# The Mediating Effect of Triglyceride/High Density Lipoprotein Cholesterol Ratio on Inflammatory Markers and the Metabolic Syndrome in the Postmenopausal Women

*7/ 25 / 2015*

*Jen-Chen Tsai, DNSc., Professor*
*School of Nursing, National Yang-Ming University*
*Taipei, Taiwan*
*jenchent@ym.edu.tw*

# Disclosures

- **Authors name**

  Jen-Chen,Tsai; Chia-Hui, Wang; Min-Huey, Chung

- **Learner objectives**
  - The leaners will be able to identify the risk factors of metabolic syndrome in postmenopausal women
  - The learner will be able to understand the important role of iterleukin-6 and triglyceride/high density lipoprotein cholesterol ratio in metabolic syndrome among postmenopausal women

- There is no conflict of interest in the present study

# Content Outline

- Introduction
- Purposes of study
- Methods
- Results
- Conclusions

# Introduction

- The metabolic syndrome (MS) is a constellation of metabolic abnormalities including abdominal obesity, dyslipidemia, hypertension, and impaired glucose tolerance.
- The MS has 2-fold increase in risk of cardiac vascular diseases and cardiac related mortality.
- The prevalence of MS increases with age, affecting up to 40% of the U.S. population older than 50 years and approaching 50% in postmenopausal women.

*(Ackermann et al., 2011; Joseph, Prigeon, Blumenthal, Ryan, & Goldberg, 2011)*

# Introduction

- The high incidence of MS in postmenopausal women is corroborated by numerous factors:
  - Sedentary lifestyles
  - Weight gain and central obesity facilitated by low levels of estrogens
  - Dyslipidemia
  - Decreased glucose tolerance
  - Elevated blood pressure
  - Increased pro-inflammatory states

*(Lee et al., 2009)*

# Introduction

- The intereulin-6 (IL-6) is a pro-inflammatory cytokine produced by adipose tissue and mediates many metabolic changes in the development of insulin resistance and atherosclerosis during menopausal transition.
- In comparison with other inflammatory makers such as TNF-α or IL-1β, increased levels of IL-6 has been observed in postmenopausal women.

*(Ackermann et al., 2011; Dinh et al., 2009; Kim et al., 2012; Saremi et al., 2009).*

# Introduction

- Adiponectin is an adipocyte-specific protein, which may enhance insulin sensitivity through increased fatty acid oxidation and inhibition of hepatic glucose production.
- Low circulating levels of adiponectin is associated with insulin resistance and the development of MS .

*(Ackermann et al., 2011; Dadson, et al., 2011;Zhuo et al., 2009).*

# Introduction

- More recently, the triglyceride/HDL- cholesterol ratio (TG/HDL-C ratio) has showed to be the best marker of dyslipidemia associations with the components of MS in both men and women.
- Insulin resistance has been shown to predict the development of dyslipidemia and the MS.

