

Dietary Patterns Are Associated with Levels of Global Genomic DNA Methylation in a Cancer-Free Population



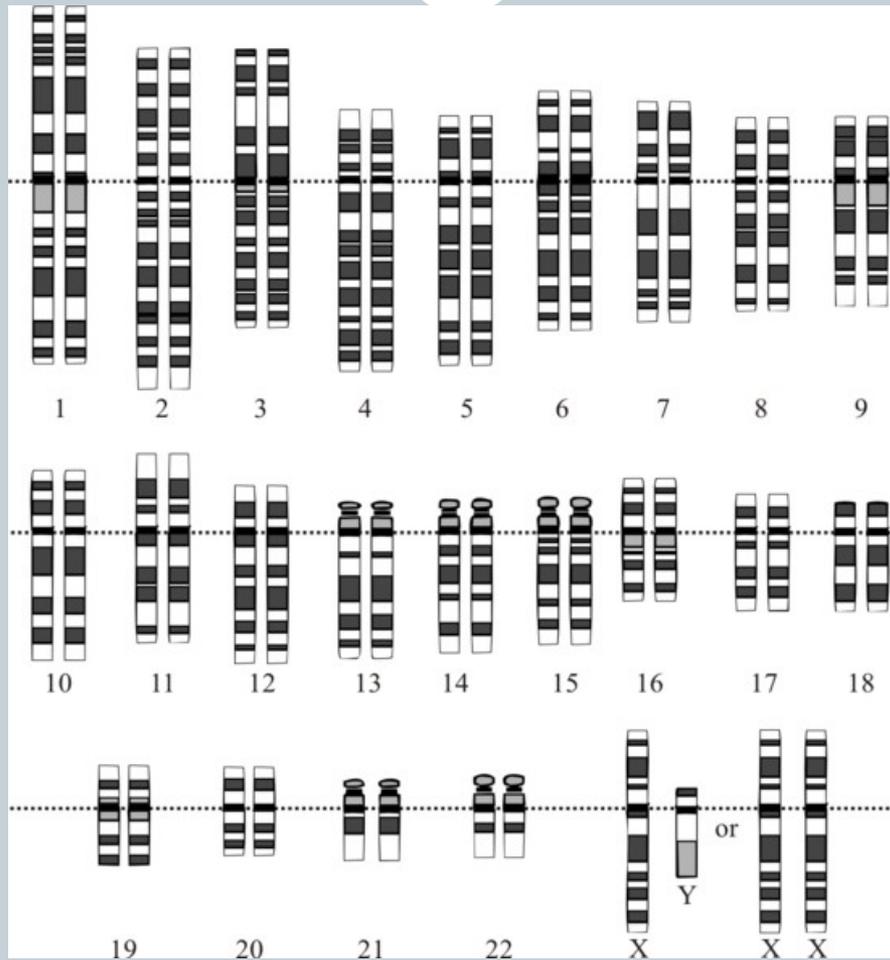
Presented By: Jamie Houdek and Kirstie Ducharme-Smith

