WNT10B Polymorphism in Korean Stroke Patients with Yin Deficiency Pattern

Mi Mi Ko, Tae-Yong Park, Ji Hye Lim, Min Ho Cha, and Myeong Soo Lee

Medical Research Division, Korea Institute of Oriental Medicine, 1672 Yuseongdae-ro, Yuseong-gu, Daejeon 305-811, Republic of Korea

Correspondence should be addressed to Min Ho Cha, mhchamin@kiom.re.kr

Received 29 May 2012; Accepted 13 June 2012

Copyright © 2012 Mi Mi Ko et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

WNT10B has been indicated as a potential regulator of adipogenesis in vivo and in vitro models of obesity. In this study, we analyzed the distribution of WNT10B polymorphism in elderly Korean subjects with cerebral infarction (CI) and Yin Deficiency pattern and Non-Yin Deficiency pattern. A total of 630 CI patients, including 75 with Yin Deficiency pattern and 555 with Non-Yin Deficiency pattern, participated in this study. SNP (G-607C) genotyping was conducted by primer extension using TaqMan probe; five percent of subjects were regenotyped by direct sequencing to confirm the accuracy of the genotyping. The results were analyzed using a multiple logistic regression model to evaluate the genetic association between the G-607C variant and Yin Deficiency pattern. The frequency of the CC genotype of G-607C in the Yin Deficiency pattern group (29.33%) was significantly higher than that in the Non-Yin Deficiency pattern group (23.96%) ($P = 0.0339$, OR = 2.005 (1.054–3.814)) in a recessive model. This is the first study to demonstrate an association between a WNT10B polymorphism and the Yin Deficiency pattern of traditional Korean medicine (TKM) in a CI patient population. These results suggest that G-607C might be used as a diagnostic genetic marker for Yin Deficiency pattern in stroke patients and in the development of personalized medical care.

1. Introduction

Patients with a specific disease exhibit various phenotypes, signs, and symptoms reflecting the cause, nature, and location of the illness, the patient's physical condition, and the patient's treatment. Traditional Korean medicine (TKM), similar to traditional Chinese medicine (TCM), categorizes these phenotypes as patterns, and the procedure for determining the pattern of a particular patient is called “pattern identification” (PI) [1, 2]. Previous reports have described the PI process for differentiating stroke victims with four TKM types: the Fire-Heat pattern, Dampness-Phlegm pattern, Yin Deficiency pattern, and Qi Deficiency pattern [3, 4].

The Yin Deficiency pattern indicates a pattern/syndrome resulting from a deficiency of yin fluid and essence, incapable of restraining yang [1]. Furthermore, the Yin Deficiency pattern is typically associated with thinness [5–7]. Conversely, patients with the Qi Deficiency or Dampness-Phlegm patterns tend to be overweight or obese [7, 8].

PI is affected by various environmental factors, such as climate, diet, and lifestyle [9]. However, some reports have shown that inherent factors, such as genetic variations, are correlated with PI [10–14]. For example, a study in Chinese patients with coronary heart disease reported that the frequencies of the $\varepsilon 4$ allele and $\varepsilon 3/4$ genotype of the apolipoprotein E gene were significantly higher in patients with the phlegm pattern than in patients with the blood stasis pattern [10]. Recently, we reported that L55M and C-2033T alleles of the Paraoxonase 1 (PON1) gene, which were correlated with stroke in an East Asian population, were associated with the Dampness-Phlegm pattern among Korean stroke patients [12]. Around the same time, our research group also reported that the C-399T variation of the neuropeptide Y (NPY) gene, which were correlated with stroke in an East Asian population, were associated with the Dampness-Phlegm pattern among Korean stroke patients [13]. However, few genetic polymorphisms have been associated with the Yin Deficiency pattern.

