotten to Live
- Preoccupied by Unending daily Activities & Deadlines, All these fatigue us
- They put tremendous pressure on the body and alters Normal Cellular events, Hormones and Genes that confers protection against various ailments
- Even in the Bible God instructed man to rest and do no work at the of the Week
- The Cell normal Metabolism (Homeostasis) is destabilized consistently and daily because we never rest and always busy.
- Daily Habitual Activities/Lifestyle has significant effect on the CELL (the Basic unit of life)
- Once the Cellular activities are altered, Diseases find a way to us, some Fatal
- My Research is understanding Molecular & Cellular events of Cell
- Most Importantly Lifestyle activities: Impact of Exercise, Glucose and Lipid Metabolism in the Cell, especially the Chromatin

THE ENEMY: CHRONIC DISEASE

- "Chronic disease" is defined as a disease that is slow in its progress and long in its continuance.
- Major examples of chronic disease are coronary heart disease (including atherosclerosis, heart failure, hypertension, and stroke), diabetes, Type 2 diabetes, some cancer, osteoporosis, and senility (frailty in old age as a result of weak muscles). It would be difficult to find anyone in our society who is exempt from the devastating effects of one or more chronic diseases.
- There has been a dramatic increase in the incidence of chronic diseases in the latter part of the 20th century.
- Obesity is considered a comorbidity of some of the most prevalent diseases of modern society (15, 40). In fact, the number of comorbidities displayed by an individual rises with increasing body weight.
- Type 2 diabetes has become so common in our society that it has been said to have reached epidemic proportions. A sixfold increase in prevalence of Type 2 diabetes occurred between 1980 and 1993. Historically, Type 2 diabetes has been considered a disease of adults and older individuals and not a pediatric condition.

WAGING THE WAR AGAINST OBESITY & DIABETES

FRANK W. BOOTH

J. Appl. Physiol. 85: 774-787, 2000, 2005
Well... You see the problem is that obesity runs in my family.

Patient: The problem is that obesity runs in our family.

Doctor: No, the problem is that nobody runs in your family.

SOCRATES & SHAKESPEARE

- Greek Physician Hippocrates wrote "That which is not used cavelops and that which is not used waste away.
- The relevance of this is in Muscle tissue.
- Shakespeare writing and plays none of them where death was through Diabetes/Obesity or chronic diseases.
From the Atom to an Organism

Cell Organelles

Biochemical Pathways
Molecular Pathways
Cellular Pathways
Skeletal muscle is an active tissue that adapts to changes in its metabolic and contractile properties, including oxidative slow-twitch (type I), mixed oxidative-glycolytic fast-twitch (type IIA), and glycolytic fast-twitch (type IIB) myofibers.

- **Type I fibers** preferentially express enzymes that oxidize fatty acids, contain slow isoforms of contractile proteins, and are more resistant to fatigue than are glycolytic fibers.
- **Type II fibers** preferentially metabolize glucose and express the fast isoforms of contractile proteins.
- **Endurance exercise training** triggers a remodeling program in skeletal muscle that progressively enhances performance in athletes such as marathon runners, mountain climbers, and cyclists.
- This involves changes in metabolic programs and structural proteins within the myofiber that alter the energy substrate utilization and contractile properties that act to reduce muscle fatigue (Fluck and Hoppeler, 2003).
- **Training-based adaptations in the muscle** are linked to increases in the expression of genes involved in the slow-twitch contractile apparatus, mitochondrial respiration, and fatty acid oxidation (Hollloszy and Coyle, 1984).
- These adaptations that improve performance can also protect against obesity and related metabolic disorders (Koves et al., 2005).
- Moreover, skeletal muscles rich in oxidative slow-twitch fibers are resistant to muscle wasting (Minnaard et al., 2005).
**THRIFTY GENES HYPOTHESIS**

Thrifty genes are genes which enable individuals to efficiently collect and process food to deposit fat during periods of food abundance in order to provide for periods of food shortage (feast and famine).

Hypothesis to explain rising epidemics of metabolic syndrome - obesity and its closely associated co-morbidities.