Outline

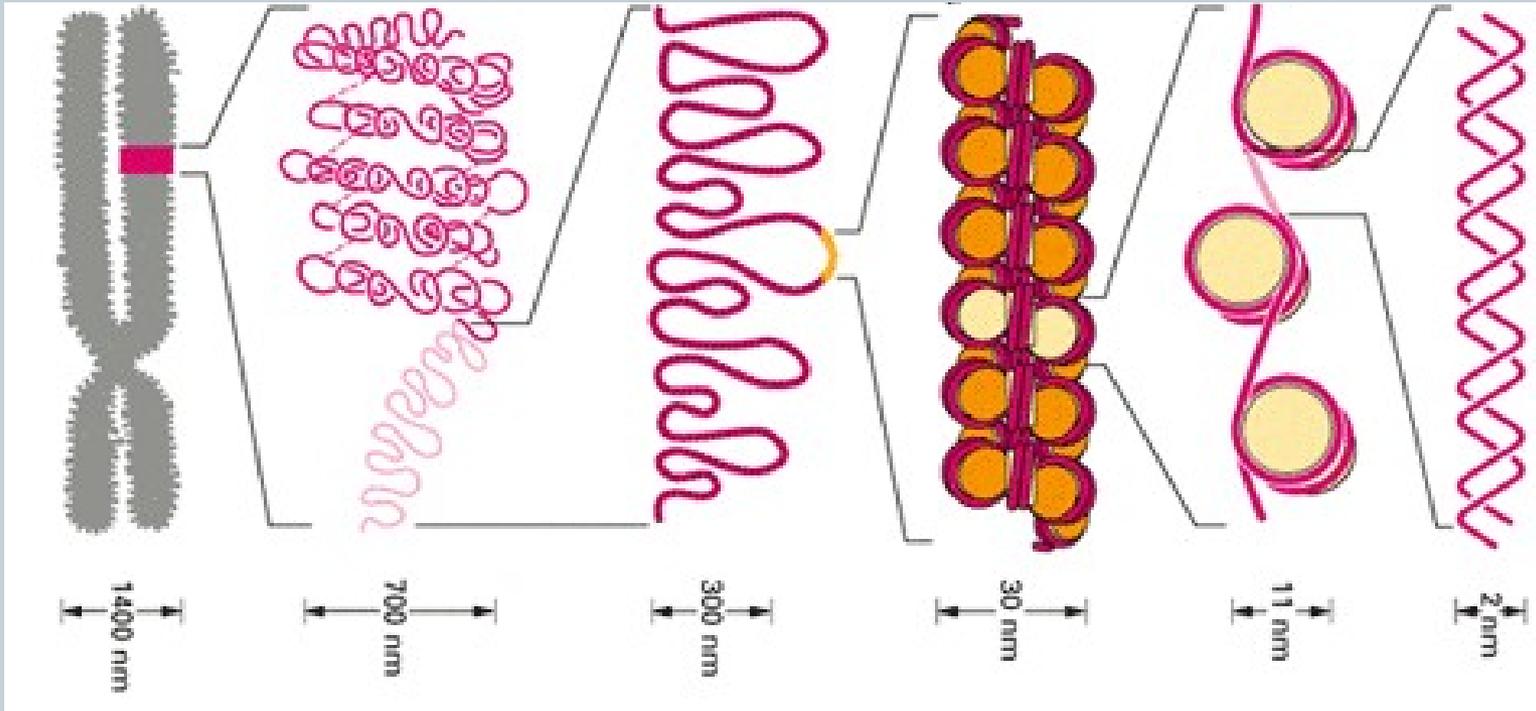


- **Background Information**
 - Human Genome and DNA Methylation
- **Study Objectives**
- **Materials and Methods**
 - Study Population
 - Dietary Assessment
 - Dietary Pattern Derivation
 - Global DNA Methylation
 - Statistical Analysis
- **Results**
 - Implications and Limitations
- **Conclusion**
- **Application to the Profession**
- **Questions??**

Human Genome



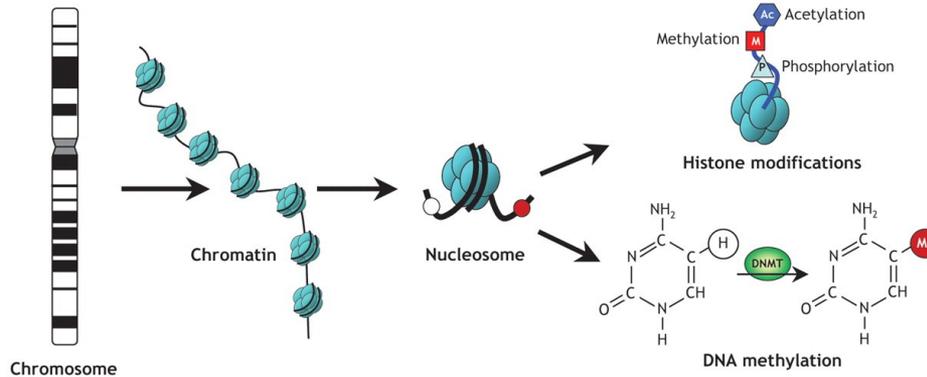
DNA Packaging



DNA Methylation



A



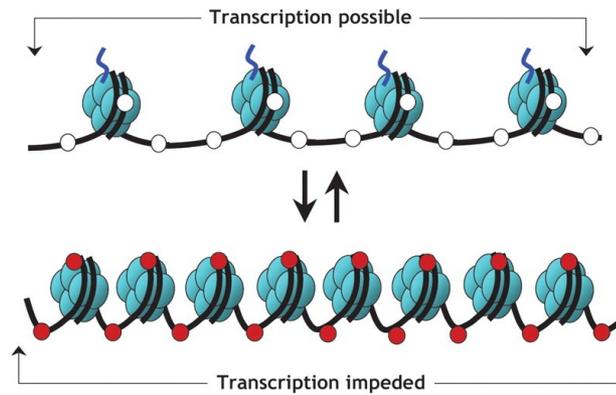
B

Gene "switched on"