*(Kang et al., 2012; Kawamoto et al., 2011; Salazar et al., 2012)*

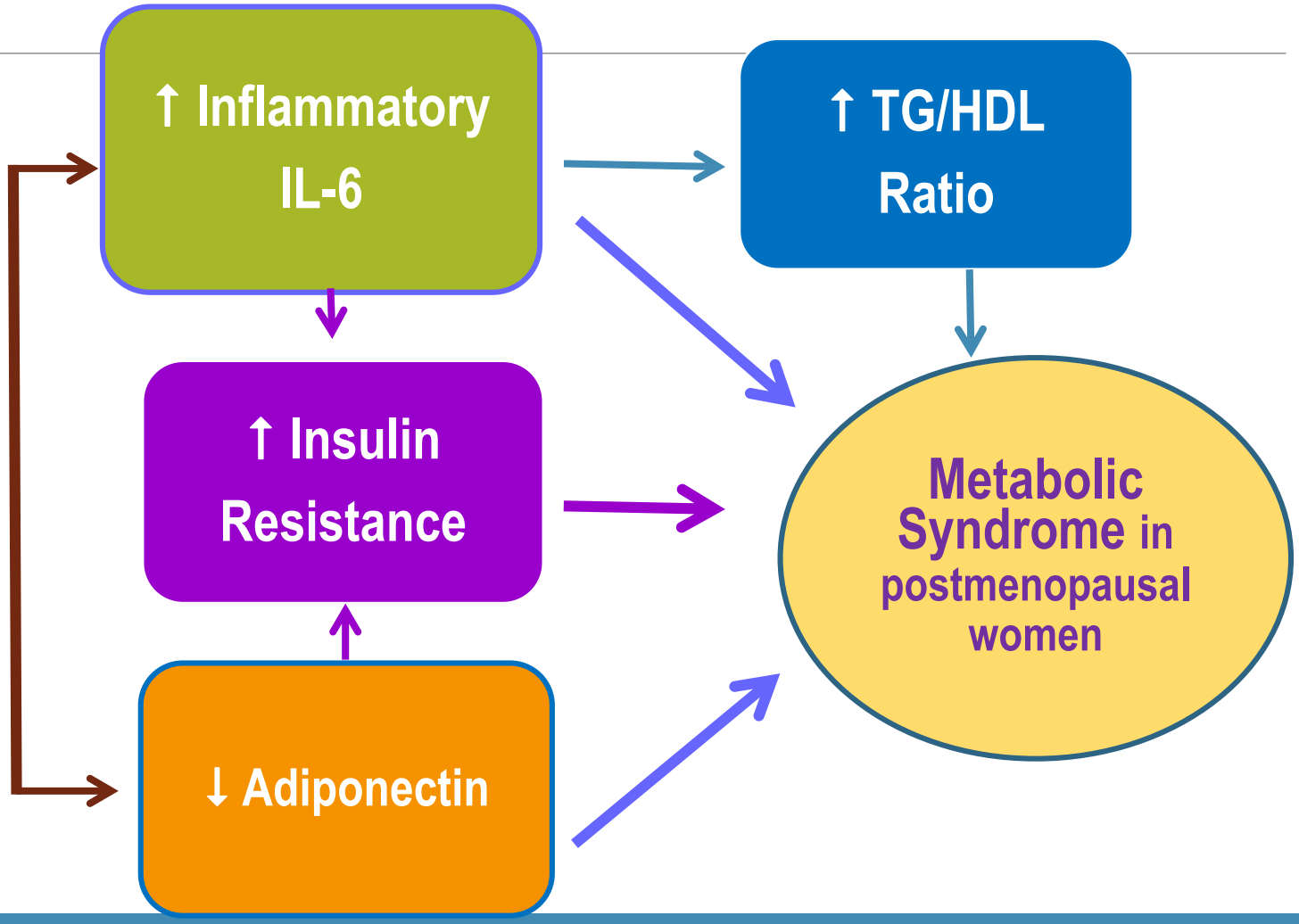
# Introduction

- The relationships among levels of IL-6, adiponectin, lipoprotein ratio, insulin resistance, and MS in postmenopausal women have not been well explored.
- Few studies have investigated the most prominent metabolic changes related to the development of MS especially in postmenopausal women.

# Purposes of the Study

- To explore the associations among inflammatory marker (mainly the IL-6), adiponectin, TG/HDL-C ratio, insulin resistance and MS in postmenopausal women
- To Identify the most prominent risk indicators of MS among postmenopausal women
- To test whether the TG/HDL-C ratio or Insulin resistance carry mediating effects on the influence of IL-6 and adiponectin on the MS in postmenopausal women

# Metabolic Changes in Postmenopausal Women



# Methods

- A cross-sectional research was performed in this study.
- Postmenopausal women were recruited in health clinics of a university-based medical center in Northern Taiwan.
- The study protocol was approved by the ethics committee of the study medical center. Informed consent was obtained from each study participant.