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**MUSCLE INACTIVITY (ATROPHY)**

- Reduced or altered contractile activity
- Raised energy expenditure
- Reduced dietary energy intake
- Negative energy balance
- Hypoaemia
- Acidosis
- Steroid treatment
- Increased pro-inflammatory cytokines

Muscle wasting

Insulin resistance
Reduced IGF-1
Reduced testosterone

β₂-adrenergic agonists
Biochemical Pathways

Physical Inactivity

- Coronary artery disease 45%
- Breast Cancer 41%
- Stroke 40%
- Diabetes 50%
- Colon Cancer 41%
- Hypertension 40%
- Osteoporosis 50%
PROBLEMS ASSOCIATED WITH INACTIVITY

- Decrease in peak consumption of O2 during maximal aerobic exercise
- Decrease in maximal cardiac output
- 25% decreases in maximal stroke volume
- Bones lose mass at 10 times their normal rate
- Skeletal muscles become weaker with less endurance for light physical effort
- Muscle lowers the capacity to oxidize fatty acids and thus obesity tricks in.
- Whole body insulin sensitivity declines within the first 3 days of inactivity, whether it is bed rest or active individuals stopping daily exercise.
- Deep vein thrombosis (DVT) can occur within a time frame as short as an international flight with possible pulmonary thromboembolism in susceptible individuals

CONSEQUENCES OF INACTIVITY

- Lack of physical activity
  - Decreased turnover of endogenous energy stores
  - Decreased muscle glycogen turnover and decreased intramyocellular lipid turnover
  - Skeletal muscle insulin resistance

- Hyperinsulinemia
  - Increased de novo lipogenesis
  - Steatosis
  - Hepatic insulin resistance
  - Hyperglycemia

- Energy partitioning to adipose tissue (Central obesity)
  - Increased hepatic VLDL production and decreased LDL levels

- Metabolic syndrome
  - Cardiovascular disease

- Type 2 diabetes
Physical Activity

- Physical activity involves the coordinated function of several Physiological and Biochemical systems:
- These include the musculo-skeletal system and cardiovascular system
- Within these physiological/Biochemical systems there are signaling and metabolic pathways.
- These metabolic pathways are involved in oxidative phosphorylation,
- where ATP is synthesized
- involving a process that transfers hydrogen to molecular O2,
- are responsible for 90% of ATP synthesized in the body
Exercise

- The BEST Medicine/Pill
- Which can not be bottled by any Drug company.
- It is FREE of charge
- Drugs are very Expensive.
- Exercise provides All these benefits at NO Cost in Rands but in Sweat to you.

20 Exercise Benefits

1. Reduces body fat
2. Increases lifespan
3. Oxygenates body
4. Strengthens muscles
5. Manages chronic pain
6. Ward off viruses
7. Reduces diabetes risk
8. Strengthens heart
9. Clears arteries
10. Boosts mood
11. Maintains mobility
12. Improves memory
13. Improves coordination
14. Strengthens bones
15. Improves complexion
16. Detoxifies body
17. Decreases stress
18. Boosts immune system
19. Lowers blood pressure
20. Reduces cancer risk

www.facebook.com/montereybayholistic

Walking

- The 30 Minutes a day
- 10,000 if you have pedometer
- 10 min of aerobic exercise
- Or in simple terms Exercise to break a SWEAT.
- This means Activities enough to enable just to break a SWEAT will accrue these benefits
- This puts a bay Metabolic disorders by 5%
- The benefits of exercise increase with increase in the workout.
- Per week its recommended you have burned 500KCal to get these benefits
- As calories expenditure increase the benefits increases too.
### Body Fat Chart for Men (%)

<table>
<thead>
<tr>
<th>AGE</th>
<th>18-29</th>
<th>30-39</th>
<th>40-49</th>
<th>50-59</th>
<th>60-69</th>
<th>70+</th>
<th>AVERAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Body Fat Chart for Women (%)

<table>
<thead>
<tr>
<th>AGE</th>
<th>18-29</th>
<th>30-39</th>
<th>40-49</th>
<th>50-59</th>
<th>60-69</th>
<th>70+</th>
<th>AVERAGE</th>
</tr>
</thead>
<tbody>
<tr>
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<td></td>
<td></td>
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</tr>
</tbody>
</table>