Wingless-type MMTV integration sites (WNTs) are secreted glycoproteins that function as signaling molecules and are involved in numerous events in human organogenesis, physiology, and pathology [15]. WNT10B, one member of the Wnt family, was first found in human breast
carcinoma [16], and several studies have been performed to identify the relationship between WNT10B and human diseases [17]. The Kirikoshi study group found that WNT10B expression was upregulated in various carcinoma cells [18–22], and Christodoulides et al. [23] and Kim et al. [24] reported that WNT10B is negatively correlated with obesity.

WNT10B is located on chromosome 12q13, and six tightly linked single nucleotide polymorphisms (SNPs) in this gene have been identified in Korean women [24]. Three of these SNPs are in promoter regions: one is in the 5′-untranslated region, and two synonymous SNPs are in exon 5 and 6. Among the three SNPs in the promoter, G-607C has functional activity, modulating the transcriptional activity of WNT10B by altering the binding activity of some transcription factors.

In this study, we analyzed the distribution of G-607C genotypes in elderly Korean subjects with Yin Deficiency pattern, which is clinically different from other patterns.

2. Materials and Methods

2.1. Study Subjects. The data for this analysis were collected as parts of the “The Fundamental Study for the Standardization and Objectification of Pattern Identification in TKM for Stroke (SOPI-Stroke)” project of the Korean Institute of Oriental Medicine (KIOM) [3, 4].

The study group was composed of patients with cerebral infarction (CI) who were admitted to one of the thirteen Korean oriental medical hospitals participating in this study from November 2006 to March 2011. To be eligible for the study, participants had to be diagnosed with stroke within 30 days of symptom onset as confirmed by imaging, such as computerized tomography (CT) or magnetic resonance imaging (MRI). Patients with traumatic stroke, such as subarachnoid, subdural, or epidural hemorrhage, were excluded from this study.

After obtaining written informed consent from all subjects, the data were collected using a case report form (CRF), including general subject characteristics, such as the diagnosis, medical history, and score on the Korean standard criteria for stroke (K-SPI-Stoke), which was developed by an expert committee organized by the KIOM [3, 4]. The PI diagnosis of each patient was made by two expert TKM doctors, and subjects who received different diagnoses from the two doctors were excluded. Subjects for whom BMI, waist circumference, or other physical data were missing were also excluded. A total of 630 CI patients were classified as having a Yin Deficiency pattern (N = 75) or Non-Yin Deficiency pattern (N = 555). The general characteristics of Non-Yin Deficiency pattern patients are shown in Supplemental Table 1 (see Supplementary Material available online at doi:10.1155/2012/798131). This study was approved by the Institutional Review Boards of the KIOM and the Oriental Medical Hospitals.

2.2. Preparation of Genomic DNA and Identification of SNP. Genomic DNA from each subject was extracted from whole blood using a GeneAll Genomic Isolation Kit (GeneAll, Seoul, Korea). The WNT10B G-607C polymorphism of each subject was genotyped using the TaQman method with polymerase chain reaction (PCR) primers and TaQman probes purchased from ABI Inc. (Applied Biosystems Inc., USA) according to the manufacturer’s protocol. Additionally, the genotypes of five percent of the participants were redetermined by direct sequencing to verify the accuracy of the TaQman method (Figure 1). The primers used for the amplification and genotyping of G-607C variation are described in Supplemental Table 2. The genotyping error rate of the TaQman method in this study was 0.3% based on the direct sequencing of PCR products (data not shown), and the Kappa value was 0.98, demonstrating good accuracy. Hardy-Weinberg equilibrium (HWE) was evaluated by chi-square test using Haploview 4.2 (http://www.genenames.org/).

2.3. Statistical Analysis. Data were statistically analyzed with SAS software, version 9.1.3 (SAS Institute Inc., Cary, NC). All continuous variables were subjected to a Kolmogorov-Smirnov normality test. Differences in continuous variables were determined using parametric (Student’s t-test) or nonparametric (Wilcoxon rank-sum test) tests. Categorical variables were compared with a chi-square test or Fisher’s exact test.