# Participants

- Inclusion criteria
  - Postmenopausal women aged 45-70 years and had not menstruated for at least one year *(Bhagat et al., 2010).*
  - Were not taking medications including lipid-lowering agents, anti-hypertension drugs, non-steroid anti-inflammatory drugs (NSAIDs) or hormone therapy.
- Exclusion criteria
  - Menopause status due to surgery or medical problems
  - The presence of chronic heart failure, clinical signs of ischemic heart disease, or evidence of cancer

# Define Metabolic Syndrome

- Metabolic syndrome in females was defined by the presence of 3 or more of the following components:
  - Waist circumference >80 cm
  - SBP ≥130mmHg or DBP ≥ 85mmHg
  - TG >150 mg/dL
  - HDL-C <50 mg/dL
  - Fasting glucose ≥ 100 mg/dL

*The NCEP/ATP III criteria of the MS (Cleeman et al., 2001) were modified for Asian population (Cheung et al., 2008; Health Public Administration; 2007).*

# Metabolic Score

- Metabolic score is the sum of risk components of MS that an individual has.
- The MS score ranges from 0 (non of the risk component) to 5 (presence of all 5 components of the MS).
- The higher MS score represents the more severe of the MS.

(Solymoss, 2003, Vidal, 2005)

# Anthropometric Measurements

- Height, body weight, and waist circumferences of participants were determined following the guidelines recommended by the ACSM (2012).
- Waist circumferences were assessed twice using a Gulick anthropometric tape measure *(Creative Health Products, Plymouth, MI)* at the minimum circumference (under clothing and next to the subject's skin). The average of the readings was obtained for data analysis

# Laboratory analyses

- The concentrations of TG, HDL-C, and fasting glucose were analyzed in the hospital laboratory using standard equipment and assays, than the TG/HDL-C ratio was calculated.

# Laboratory Analyses

- Fasting insulin was measured by Human Insulin Kits *(Mercodia, Uppsala, Sweden).* The intra-assay and inter-assay coefficients of variability (CV) of insulin were 3.5% and 4% in our study.
- Homeostatic model assessment estimate for insulin resistance (HOMA-IR) was used to measure insulin resistance.
- The value of HOMA-IR was calculated by the formula: fasting insulin (μU/mL) × fasting glucose (mg/dL)/405

*(Metthews et al., 1985)*

# Laboratory Analyses

- The concentration of IL-6 of the subjects was determined using enzyme-linked immunosorbent assay (ELISA) kits *(R&D Systems, Minneapolis, USA)*. The intra-assay and inter-assay CV were 6% and 8% in our study.
- The serum adiponectin was determined using commercial available ELISA kits *(Linco Systems, Minneapolis, USA)*. The intra-assay and inter-assay CV were 5.4% and 6% in our study.

# Procedures

- Participants reported to the laboratory in the morning. Blood samples were collected after an overnight fast and abstained from caffeine, tea, and alcohol for 12 hours.
- Cardiovascular measurements were performed after at least 10 minutes of rest in the supine position in a quiet, temperature-controlled room (approximately 24°C).

# Data Analysis

- Hierarchical multiple regressions were used to analyze the effect of IL-6, adiponectin, insulin resistance and TG/HDL-C ratio on the MS score.
- A forward hierarchical procedure was applied for the model selection.
- The bootstrap resampling procedure was used to test the significance of the specific effects of measured variables on the MS.

# Results (1)

- One hundred and two women meet the study criterion.
- Eighty-nine women (mean age: 56.9±6.2 years) consent to participate in this study.
- 58 women (65%) had MS according to the study criteria.
- Major risk indicators of MS for postmenopausal women include: greater mean waist circumference, increased fasting glucose, higher levels of IL-6 and insulin resistance , but low levels of HDL-C and adiponectin.