1. Body Fat Charts provided by bodyfatcharts.com
2. Data untested by number of Anschutz Medical, LLC

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### Legion

**Female Body Fat Chart**

- **Essential**
  - 10-12%
- **Athlete**
  - 13-20%
- **Fit**
  - 21-24%
- **Normal**
  - 25-31%
- **Overweight/Obese**
  - 32%+

www.legionathletics.com
OBESITY

- It's due to excess calorie intake than its expenditure.
- Excess carbohydrates are stored as glycogen but when its Max is reached they are converted into fat.
- Obesity causes diabetes, insulin resistance, metabolic syndrome, cancer, hypertension, endometrial, breast, prostate, and colon cancers etc.
- Metabolic syndrome: characterized by insulin resistance, excess triglycerides, inflammation e.g. diabetes, hypertension, atherosclerosis, coronary heart diseases.
- Individuals of these diseases have excess visceral fat.
- Exercise, Metformin etc are used to curb the syndrome.
ABUNDANCE OF FOOD IN MOST PARTS OF WORLD

We are faced with difficult decisions about our health daily, especially choices on food to eat. In a world with plenty options of Fast food, refined carbohydrates, high sodium content foods we buy.

The healthy food choices is usually not tasty or looking any fancy but dull plate of veggies and fruits.
Table 22-2: Fuel Reserves for a Normal 70-kg Man

<table>
<thead>
<tr>
<th>Fuel</th>
<th>Mass (kg)</th>
<th>Caloriesa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tissues</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fat (adipose triacylglycerols)</td>
<td>15</td>
<td>141,000</td>
</tr>
<tr>
<td>Protein (mainly muscle)</td>
<td>6</td>
<td>24,000</td>
</tr>
<tr>
<td>Glycogen (muscle)</td>
<td>0.150</td>
<td>600</td>
</tr>
<tr>
<td>Glycogen (liver)</td>
<td>0.075</td>
<td>300</td>
</tr>
<tr>
<td>Circulating fuels</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucose (extracellular fluid)</td>
<td>0.020</td>
<td>80</td>
</tr>
<tr>
<td>Free fatty acids (plasma)</td>
<td>0.0003</td>
<td>3</td>
</tr>
<tr>
<td>Triacylglycerols (plasma)</td>
<td>0.003</td>
<td>30</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>166,000</td>
</tr>
</tbody>
</table>

a1 (dieter's) Calorie = 1 kcal = 4.184 kJ.
ITS ALL IN CALORIES & the GENETICS

Calories In
Must Equal
Calories Out

Average needed Calories 1800 - 2500KCal required per Day

BMR = 75%

# ENERGY EXPENDITURE

A schematic representation of the balance between gene interactions with physical activity and inactivity on metabolic functioning is shown.

Top: an environment of daily physical activity recapitulates the milieu during natural selection, producing metabolic homeostasis.

Bottom: when physical activity is removed from the environment, there is a loss of an integral environment-gene interaction that causes a maladaptive response leading to multiple metabolic dysfunctions and contributing to many chronic diseases.
Causes of Obesity

There's usually not just one cause of obesity. Multiple factors may interact and contribute to the condition.

- Eating more calories than the body requires
- Pregnancy
- Certain medical conditions such as polycystic ovary syndrome, a hormonal disorder
- Emotional or psychological factors such as stress, depression or low self-esteem
- Lack of sleep
- Genetics and family history
- Medications such as some antidepressants
- Lack of exercise

STATS FOR OBESITY & OVERWEIGHT

Normal Fat
14-28%
† 1-2 billion

Underfat
9-10%
† 675-750 million

Overfat
62-76%
+++++ 4.5-5.8 billion
2 in 3 adults are overweight or obese.

Body Mass Index (Kg/m²)

BMI to describe various levels of body fat:
- Normal weight: 18.5 - 24.9
- Overweight: 25.0 - 29.9
- Obese: 30.0 - 39.9
- Extreme obesity: 40.0 and above
Impact of a meal rich in saturated fatty acids

- Intramuscular lipids
- Liver Transport FA to Tissue inducing Insulin Resistance
- Adipose Formation/Increase

Obesity results in metabolic syndrome

- Oxidative stress to remote tissues
- NADPH oxidase
- Antioxidative enzymes

Oxidative stress in WAT

- Dysregulation of adipocytokines
- PAI-1, TNF-α, MCP-1
- Adiponectin

- Insulin resistance
- Diabetes
- Atherosclerosis

Metabolic syndrome
UNDER THE SKIN FAT THE MOST DANGEROUS FAT. Produces Inflammation