The association of the SNP with the Yin Deficiency pattern versus Non-Yin Deficiency pattern was performed by multiple logistic regression adjusted for age, body mass index (BMI), waist-hip ratio (WHR), and triglyceride levels, and odds ratios (ORs) with 95% confidence intervals (95% CI) were calculated. To investigate whether the G-607C polymorphism is associated with the clinical parameters of the study subjects, we performed a statistical analysis using a general linear model adjusted for sex, age, smoking, and drinking status. Statistical significance was set at P < 0.05.
The general characteristics of CI patients classified as Yin Deficiency pattern or Non-Yin Deficiency pattern according to the PI of TKM are shown in Table 1. The mean age of the Yin Deficiency pattern patients was significantly higher than that of the Non-Yin Deficiency pattern patients ($P = 0.0002$). Additionally, triglyceride levels were significantly lower in the Yin Deficiency pattern group than in the Non-Yin Deficiency pattern group ($P = 0.0009$).

The clinical differences in the body characteristics between Yin Deficiency and Non-Yin Deficiency patterns are shown in Table 2. The mean weight, BMI and waist circumference of the Yin Deficiency pattern patients, was significantly lower than that of the Non-Yin Deficiency pattern subjects after adjustment for age and triglyceride levels ($P = 0.0002$, $P = 0.0020$ and $P < 0.0001$, resp.).

The G-607C SNP in WNT10B was in HWE ($P = 0.0339$, OR $= 0.999$) according to the International HapMap Project guidelines, and the minor allele frequency (MAF) in this study was 0.495, which is slightly higher than that observed in previous studies among Korean females and Caucasians in USA [24, 25].

Table 3 shows the G-607C SNP distribution in the Non-Yin Deficiency and Yin Deficiency patients. The ratio of subjects with the CC genotype in the Yin Deficiency pattern group (29.33%) is smaller than that in the Non-Yin Deficiency pattern group (23.96%), after adjustment for age, BMI, WHR, and triglyceride levels ($P = 0.0339$, OR = 2.005 (1.054–3.814)). The frequency of the C allele in Yin Deficiency pattern patients is 50.0%, slightly higher than that in the Non-Yin Deficiency pattern patients (49.46%), although this difference was not significant. Additionally, the frequency of the G allele in the Yin Deficiency pattern group (70.67%) is smaller than that in the Non-Yin Deficiency pattern group (74.95%), but this difference was not statistically significant.

### 3. Result

The general characteristics of CI patients classified as Yin Deficiency pattern or Non-Yin Deficiency pattern according to the PI of TKM are shown in Table 1. The mean age of the Yin Deficiency pattern patients was significantly higher than that of the Non-Yin Deficiency pattern patients ($P = 0.0002$). Additionally, triglyceride levels were significantly lower in the Yin Deficiency pattern group than in the Non-Yin Deficiency pattern group ($P = 0.0009$).

The clinical differences in the body characteristics between Yin Deficiency and Non-Yin Deficiency patterns are shown in Table 2. The mean weight, BMI and waist circumference of the Yin Deficiency pattern patients, was significantly lower than that of the Non-Yin Deficiency pattern subjects after adjustment for age and triglyceride levels ($P = 0.0002$, $P = 0.0020$ and $P < 0.0001$, resp.).

The G-607C SNP in WNT10B was in HWE ($P = 0.0339$, OR $= 0.999$) according to the International HapMap Project guidelines, and the minor allele frequency (MAF) in this study was 0.495, which is slightly higher than that observed in previous studies among Korean females and Caucasians in USA [24, 25].