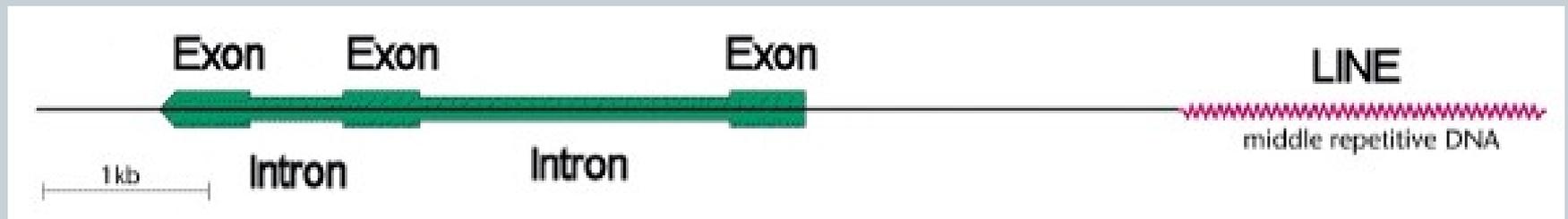
- Active (open) chromatin
- Unmethylated cytosines (white circles)
- Acetylated histones

Gene "switched off"

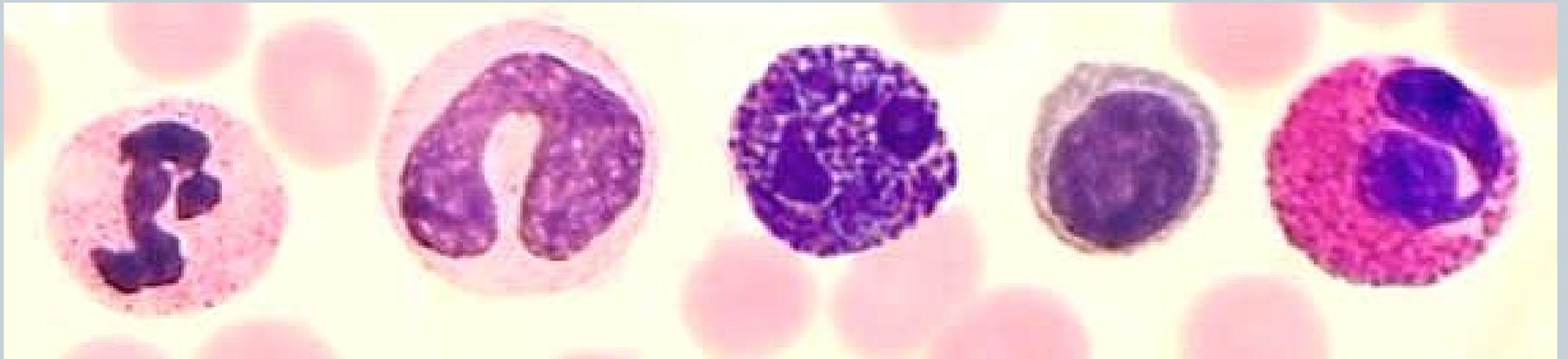
- Silent (condensed) chromatin
- Methylated cytosines (red circles)
- Deacetylated histones