INFLAMMATION

- Excessive Lipids or being Obese results in INFLAMMATION
- Inflammation itself results in DISRUPTION of Metabolic Processes/pathways
- Altered Processes leads to various Ailments such Insulin Resistance, Diabetes, Cardiovascular Diseases, Brain disorders.
TYPE 2 DIABETES
[Constitutes >90%] of All Diabetes cases.
DIABETES MELLITUS

Healthy

Type 1

Type 2

Insulin receptor

Pancreas failure to produce insulin

Insulin

Cells fail to respond to insulin properly

Glucose

Glucose

TYPE 2 DIABETES

- Most common
- >40 years of age
- Normal or overweight
- Genetic factor
  - Strongly positive

Non Diabetic (No Family History)

Non Diabetic (With Family History)

Diabetic
Type 2 Diabetes

- Increased glucose in the bloodstream
- Sufficient insulin secreted in the bloodstream
- Muscle unable to use glucose due to insulin resistance

Factors leading to insulin resistance:
- Obesity
- Inheritance
- Other factors

Diabetes Complications:
- Stroke
- Heart attack
- Eye complications
- Nerve damage
- Foot complications
- Kidney complications
- Cataracts

Glaucoma
Drugs Decrease Insulin Resistance

Metformin

A thiazolidinedione (TZD)

Diabetes a 3rd leading cause of death in the US after Heart disease & cancer.

2 types of diabetes:
1. Insulin dependent and Non insulin dependent (>90%).
Drugs such as Metformin & TZD are used.
Some Increase Risk of Heart Attack & Stroke

Rosiglitazone (Avandia)
Three major biological functions

1. Essential component of biological membranes forming lipid-bilayer
2. Lipid containing hydrocarbons serve as energy source
3. Many intra- and intercellular signaling events involve lipid molecules. E.g inositol, phospholipids

Plasma triacylglycerol & cholesterol transport in humans
COMPARISON OF METABOLISM IN
a) Fed (abundance of energy)
b) Starvation (Scarcity of energy)

Adipocytes Containing Fat Globule
LIPIDS PLAY VITAL ROLE IN MEMBRANE FORMATION
CRITICAL IN MANY PATHWAYS e.g. ATP SYNTHESIS

\[
\begin{align*}
\text{Intermembrane space} & : & 4H^+ \\
\text{Inner mitochondrial membrane} & : & \text{FeS} \\
\text{Matrix} & : & \text{NADH} \\
\text{Complex I} & : & \text{FMN} \\
\text{Complex III} & : & \text{Cyt b} \\
\text{Complex IV} & : & \text{Cyt c} \\
\end{align*}
\]

\[
\begin{align*}
\frac{1}{2}O_2 + 2H^+ & \rightarrow H_2O \\

4H^+ & \rightarrow 2H_2O \\
\end{align*}
\]
LIPIDS play vital role in Signal Transduction Pathways

Metabolic Homeostasis

AMPK-The Cell Fuel Gauge

- It activates Glycolysis in cardiac muscle
- Inhibits Lipogenesis & gluconeogenesis in liver
- Promotes fatty acid oxidation & glucose uptake in muscle
- Inhibits Lipolysis in adipocytes
- AMPK is regulated by adiponectin hormone found in adipocytes.
- Leptin also controls metabolism as a satiety hormone by promoting energy expenditure.
- Leptin resistance lead to increase of Neuropeptide Y which stimulate appetite & fat accumulation (Inhibited by Leptin+Insulin)
- Grehlin is an appetite stimulating hormone secreted by empty stomach
Gene "switched on"
- Active (open) chromatin
- Unmethylated cytosines (white circles)
- Acetylated histones

Transcription Factors / Co-activators

Gene "switched off"
- Silent (condensed) chromatin
- Methylated cytosines (red circles)
- Deacetylated histones

Transcription possible

Transcription impeded

HOW DOES EXERCISE PROTECT INDIVIDUALS FROM OBESITY & DIABETES?
Exercise protects individuals from type 2 diabetes

Delays/Prevents the onset of T2D/Obesity

Our Hypothesis Model

Exercise → CaMKII → NRF-1

Glucose transport (GLUT4)
DIPHO (NRF-1)
CaMKII is activated by exercise

Mechanisms by which exercise protect individuals.