Table 3 shows the G-607C SNP distribution in the Non-Yin Deficiency and Yin Deficiency patients. The ratio of subjects with the CC genotype in the Yin Deficiency pattern group (29.33%) is smaller than that in the Non-Yin Deficiency pattern group (23.96%), after adjustment for age, BMI, WHR, and triglyceride levels ($P = 0.0339$, OR = 2.005 (1.054–3.814)). The frequency of the C allele in Yin Deficiency pattern patients is 50.0%, slightly higher than that in the Non-Yin Deficiency pattern patients (49.46%), although this difference was not significant. Additionally, the frequency of the G allele in the Yin Deficiency pattern group (70.67%) is smaller than that in the Non-Yin Deficiency pattern group (74.95%), but this difference was not statistically significant.
genetic factors have been correlated to PI in certain diseases by regression analysis. The Wnt/β-catenin signaling pathway has been involved in many physiological pathways by regulating protein expression of WNT10B and was significantly associated with Yin Deficiency pattern in a Chinese Han population [14]. However, no genetic factors associated with the Yin Deficiency pattern have yet been established in a Korean population.

### 4. Discussion

PI is a traditional diagnosis system developed over several hundred years in East Asia [1, 2], but there remains a lack of scientific evidence supporting its use due to its high dependence on subjective diagnostic indicators. Recently, some clinical differences among patterns were reported, and genetic factors have been correlated to PI in certain diseases [10–14].

WNTs are the ligands of Frizzled receptors and are involved in many physiological pathways by regulating the Wnt/β-catenin signaling pathway [26]. Among WNT family genes, WNT10B is expressed in carcinoma cells [18–22] and adipocytes [27, 28] and is known to inhibit adipogenesis. Ross et al. showed that the overexpression of WNT10B in 3T3L1 preadipocytes inhibited adipogenesis by inhibiting C/EBPα and PPARγ2, transcription factors that accelerate adipogenesis [27], and Bennett et al. also reported that WNT10B inhibits adipogenesis by inhibiting glycogen synthase kinase 3 activity [28]. Another study also showed that transgenic mice with WNT10B driven by the FABP4 promoter have 50–70% less adipose tissue weight than ob/ob mice [29]. Recently, G-607C SNP, which is located in a promoter region of WNT10B, was found to alter the expression of WNT10B and was significantly associated with a decrease in abdominal fat area in Korean female subjects [24].

Table 4 categorizes stroke patients according to their related internal disease symptoms. Previously, Lee et al. described PI for four patterns in stroke patients (Fire-Heat pattern, Dampness-Phlegm pattern, Yin Deficiency pattern, and Qi Deficiency pattern) as part of the “SOPI-Stroke” project of the KIOM [3, 4]. The Yin Deficiency pattern in stroke patients results from a deficiency of yin with diminished moistening and inability to restrain yang, which is usually manifested as emaciation, dizziness, tinnitus, dryness of the mouth and throat, constipation, dark urine, afternoon fever, malar flush, night sweats, reddened tongue with scanty coating, and rapid fine pulse [1–4]. Moreover, patients with Yin Deficiency pattern are nonobese and gaunt [1, 5, 8]. Wu et al. reported that Pro12Ala SNP of PPARγ polymorphism were associated with Yin Deficiency pattern in a Chinese Han population [14]. However, no genetic factors associated with the Yin Deficiency pattern have yet been established in a Korean population.

### Table 3: Genotype distribution of the G-607C polymorphism in patients with Yin Deficiency pattern versus non-Yin Deficiency pattern.

| Model | Genotype | Non-YD | YD | OR [95% CI] | P |
|-------|----------|--------|----|------------|---|
| Allele | G        | 561 (50.54) | 75 (50.0) | 1.262 [0.818, 1.946] | 0.2927 |
|        | C        | 549 (49.46) | 75 (50.0) | 1.262 [0.818, 1.946] | 0.2927 |
| §Do    | GG       | 139 (25.05) | 22 (29.33) | 0.850 [0.422, 1.712] | 0.6495 |
|        | GC + CC  | 416 (74.95) | 53 (70.67) | 2.005 [1.054, 3.814] | 0.0339 |
| §R     | GG + GC  | 422 (76.04) | 53 (70.67) | 2.005 [1.054, 3.814] | 0.0339 |
|        | CC       | 133 (23.96) | 22 (29.33) | 0.850 [0.422, 1.712] | 0.6495 |

The data are presented as frequencies (percentages). §ORs after adjustment for age, BMI, WHR, and triglyceride levels. P values were calculated by logistic regression analysis. §Do and R denote dominant and recessive models, respectively. P values with statistical significance are shown in bold (<0.05).