# Table 1 Metabolic risks of postmenopausal women in the study (n=89)

| | ALL (n=89) | MS (n=58) | Non-MS (n=31) | p |
|---|---|---|---|---|
| **Age** (years) | 56.9±6.2 | 58.1±6.1 | 54.8±5.9 | 0.02 |
| **BMI** (kg/m$^2$) | 26.0±3.3 | 26.6±3.2 | 24.8±3.3 | 0.02 |
| **IL-6** (pg/ml) | 3.1±1.8 | 3.7±1.9 | 2.0±1.1 | <0.01 |
| **HOMA-IR** | 2.7±2.2 | 3.4±2.4 | 1.4±1.0 | <0.01 |
| **Adiponectin** (ug/ml) | 11.8±6.5 | 9.2±3.7 | 16.2±7.9 | <0.01 |
| **TG/HDL-C Ratio** | 3.1±1.9 | 3.8±2.0 | 2.1±1.0 | <0.01 |

BMI, body mass index; IL-6, intereulin-6; HOMA-IR, Homeostasis model assessment insulin resistance; TG, triglyceride; HDL, high density lipoprotein

# Table 1 Metabolic risks of postmenopausal women in the study (n=89)

| | ALL (n=89) | MS (n=58) | Non-MS (n=31) | p |
|---|---|---|---|---|
| **MS Components** | | | | |
| **WC** (cm) | 90.1±9.4 | 93.0±9.0 | 85.2±7.9 | <0.01 |
| **SBP**(mmHg) | 124.1±12.7 | 127.0±13.0 | 119.3±10.8 | <0.01 |
| **DBP**(mmHg) | 74.8±9.4 | 76.5±9.0 | 72.1±9.6 | 0.05 |
| **Fasting glucose** (mg/dL) | 104.9±13.6 | 108.4±14.9 | 98.9±8.2 | <0.01 |
| **HDL-C** (mg/dL) | 48.8±11.9 | 43.5±7.8 | 57.7±12.4 | <0.01 |
| **TG** (mg/dL) | 138.2±57.8 | 152.6±60.3 | 113.9±44.6 | <0.01 |
| **MS Score** | 3.0±1.1 | 3.7±0.7 | 1.8±0.4 | <0.01 |

WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure

# Table 2 Correlations among IL-6, HOMA-IR, adiponectin, TG/HDL-C ratio and MS score in postmenopausal women

| | Age | BMI | IL-6 | Adipon-ectin | HOMA | TG/LDL-C |
|---|---|---|---|---|---|---|
| Age | 1.00 | | | | | |
| BMI | -0.09 | 1.00 | | | | |
| IL-6 | 0.10 | 0.25* | 1.00 | | | |
| Adiponectin | -0.17 | -0.20 | -0.29* | 1.00 | | |
| HOMA-IR | 0.01 | 0.51** | 0.28** | -0.21 | 1.00 | |
| TG/HDL-C ratio | -0.20 | 0.36* | 0.48** | -0.31** | 0.37** | 1.00 |
| MS Score | 0.24* | 0.37* | 0.42** | -0.48** | 0.52** | 0.50** |

** p< 0.01; * p< 0.05.

# Results (2)

- The MS score was significantly positive correlated with age, BMI, insulin resistance, TG/HDL-C ratio and IL-6 levels, and was negative correlated with adiponectin.
- IL-6 level was significantly positive correlated with BMI, insulin resistance, TG/HDL-C ratio and negative correlated with adiponectin.
- Adiponectin was not significant correlated with insulin resistance but was significantly negative correlated with IL-6 and TG/HDL-C ratio.

# Table 3 Hierarchical regression analysis predicting MS among IL-6, HOMA-IR, adiponectin, and TG/HDL-C ratio (n=89)

| | B | SE | β | t | p | $R^2$ |
|---|---|---|---|---|---|---|
| **Step 1** | | | | | | **0.23** |
| Age | 0.04 | 0.02 | 0.24 | 2.56 | 0.01 | |
| BMI | 0.12 | 0.04 | 0.36 | 3.65 | <0.001 | |
| **Step 2** | | | | | | **0.31** |
| Age | 0.04 | 0.02 | 0.22 | 2.04 | 0.04 | |
| BMI | 0.10 | 0.03 | 0.29 | 2.78 | 0.01 | |
| Level of IL-6 | 0.18 | 0.06 | 0.30 | 2.83 | 0.01 | |
| **Step 3** | | | | | | **0.40** |
| Age | 0.04 | 0.02 | 0.21 | 2.22 | 0.04 | |
| BMI | 0.04 | 0.04 | 0.12 | 1.13 | 0.26 | |
| Level of IL-6 | 0.15 | 0.06 | 0.26 | 2.55 | 0.01 | |
| HOMA-IR | 0.18 | 0.06 | 0.36 | 3.13 | <0.001 | |