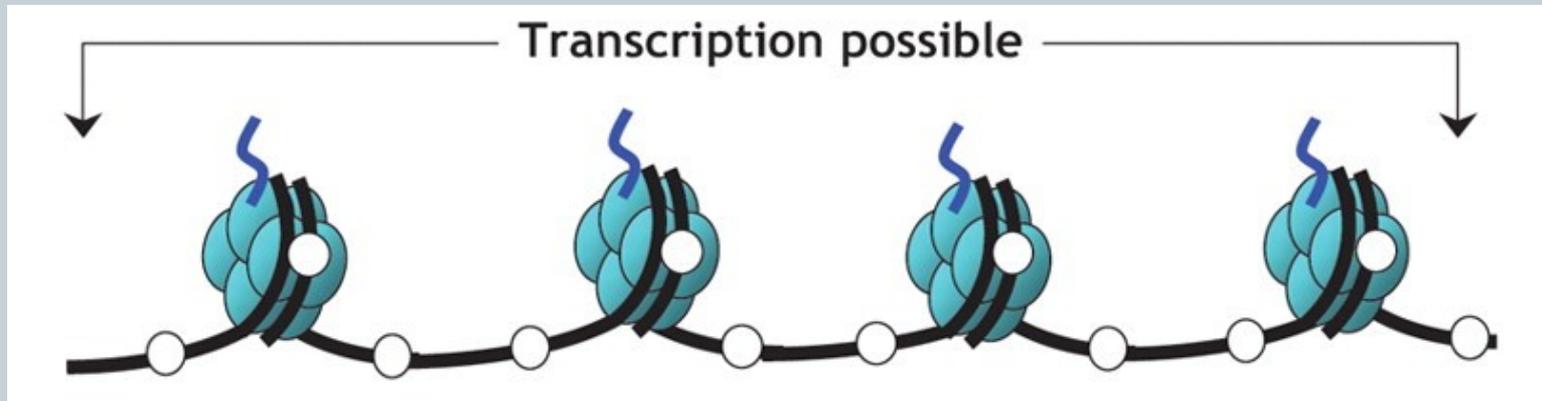
Long Interspersed Elements (LINEs)