### Table 4: Association of G-607C polymorphism with clinical parameters among study subjects.

| Variable             | GG (n = 161) | Genotype | CC (n = 155) | §Co | P |
|----------------------|--------------|----------|--------------|-----|---|
| Body characteristics |              |          |              |     |   |
| Weight (kg)          | 61.44 ± 11.91| GC (n = 314) | 60.89 ± 10.50 | 0.2681 | 0.7136 | 0.1711 |
| BMI (kg/m²)          | 24.23 ± 3.44 | CC (n = 155) | 24.01 ± 3.17 | 0.7647 | 0.8895 | 0.4686 |
| Waist circumference (cm) | 87.86 ± 9.91 | GC + CC | 87.13 ± 8.52 | 0.5062 | 0.9268 | 0.2835 |
| WHR                  | 0.95 ± 0.10  | GC (n = 314) | 0.95 ± 0.18  | 0.9547 | 0.9179 | 0.7606 |
| Serum parameters     |              |          |              |     |   |
| GOP (U/mL)           | 24.89 ± 10.78| GC (n = 314) | 27.70 ± 15.66 | 0.1623 | 0.0602 | 0.3483 |
| GPT (U/mL)           | 23.73 ± 16.78| CC (n = 155) | 24.92 ± 18.10 | 0.7313 | 0.4291 | 0.8164 |
| Total cholesterol (mg/dL) | 191.67 ± 52.29 | GC + CC | 181.55 ± 43.91 | 0.1521 | 0.2479 | 0.0634 |
| Triglyceride (mg/dL) | 171.75 ± 150.35 | GC (n = 314) | 151.08 ± 96.69 | 0.3631 | 0.2243 | 0.2685 |
| HDL-cholesterol (mg/dL) | 43.79 ± 11.15 | CC (n = 155) | 42.41 ± 11.40 | 0.5582 | 0.5173 | 0.3029 |
| FBS (mg/dL)          | 108.86 ± 29.41| CC (n = 155) | 112.46 ± 36.74 | 0.1569 | 0.1072 | 0.6417 |

The data are presented as the mean ± standard deviation. P values: adjusted for sex, age, smoking, and drinking status using a general linear model. §Co, Do and R denote codominant, dominant, and recessive models, respectively.

Table 4 shows the comparison of obesity phenotypes and serum parameters according to G-607C genotype. The mean level of total cholesterol tended to decrease in subjects with the CC genotype compared with the GG or GC genotype in the recessive model (P = 0.0634).
In this study, we analyzed the association of the G-607C SNP of WNT10B with a Yin Deficiency pattern in elderly Korean subjects with CI.

The Yin Deficiency pattern was related with decreased obesity. In this study population, the BMI and waist circumference in the Yin Deficiency group were significantly lower than that in the Non-Yin Deficiency group, and WHR was also slightly lower in subjects with Yin Deficiency (Table 2). This result was similar to those of other studies [7, 8]. Zhu et al. showed that frequency of overweight in subjects with Yin Deficiency was slightly lower than that in the overall study population [7], and Yin Deficiency patients with type 2 diabetes mellitus were usually non-obese [8].

In this study, the frequency of subjects with the CC genotype in the G-607C SNP of WNT10B was 29.33% in the Yin Deficiency group, significantly higher than that in the Non-Yin Deficiency group (23.96%) (OR = 2.005) in the recessive model (Table 3). We also confirmed that the G-607C SNP was associated with a trend toward decreased serum lipids and total cholesterol (Table 4). These data showed that correlation between the G-607C SNP and Yin Deficiency in elderly Korean subjects with CI might be related to a decrease in obesity indices. This result suggests that PI may be affected not only by environmental factors but also by inherent factors such as genetic variations.