- Increases in glucose transport - GLUT4 protein.
- Increases in mitochondrial oxidative capacity - NRF-1.
- By remodeling the chromatin increasing NRF-1 epigenetics
- The result is increased expression of genes & protects individuals from these ailments.
The GLUT4 Protein

- Insulin responsive protein
- The major glucose transporter in skeletal muscle.
- Essential in glucose transport

Blood glucose

<table>
<thead>
<tr>
<th>Glucose transporters</th>
<th>Location</th>
<th>Insulin</th>
</tr>
</thead>
<tbody>
<tr>
<td>GLUT1</td>
<td>skeletal muscle, brain, erythrocytes</td>
<td>No</td>
</tr>
<tr>
<td>GLUT2</td>
<td>liver, pancreas, kidney, intestine</td>
<td>No</td>
</tr>
<tr>
<td>GLUT3</td>
<td>Brain</td>
<td>No</td>
</tr>
<tr>
<td>GLUT4</td>
<td>skeletal muscle, heart, brain, adipose</td>
<td>Yes</td>
</tr>
</tbody>
</table>

Hypoglycemia

<table>
<thead>
<tr>
<th>6</th>
<th>8</th>
<th>10</th>
<th>12......mmol/L</th>
<th>Hyperglycemia</th>
</tr>
</thead>
</table>

Hypoglycemia

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<tr>
<th>6</th>
<th>8</th>
<th>10</th>
<th>12......mmol/L</th>
<th>Hyperglycemia</th>
</tr>
</thead>
</table>

Hypoglycemia
Glucose transport by GLUT4 protein

Signals that induce GLUT4 expression

Exercise activates CaMKII

Mechanisms involved in pathways that impair or induce GLUT4 expression are not yet fully understood.

GLUT4 Protein

Increases glucose transport
Regulation of the *Glut4* gene expression

- **Glut4** gene
  - Glut4 promoter
  - G4 mRNA
  - GLUT4 protein

NRF-1, the mitochondrial transcription factor

- NRF-1
- Exercise

- Oxidation of LCACOA, Ceramides, Diacylglycerol
- Increased Insulin sensitivity
CaMKII regulate NRF-1

CaMKII regulate lipid metabolizing genes
Although Exercise is not always the easiest method to follow in controlling Diabetes, we want to design alternative methods that simulate Exercise induced benefits using organic compounds in this paper.

**Figure 22.2**

ETC (electron transport chain)

THERE EXCESSIVE CARBOHYDRATE IN OUR MODERN DIET. CHO ARE CONVERTED TO FATS.

EXCESS GLUCOSE/CARBHYDRATES LEADS TO METABOLIC DISORDERS
EVERY BITE YOU TAKE IS EITHER FIGHTING DISEASE OR FEEDING IT.

SUGAR ALTERS GENE TRANSCRIPTION

The results of altered gene transcription:
- Silencing critical genes that protects individuals from many diseases.
G-6-P crossroad of carbohydrate metabolism

1. Glucose $\rightleftharpoons$ G6P
2. Glycogen $\rightarrow$ NADPH
3. G6P $\rightarrow$ Acetyl-CoA
4. G6P $\rightarrow$ RSP

- fatty acids
- phospholipids
- cholesterol

ATP

ENERGY STORAGE

Fat accumulation - a way of storing energy for later use.

1. Fatty acid synthesis - for energy storage
2. Fatty acid degradation - for energy use
3. Two pathways are reverse of each other.
Energy is a tightly control process leading to on and off ATP synthesis.

Regulation of the Citric Acid Cycle

1. PDH
2. Citrate synthase.
3. NAD isocitrrate dehydrogenase.
4. Keto glutarate dehydrogenase

1. Substrate availability
2. Product inhibition (Citrate & OAA)
3. Competitive feedback inhibition
The effect of obese gene knockout

- Obese gene codes for leptin.
- Its lack leads to overeating.
- Excess energy is stored as fat & glycogen in the body for future use.
- Persistent excess calories leads to obesity.

Hormones Play key Role in Controlling Metabolism

1. Leptin = Regulate Appetite
2. Adiponectin regulate Glucose & Lipids
CONTROL OF FOOD INTAKE

Adipose tissue
Pancreas
Stomach

Leptin
Insulin
Ghrelin

Expenditure
Intake
Energy balance

CORTISOL IS INCREASED BY STRESS KILLING LOT OF PEOPLE
Effects of Excess Cortisol to the Body

Cortisol - The Stress Hormone

IMPACT OF CORTISOLON DIABETES & OBESITY
Neuropeptide Y
- stimulates appetite and is released by the Hypothalamus
- Diminished response to Leptin results to high Neuropeptide