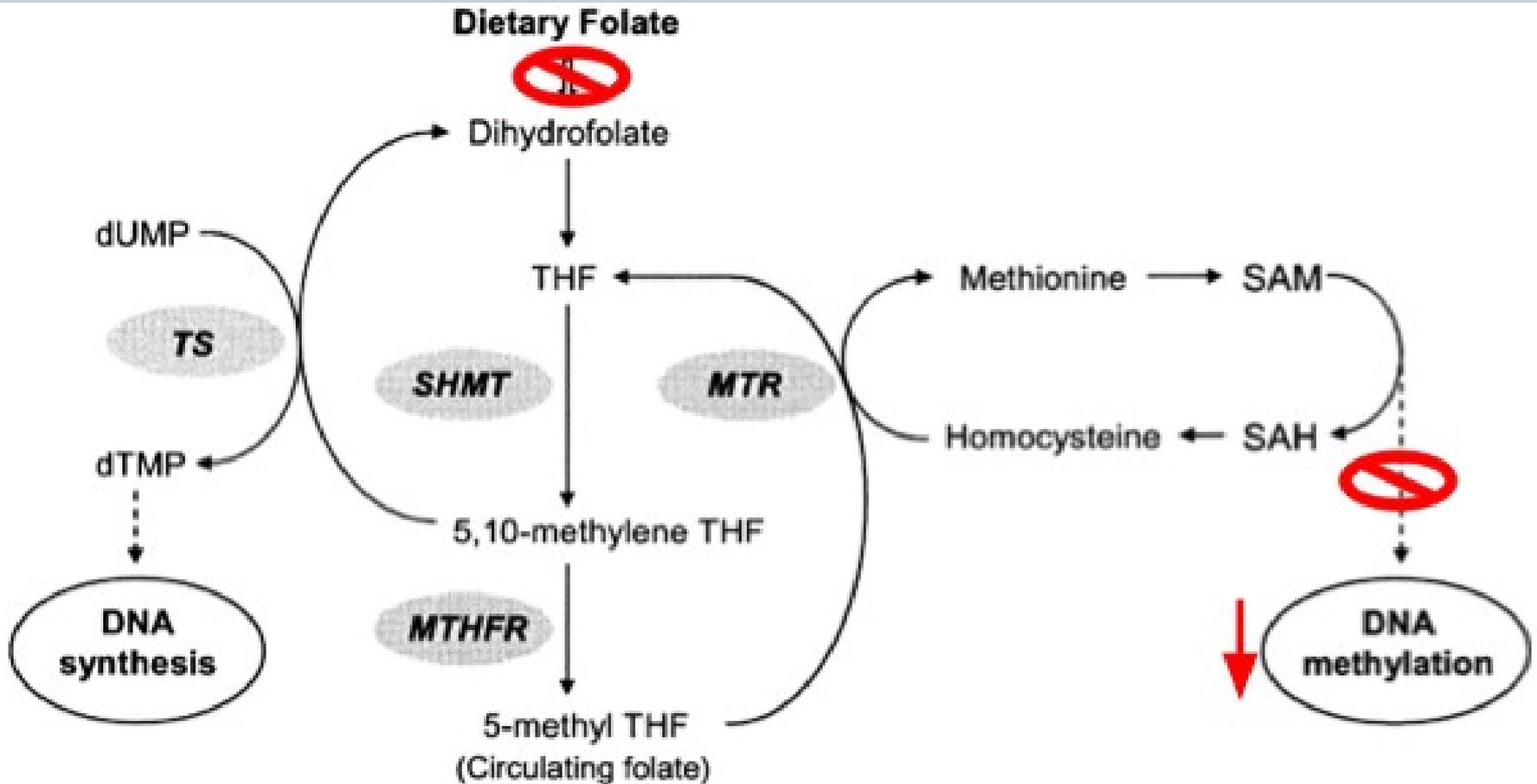
Leukocytes



Hypomethylation



Dietary Factors: Folate Deficiency



Limitations of Previous Studies



- Inconsistent findings in humans
 - *Many animal studies*

- Focus only on single nutrients
 - *Ignores the complex diet of humans*

Study Objectives



To examine the association between dietary patterns and leukocyte DNA methylation in a cancer-free population

Study Population



● Recruitment

- The North Texas Healthy Heart Study (2006)
- Assessed racial/ethnic differences in cardiovascular serum markers and calcium scores
- Inclusion Criteria:
 - Males and Females > 45 years old
 - Caucasian, Hispanic, African-American
 - No history of stroke, peripheral arterial disease, renal failure, heart failure or coronary disease, or cancer
- Second Study Visit (2008)
- 161 participants
- Survey
 - Measured BW, Ht, Waist and Hip Circumference
- Blood Sample

Dietary Assessment



- During the 2008 Visit:
- 108-item block Food Frequency Questionnaire (FFQ)
 - 8 Categories to assess frequency
 - 3 Categories to assess portion size
- Previously validated by three 24-hour recalls

Dietary Pattern Derivation



- Identified 13 food groups
 - Used exploratory factor analysis
- Individual Factor Scores
 - High score = High Adherence
 - Individuals categorized into 4 groups
 - High quartiles = High Factor Score = High Adherence