This study showed, for the first time, that WNT10B polymorphism is associated with Yin Deficiency pattern in a CI patient population. Thus, this SNP can be used as a diagnostic genetic marker for Yin Deficiency pattern in stroke patients and in the development of personalized medical care. However, this study included several limitations. First, this is a simple cross-sectional study, not a longitudinal study. Second, this study does not have a sufficient sample size to generalize the relationship between WNT10B and Yin Deficiency. Further studies should be performed in subjects of other ethnicities to generalize the conclusions of this study.

Acknowledgment

This research was supported by a Grant from the Korea Inst. of Oriental Medicine (K12130).

References

[1] WHO International Standard Terminologies on Traditional Medicine in the Western Pacific Region, WHO Western Pacific Regional Office, 2007.

[2] H. J. Kim, H. S. Bae, S. U. Park, S. K. Moon, J. M. Park, and W. S. Jung, “Clinical approach to the standardization of oriental medical diagnostic pattern identification in stroke patients,” Evidence-based Complementary and Alternative Medicine, vol. 2011, Article ID 768492, 7 pages, 2011.

[3] J. A. Lee, I. S. Lee, B. K. Kang et al., “Report on the Korean standard pattern identification for the stroke-III,” Korean Journal of Oriental Internal Medicine, vol. 32, no. 2, pp. 232–242, 2011.

[4] T. Y. Park, J. A. Lee, M. H. Cha et al., “The fundamental study of the standardization and objectification of pattern identification in traditional Korean medicine for stroke (SOPH-Stroke): an overview of phase 1,” European Journal of the Integrative Medicine, vol. 4, no. 2, pp. 125–131, 2012.

[5] B. K. Lee, Diagnosis in Oriental Medicine, Seongbosa, Seoul, South Korea, 3rd edition, 1992.

[6] The pathology association of oriental Medicine colleges, Pathology of Oriental Medicine, Iljoonsa, Seoul, South Korea, 4th edition, 2004.

[7] Y. B. Zhu, Q. Wang, C. Y. Wu et al., “Logistic regression analysis on relationships between traditional Chinese medicine constitutional types and overweight or obesity,” Journal of Chinese Integrative Medicine, vol. 8, no. 11, pp. 1023–1028, 2010.

[8] C. L. Qiu, C. Y. Tian, and J. A. Li, “Study progress of TCM syndrome of type 2 diabetes mellitus,” China Journal of Traditional Chinese Medicine and Pharmacy, vol. 25, no. 4, pp. 517–519, 2010.

[9] L. Z. Chun, Basic Theory of Traditional Chinese Medicine, Higher Education Press, 2007.

[10] S. An, E. Li, and X. Tong, “Study on relationship between estrogen receptor gene polymorphism and syndrome differentiation typing of female postmenopausal osteoporosis in Traditional Chinese medicine,” Chinese Journal of Integrated Traditional and Western Medicine, vol. 20, no. 12, pp. 907–910, 2000.

[11] S. Chen, F. Lv, J. Gao et al., “HLA class II polymorphisms associated with the physiologic characteristics defined by traditional Chinese medicine: linking modern genetics with an ancient medicine,” Journal of Alternative and Complementary Medicine, vol. 13, no. 2, pp. 231–239, 2007.

[12] J. H. Lim, M. M. Ko, J. S. Lee, O. S. Bang, and M. H. Cha, “Genetic association of SNPs located at PON1 gene with dampness and phlegm pattern identification among Korea stroke patients,” Korean Journal of Oriental Internal Medicine, vol. 31, no. 4, pp. 752–762, 2010.

[13] M. M. Ko, B. K. Kang, J. H. Lim, M. S. Lee, and M. H. Cha, “Genetic association of NPY gene polymorphisms with Dampness-Phlegm pattern in Korea stroke patients,” Evidence-Based Complementary and Alternative Medicine, vol. 2012, Article ID 109796, 7 pages, 2012.