1
Tyr — Pro — Ser — Lys — Pro — Asp — Asn — Pro — Gly — Asp — Ala —

10
Pro — Ala — Glu — Asp — Met — Ala — Arg — Tyr — Tyr — Ser — Ala — Leu —

20
Arg — His — Tyr — Ile — Asn — Leu — Ile — Thr — Arg — Gln — Arg — Tyr — NH₂

Neuropeptide Y
(The C-terminal carboxyl is amidated)

Ghrelin
Appetite stimulating hormone secreted by empty stomach

Gly — Ser — X — Phe — Leu — Ser — Pro — Glu — His — Gln —

10
Arg — Val — Gln — Gln — Arg — Lys — Glu — Ser — Lys — Lys —

20
Pro — Pro — Ala — Lys — Leu — Gln — Pro — Arg

Ghrelin
(X = Ser modified with n-octanoic acid)
PYY
An appetite suppressing hormone found in the GIT

\[
\begin{align*}
3 & \quad \text{Ile} \quad \text{Lys} \quad \text{Pro} \quad \text{Glu} \quad \text{Ala} \quad \text{Pro} \quad \text{Gly} \quad \text{Glu} \\
10 & \\
19 & \quad \text{Asp} \quad \text{Ala} \quad \text{Ser} \quad \text{Pro} \quad \text{Glu} \quad \text{Glu} \quad \text{Leu} \quad \text{Asn} \quad \text{Arg} \quad \text{Tyr} \\
20 & \\
29 & \quad \text{Tyr} \quad \text{Ala} \quad \text{Ser} \quad \text{Leu} \quad \text{Arg} \quad \text{His} \quad \text{Tyr} \quad \text{Leu} \quad \text{Asn} \quad \text{Leu} \\
30 & \\
36 & \quad \text{Val} \quad \text{Thr} \quad \text{Arg} \quad \text{Gln} \quad \text{Arg} \quad \text{Tyr} \\
\text{PYY}_{3-36}
\end{align*}
\]

NRF-1

MITOCHONDRION

1. DNA
2. Granules
3. Ribosomes
4. ATP synthase

Matrix

Outer membrane

Pores

Inner membrane
Changes in Insulin Sensitivity and Muscle Oxidative Capacity Kevin R. Short., 2003

1. Aerobic capacity decrease 10% per Decade
   As measured by VO2peak declined linearly with age

2. Insulin sensitivity decrease 8% per Decade

Insulin resistance increases and muscle oxidative capacity decreases during aging, but lifestyle changes—especially physical activity—may reverse these trends
NRF-1 = Nuclear Respiratory Factor 1

- Nuclear respiratory factor 1 (NRF-1) encodes a protein that homodimerizes and functions as a transcription factor which activates the expression of some key metabolic genes regulating cellular growth and nuclear genes required for respiration, heme biosynthesis, and mitochondrial DNA transcription and replication.

MITOCHONDRIAL GENE EXPRESSION IN RESPONSE TO EXERCISE BOTH IN MEN AND WOMEN
NRF-1 = Nuclear Respiratory Factor -1

- Transcription Factor For Mitochondrial Biogenesis
- Control Genes involved in OXPHOS Oxidative Phosphorylation
- Most genes involved metabolism have NRF-1 binding sites
- Now it is known to involved in Glucose Metabolism
- Thus in control both Lipids and Glucose in parallel
- Directly links it to Metabolic Disorders such Diabetes & Obesity

Methods We Employ

- Western Blotting = Protein expression
- Qpcr = Gene expression
- LC/GC-MS – We assess lipid species metabolites
- Chromatin Immunoprecipitation = assess Transcription binding to Cis/Trans Elements
- Accessibility Asssay to assess DNA/Protein accessibility
- Cloning.
- Mammalian Cell culture- C2C12, Hepatocytes, Adipocytes
- Animal models – Rats and Mice
RATS WONDERFUL EXPERIMENTAL TOOL IN MEDICINE, BIOCHEMISTRY & PHYSIOLOGY

They provide us vital tools in elucidating pathways/mechanisms involved in leading Diabetes and Obesity. These provide with information to develop therapeutics for these metabolic disorders, especially in the Chromatin where our focus is at.
Various Rats and Mice models and how we can manipulate their genetics, altering or overexpressing various genes in them.
Questions

- Does NRF-1 bind the Mef2α gene & increase its expression?