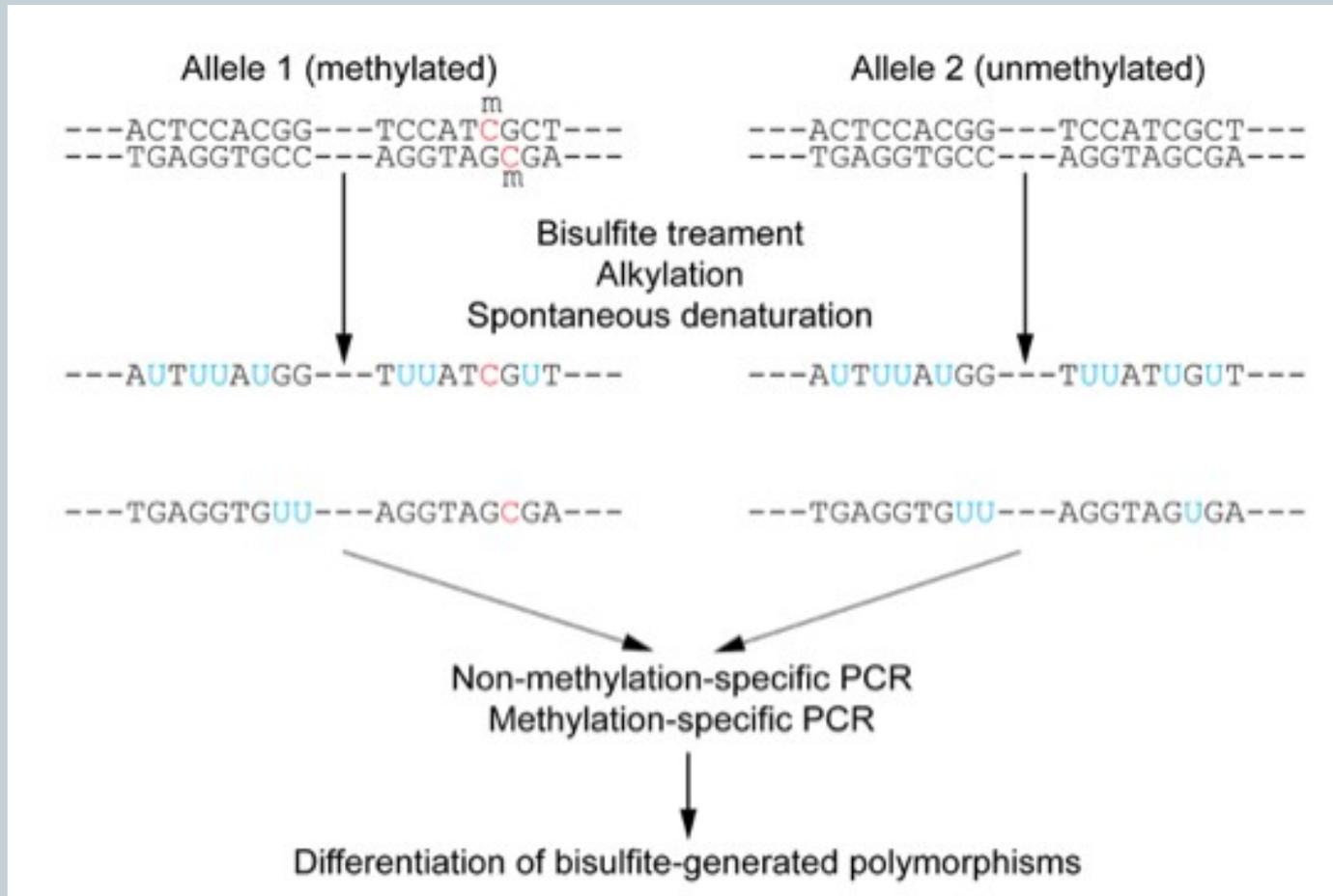
Global DNA Methylation



- Bisulfite conversion of DNA and pyro sequencing for LINE-1
 - Used sample of peripheral blood

- Assessment of DNA Methylation
 - Degree of methylation expressed for each DNA locus
 - $\% \text{ methylated cytosine} / (\text{methylated} + \text{unmethylated cytosine})$

Bisulfate Sequencing



Statistical Analysis



- **Chi-squares**

- characteristics of study population
- categorical variables

- **ANOVA**

- characteristics of study population
- continuous variables

- **Unconditional Logistic Regression**

- DNA methylation associated with each dietary pattern

Identification of 2 Dietary Patterns

TABLE 1 Factor loadings of 2 dietary patterns for 149 participants in the North Texas Healthy Heart Study, 2008

Food group intake, ¹ <i>unit/d</i>	Dietary patterns	
	Prudent	Western
Fruits, total including juice, <i>cup</i>	0.42	0.17
Vegetables not including legumes/potatoes, <i>cup</i>	0.97	0.19
Dark green vegetables, <i>cup</i>	<u>0.91</u>	-0.03
Orange vegetables, <i>cup</i>	<u>0.80</u>	0.07
Legumes and soy, <i>1-cup equivalent</i>	0.19	0.26
Potato, <i>cup</i>	0.31	<u>0.58</u>
Other vegetables including tomatoes, <i>cup</i>	<u>0.88</u>	0.28
Total grains, <i>oz. equivalent</i>	-0.06	<u>0.75</u>
Whole grains, <i>1-oz. equivalent</i>	0.14	<u>0.43</u>
Meat, <i>oz.</i>	0.06	<u>0.65</u>
Nuts and seeds, <i>1-oz. equivalent</i>	0.22	<u>0.43</u>
Dairy, <i>1-cup equivalent</i>	0.01	<u>0.47</u>
Oils, <i>tsp.</i>	0.40	<u>0.56</u>

¹ 1 cup or 1-cup equivalent (chopped vegetables and fruits) = 150 g; 1 oz. or 1-oz. equivalent (grains, meat, nuts, and seeds) = 28.3 g; 1-cup equivalent (dairy) = 237 mL; 1 tsp. (oils) = 4.93 mL.