| | B | SE | β | t | p | $R^2$ |
|---|---|---|---|---|---|---|
| Step 4 | | | | | | 0.46 |
| Age | 0.03 | 0.02 | 0.18 | 2.00 | 0.06 | |
| BMI | 0.03 | 0.04 | 0.10 | 0.98 | 0.33 | |
| Level of IL-6 | 0.12 | 0.06 | 0.20 | 2.03 | 0.04 | |
| HOMA-IR | 0.17 | 0.05 | 0.34 | 3.12 | <0.001 | |
| Adiponectin | -0.05 | 0.02 | -0.27 | -2.75 | 0.01 | |
| Step 5 | | | | | | 0.52 |
| Age | 0.05 | 0.02 | 0.27 | 2.85 | 0.01 | |
| BMI | 0.02 | 0.03 | 0.07 | 0.64 | 0.53 | |
| Level of IL-6 | 0.05 | 0.06 | 0.08 | 0.74 | 0.47 | |
| HOMA-IR | 0.14 | 0.05 | 0.29 | 2.75 | 0.01 | |
| Adiponectin | -0.04 | 0.02 | -0.22 | -2.30 | 0.03 | |
| TG/HDL-C ratio | 0.19 | 0.06 | 0.32 | 2.93 | 0.01 | |

# Results (3)

- Results from the hierarchical multiple regression models showed that age, insulin resistance, adiponectin and TG/HDL-C ratio were significantly associated with the MS in postmenopausal women.
- After entering TG/HDL-C ratio into the regression model in step 5, the correlation between IL-6 and MS score became non-significantly, indicating TG/HDL-C ratio mediate the effect of IL-6 on the MS.

## Table 4. Mediating Model of IL-6, TG/HDL-C ratio and MS when controlling for age, BMI (n=89)

| Dependent variable | Predictor | B | SE | p | Indirect effect | Bootstrap with bias-corrected 95% CI | Percentage mediated |
|---|---|---|---|---|---|---|---|
| MS | IL-6 | 0.19 | 0.06 | <0.01 | | | |
| TG/HDL-C ratio | IL-6 | 0.46 | 0.10 | <0.001 | | | |
| MS | IL-6 | 0.08 | 0.06 | 0.22 | 0.11 | 0.05 -0.22 | 0.41 |
| | TG/HDL-C ratio | 0.24 | 0.07 | <0.01 | | | |
| Covariates | | | | | | | |
| | Age | 0.06 | 0.02 | <0.01 | | | |
| | BMI | 0.07 | 0.03 | 0.04 | | | |