- Does the increase in MEF2A result in increased binding to the Glut4 gene?
Experimental Design

- C2C12 myotubes
  - Control
  - Caffeine
  - Caffeine+ KN93

- ChIP assay to detect acetylation & binding.
  - NRF-1/Mef2A
  - MEF2A/Glut4
  - NRF-1/Alas

Methodology

- Control (DMSO)
- Exercise
- Exercise + KN93
MEF2A
8-Tubulin

Wild-type Control 0 30 60 90 pmol MEF2A siRNA

Primer design for Mef2a gene
Tet-On gene expression system
MEF2A RNA Interference

C2C12-Tet-On-NRF-1

Overexpression of NRF-1

A

C2C12-Tet-On-NRF-1 myotubes

NRF-1 protein (relative units)

Vehicle Dox

B

Wildtype C2C12 myotubes

NRF-1 protein (relative units)

Vehicle Dox

α-tubulin
Overexpression of NRF-1

Effect of MEF2A silencing on NRF-1 overexpression
Dox increases NRF-1 binding to the Mef2a gene

Chromatin remodeling: CaMKII
Time line analysis of NRF-1 overexpression
CaMKII activation exports HDAC5

CaMKII activation increases Lauric acid
IN PERORATION

- Humans likely have the potential for the most advantageous environment-gene interaction in their history on Earth; we are quite fortunate to be alive in this era.
- In a period of less than a half-century, knowledge has advanced from Watson and Crick (97) to a complete sequence of the human genome.
- However, humans physical inactivity into their lifestyle has had the adverse effect of increasing the incidence of inactivity-induced chronic disease.
- The solution to this critical societal problem lies in a more advanced understanding of the maladaptations due to physical inactivity-gene interactions.
- Understanding physical inactivity-gene interactions and impact of western diet will further our knowledge of gene-environment interactions, provide the molecular evidence required for the prevention of chronic diseases through physical activity and healthy diet.
- We must identify those molecules that will allow early disease detection, and provide society with the information needed to counter the current strategy of increasing physical inactivity in our lives.
- Therefore, a fundamental question of biology is, how and why does the body adapt to physical inactivity?

Conclusion

- The Abundance of Food has becomes our worst Enemy and our biggest Weapon of Mass Destruction
- Majority of Death Today accounts for what we eat or not Ate
- Our comfort (Inactivity) has become death bed and slowly Journeying to Death
- Our Metabolic Functionality decrease 10% per each decade from the age of 30 (Short et al., 2003).
- Our Cell Oxidative capacity decrease as we age hence Most disease catches up with us with our bodies are less capacitated.
WAR III:
METABOLIC DISORDER BATTLE

- In Ancient times people died of Wars, Leprosies, infections
- World War I & II Led to Millions of losses in Life world wide
- World War III: METABOLIC DISORDER WAR (diseases emanating from these disorders)
- It is a battle within us
- If not suffering from it, you either have a family member or friend from Metabolic disorder/chronic diseases.
- It requires Weapons needing NO Nuclear armaments
- The use of Exercise & Diet to Survive/Adapt
- “If we don’t end war, war will end us.” - H. G. Wells
- “Mankind must put an end to war -- or war will put an end to mankind” (John F Kennedy).
- Chronic Diseases are killings millions of people, will they end man or man put an end to habits that promote these diseases?

Graduates in 2018

MSc
1. Bonolo Masilo
2. Mashudu Matumba
3. Victoria Fasiku
4. Brian Munansangu
5. Simon Isaiah
6. Mogorosi Masenye

PhDs
1. Dr Betty Ayeleso
2. Dr Shesan John Owonubi
3. Trevor Nyakunda [WITS]
POST DOC FELLOWS 2015-2018 IN NWU

1. Dr Sandile Fuku
   (now Senior Lecturer in Biochemistry)
2. Dr Ademola Ayeleso
   (now Senior Lecturer in Biochemistry)
3. Dr Amil Zohir
4. Dr Dr Tella Toluwani
PhDs 2018

GLORY BE GOD WHO ALWAYS CAUSE TO TRIUMPH THROUGH CHRIST JESUS

2 CORINTH 2:14
References

PUBLICATIONS

Some of the Publications Since Joining NWU in October 2014- June 2018


    [http://dx.doi.org/10.1155/2015/515042](http://dx.doi.org/10.1155/2015/515042)