Table 2 Presented by: Kelly Tasky



Characteristics of Study Population in Quartiles of the 2
Dietary Patterns

Population Characteristics



TABLE 2 Characteristics of the study population for quartiles of the 2 dietary patterns in the North Texas Healthy Heart Study, 2008¹

	Dietary patterns	Q1	Q2	Q3	Q4	P-value
<i>n</i>		37	37	37	38	
Age, <i>y</i>	Prudent	56.1 ± 6.6	57.8 ± 8.6	57.8 ± 8.2	57.3 ± 8.3	0.76
	Western	56.1 ± 7.4	56.6 ± 7.9	57.5 ± 7.1	58.6 ± 9.2	0.54
Male, <i>n</i> (%)	Prudent	13 (22.8)	17 (29.8)	15 (26.3)	12 (21.1)	0.60
	Western	10 (17.5)	12 (21.1)	16 (28.1)	19 (33.3)	0.16
Hispanic, <i>n</i> (%)	Prudent	10 (18.9)	14 (26.4)	17 (32.1)	12 (22.6)	0.34
	Western	18 (34.0)	17 (32.1)	10 (18.9)	8 (15.1)	0.02
Some college/college graduate, <i>n</i> (%)	Prudent	26 (26.5)	25 (25.5)	17 (17.4)	30 (30.6)	0.03
	Western	17 (17.4)	20 (20.4)	31 (31.6)	30 (30.6)	<0.001
BMI, ¹ <i>kg/m</i> ²	Prudent	31.5 ± 6.3	31.1 ± 4.6	30.5 ± 8.0	31.3 ± 6.9	0.93
	Western	30.7 ± 6.3	30.3 ± 5.2	29.7 ± 5.4	33.6 ± 8.2	0.04
Smoker, <i>n</i> (%)	Prudent	12 (21.4)	11 (19.6)	15 (26.8)	18 (32.1)	0.41
	Western	13 (23.2)	15 (26.8)	13 (23.2)	15 (26.8)	0.92
Drinker, <i>n</i> (%)	Prudent	22 (24.4)	21 (23.3)	19 (21.1)	28 (31.1)	0.23
	Western	21 (23.3)	20 (22.2)	27 (30.0)	22 (24.4)	0.34
Physically inactive, <i>n</i> (%)	Prudent	31 (27.4)	27 (23.9)	30 (26.6)	25 (22.1)	0.25
	Western	29 (25.7)	24 (21.2)	32 (28.3)	28 (24.8)	0.18

¹ Values are mean ± SD or *n* (%).

Table 3 Presented by: Olivia Moss



My Pyramid Food Groups and Nutrients Daily Intake by Levels of Leukocyte DNA Methylation

Examination of Food Items



TABLE 3 My Pyramid food groups and nutrients daily intake by levels of global leukocyte DNA methylation in the North Texas Healthy Heart Study population, 2008¹

	Global leukocyte DNA methylation		P-value
	Low	High	
n	72	77	
Food groups²			
Fruits total including juice, cup	0.72 (0.80)	0.63 (0.82)	0.74
Vegetables not including legumes/potatoes, cup	0.86 (0.97)	0.89 (1.14)	0.12
Dark green vegetables, cup	0.17 (0.36)	0.25 (0.39)	0.04
Orange vegetables, cup	0.07 (0.11)	0.08 (0.12)	0.33
Legumes and soy, 1-cup equivalent	0.08 (0.19)	0.11 (0.27)	0.16
Potato, cup	0.14 (0.18)	0.12 (0.14)	0.44
Other vegetables including tomatoes, cup	0.62 (0.65)	0.63 (0.68)	0.22
Total grains, 1-oz. equivalent	4.13 (3.31)	4.73 (3.36)	0.55
Whole grains, 1-oz. equivalent	0.77 (0.68)	0.91 (0.68)	0.56
Meat, oz.	2.86 (2.68)	2.95 (2.03)	0.50
Nuts and seeds, 1-oz. equivalent	0.21 (0.24)	0.24 (0.28)	0.74
Dairy, 1-cup equivalent	0.66 (0.88)	0.55 (0.72)	0.29
Oils, tsp.	1.67 (1.95)	2.11 (2.22)	0.44
Nutrients			
Protein, g/1000 kJ	9.34 (1.99)	9.00 (2.52)	0.66
Carbohydrate, g/1000 kJ	28.5 (5.83)	29.6 (5.99)	0.39
Fat, g/1000 kJ	9.55 (2.05)	9.13 (2.33)	0.58
Saturated fat, g/1000 kJ	2.86 (0.85)	2.57 (0.69)	0.02
Monounsaturated fat, g/1000 kJ	3.87 (0.87)	3.58 (1.23)	0.68
Polyunsaturated fat, g/1000 kJ	2.08 (0.68)	2.14 (0.72)	0.21
Dietary folate equivalents, $\mu\text{g}/1000 \text{ kJ}$	64.8 (26.4)	66.2 (22.8)	0.74
Dietary folate, $\mu\text{g}/1000 \text{ kJ}$	33.9 (23.2)	36.0 (16.9)	0.26
Supplemental folic acid, $\mu\text{g}/1000 \text{ kJ}$	37.7 (88.2)	43.3 (78.2)	0.45
Vitamin B-6, mg/1000 kJ	0.22 (0.08)	0.25 (0.10)	0.38
Riboflavin, mg/1000 kJ	0.26 (0.09)	0.24 (0.10)	0.08
Vitamin B-12, $\mu\text{g}/1000 \text{ kJ}$	0.64 (0.25)	0.51 (0.27)	0.08
Methionine, mg/1000 kJ	0.16 (0.06)	0.19 (0.06)	0.44