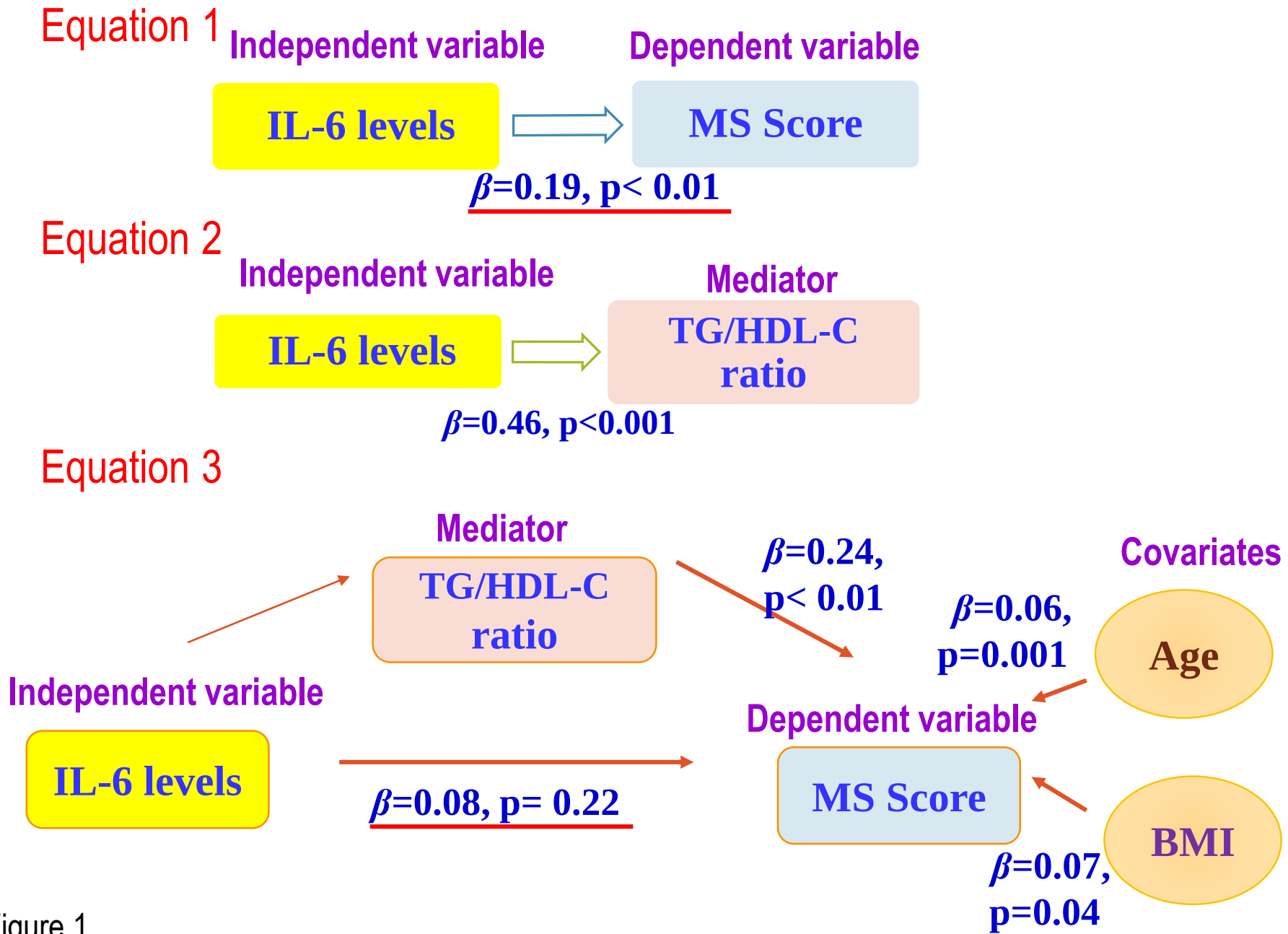



Figure 1

Mediation models of the MS on the relationship between IL-6 and TG/HDL-C of the postmenopausal women (N = 89)

# Results (4)

- Results from the bootstrap resampling procedure confirmed that IL-6 levels had a significant indirect effect on MS, with TG/HDL-C ratio carrying the influence of IL-6 on the dependent variable of MS.
- No additional mediating effect of insulin resistance carry the influence of IL- 6 and adiponectin on MS.