[14] Y. Wu, Y. Cun, J. Dong et al., “Polymorphisms in PPARD, PPARG and APOA1 associated with four types of Traditional Chinese Medicine constitutions,” Journal of Genetics and Genomics, vol. 37, no. 6, pp. 371–379, 2010.

[15] K. Willert, J. D. Brown, E. Danenberg et al., “Wnt proteins are lipid-modified and can act as stem cell growth factors,” Nature, vol. 423, no. 6938, pp. 448–452, 2003.

[16] T. D. Bui, J. Rankin, K. Smith et al., “A novel human Wnt gene, WNT10B, maps to 12q13 and is expressed in human breast carcinomas,” Oncogene, vol. 14, no. 10, pp. 1249–1253, 1997.

[17] P. Wend, K. Wend, S. A. Krum, and G. A. Miranda-Carboni, “The role of WNT10B in physiology and disease,” Acta Physiologica, vol. 204, no. 1, pp. 34–51, 2011.

[18] H. Kirikoshi and M. Katoh, “Expression and regulation of WNT10B in human cancer: up-regulation of WNT10B in MCF-7 cells by beta-estradiol and down-regulation of WNT10B in NT2 cells by retinoic acid,” International Journal of Molecular Medicine, vol. 10, no. 4, pp. 507–511, 2002.

[19] H. Kirikoshi and M. Katoh, “Expression of WNT7A in human normal tissues and cancer, and regulation of WNT7A and WNT7B in human cancer,” International Journal of Oncology, vol. 21, no. 4, pp. 895–900, 2002.
[20] H. Kirikoshi, S. Inoue, H. Sekihara, and M. Katoh, “Expression of WNT10A in human cancer,” *International Journal of Oncology*, vol. 19, no. 5, pp. 997–1001, 2001.

[21] H. Kirikoshi, H. Sekihara, and M. Katoh, “Expression of WNT14 and WNT14B mRNAs in human cancer, up-regulation of WNT14 by IFN gamma and up-regulation of WNT14B by beta-estradiol,” *International Journal of Oncology*, vol. 19, no. 6, pp. 1221–1225, 2001.

[22] H. Kirikoshi, H. Sekihara, and M. Katoh, “Up-regulation of WNT10A by tumor necrosis factor alpha and helicobacter pylori in gastric cancer,” *International Journal of Oncology*, vol. 19, no. 3, pp. 533–536, 2001.

[23] C. Christodoulides, A. Scarda, M. Granzotto et al., “WNT10B mutations in human obesity,” *Diabetologia*, vol. 49, no. 4, pp. 678–684, 2006.

[24] I. C. Kim, M. H. Cha, D. M. Kim et al., “A functional promoter polymorphism –607G>C of WNT10B is associated with abdominal fat in Korean female subjects,” *Journal of Nutritional Biochemistry*, vol. 22, no. 3, pp. 252–258, 2011.

[25] J. M. Zmuda, L. M. Yerges, C. M. Kammerer et al., “Association analysis of WNT10B with bone mass and structure among individuals of African ancestry,” *Journal of Bone and Mineral Research*, vol. 24, no. 3, pp. 437–447, 2009.

[26] http://www.stanford.edu/group/nusselab/cgi-bin/wnt/pathway_diagram.

[27] S. E. Ross, N. Hemati, K. A. Longo et al., “Inhibition of adipogenesis by Wnt signaling,” *Science*, vol. 289, no. 5481, pp. 950–953, 2000.

[28] C. N. Bennett, S. E. Ross, K. A. Longo et al., “Regulation of Wnt signaling during adipogenesis,” *Journal of Biological Chemistry*, vol. 277, no. 34, pp. 30998–31004, 2002.

[29] W. S. Wright, K. A. Longo, V. W. Dolinsky et al., “Wnt10b inhibits obesity in ob/ob and agouti mice,” *Diabetes*, vol. 56, no. 2, pp. 295–303, 2007.