¹ Values are median (IQR) of daily intake of food groups or nutrients.

² 1 cup or 1-cup equivalent (chopped vegetables and fruits) = 150 g; 1 oz. or 1-oz. equivalent (grains, meat, nuts, and seeds) = 28.3 g; 1-cup equivalent (dairy) = 237 mL; 1 tsp. (oil) = 4.93 mL.

Table 4 Presented by Molly DePrengner



Association Between Dietary Patterns and Levels of Global Leukocyte DNA Methylation

Association of Dietary Patterns



TABLE 4 The association between dietary patterns and levels of global leukocyte DNA methylation in the North Texas Healthy Heart Study population, 2008

	Global DNA methylation		OR (95%CI) (crude)	OR (95%CI) (step-wise selection)
	Low (< median)	High (\geq median)		
Prudent dietary pattern	<i>n (%)</i>			
Q1 (< -0.73)	25 (34.7)	12 (15.6)	1.00	1.00
Q2 (-0.73, -0.26)	17 (23.6)	20 (26.0)	0.41 (0.16-1.05)	0.38 (0.41-1.04)
Q3 (-0.26, 0.40)	15 (20.8)	22 (28.6)	0.33 (0.13-0.85)	0.38 (0.14-1.04)
Q4 (\geq 0.40)	15 (20.8)	23 (29.9)	0.31 (0.12-0.81)	0.33 (0.12-0.92)
			<i>P</i> -trend = 0.02	<i>P</i> -trend = 0.04
Western dietary pattern				
Q1 (< -0.66)	18 (25.0)	19 (24.7)	1.00	1.00
Q2 (-0.66, -0.16)	16 (22.2)	21 (29.2)	0.80 (0.32-2.01)	1.10 (0.40-3.01)
Q3 (-0.16, 0.46)	19 (26.4)	18 (23.4)	1.14 (0.45-2.77)	1.39 (0.52-3.73)
Q4 (\geq 0.46)	19 (26.4)	19 (24.7)	1.06 (0.43-2.31)	1.28 (0.47-3.47)
			<i>P</i> -trend = 0.74	<i>P</i> -trend = 0.55

Study Implications



- **Dietary Pattern and Health Implications**
 - Cohort Study
 - Nurses' Health Study

- **Median Level of LINE-1 Methylation**
 - Cancer-free population (73.7%)
 - Breast cancer case-control study (73.5%)
 - Head and neck squamous cell carcinoma case-control study (74.7%)

- **Lower Dietary Folate Intake and Methylation?**

Study Limitations



- Cross-sectional design
- Factor analysis
- Population characteristics

Conclusions



- **Author's Conclusion:**

- A dietary pattern characterized by a high intake of vegetables and fruits may protect against global DNA hypomethylation

- **Our Conclusions:**

- Hypomethylation may be impacted by dietary intake of individual
- More research should explore the relationship between dietary patterns and global DNA hypomethylation

Application to the Profession



- Intervention on the overall dietary pattern rather than on single nutrients or food groups may be a more effective way to protect against cancer risk through epigenetic regulation
- Recommendation of a Prudent diet
 - loaded heavily on vegetables
 - dark green, orange, and other non-starchy vegetables
 - loaded heavily on fruits

Questions??



Thank you for Listening!!

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