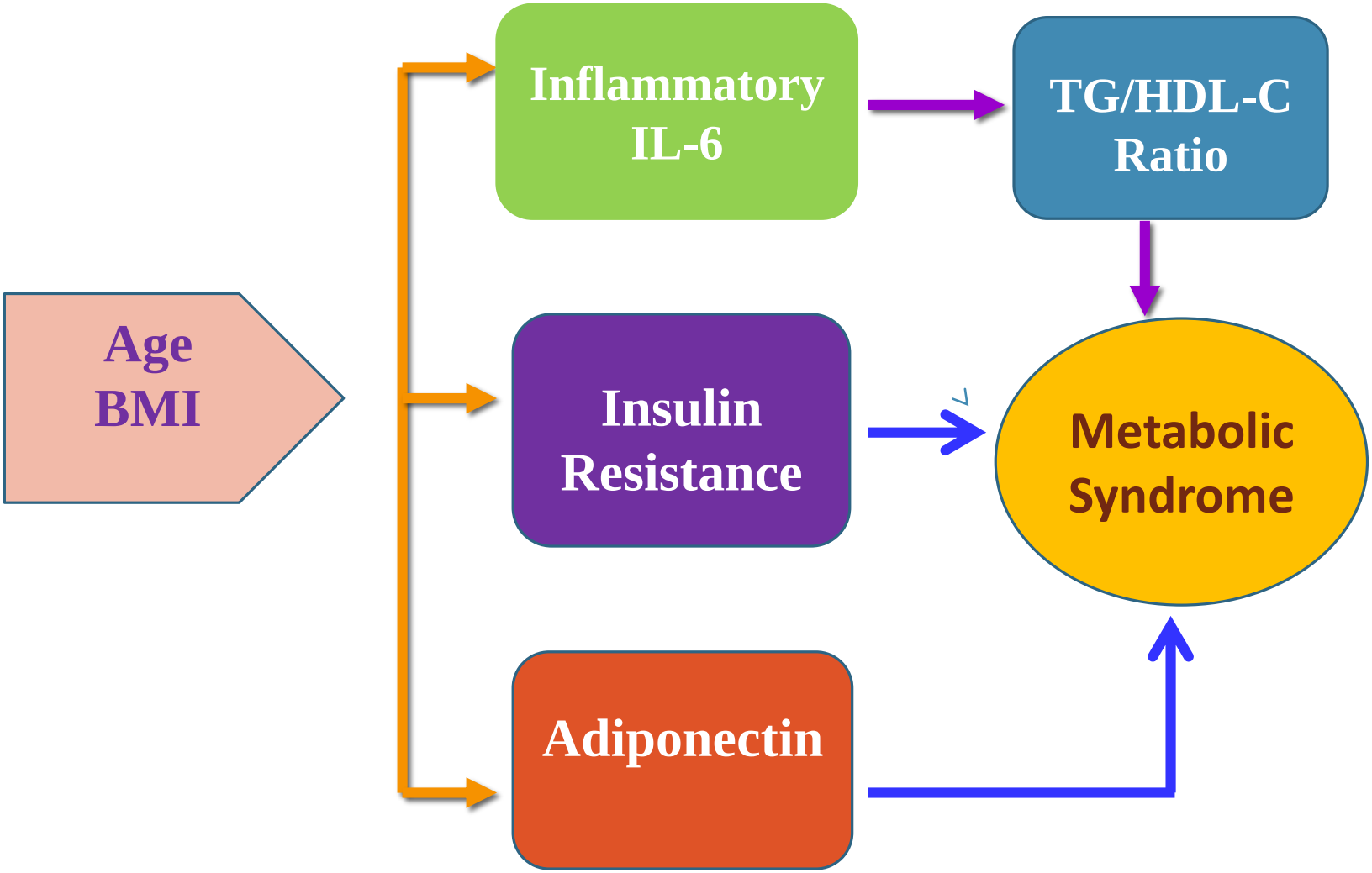



Figure 2 A framework of related factors of MS among postmenopausal women

# Conclusion

- We found significant correlations of serum IL-6 concentrations with BMI, HOMA-IR, TG/HDL-C ratio, adiponectin, and MS score in postmenopausal women.
- The TG/HDL-C ratio is a mediator of the effect of IL-6 on MS.
- Result of this study supports the important role of IL-6 and TG/HDL-C ratio in the development of MS among postmenopausal women.

# Conclusion

- In addition to the components of MS proposed by "*National Cholesterol Education Program Adult Treatment Panel III*" (2011), the concentration of IL-6 and adiponectin should be evaluated in females during postmenopausal stage.
- The intervention to improve inflammatory state, adiponectin, the TG/HDL-C ratio would be an important primary and secondary prevention for health promotion in postmenopausal women.

# *Thank you*