Relationships between MGMT promoter methylation and gastric cancer: a meta-analysis

Dan Yu
Tao Cao
Ya-Di Han
Fu-Sheng Huang

Department of Laboratory Medicine, Center for Gene Diagnosis, Zhongnan Hospital of Wuhan University, Wuhan, People’s Republic of China

Abstract: A DNA repair enzyme, O6-methylguanine-DNA methyltransferase (MGMT), plays an important role in the development of gastric cancers. However, the role of MGMT promoter methylation in the occurrence of gastric cancer and its relationships with clinicopathologic characteristics has not been fully clarified. Thus, we performed a meta-analysis to evaluate the associations between MGMT promoter methylation and gastric cancer. Electronic databases, including PubMed and Web of Science, were used to systematically search related clinical studies published in English until April 1, 2016. Odds ratios (ORs) and 95% confidence intervals (95% CIs) were calculated to evaluate the associations between MGMT promoter methylation and gastric cancer risk or clinicopathologic characteristics. A total of 16 studies including 1,935 patients and 1,948 control persons were included in the analysis. Our study suggested that MGMT promoter methylation frequency was associated with gastric cancer (OR=3.46, 95% CI: 2.13–5.61, P<0.001). Moreover, the frequency of MGMT promoter methylation in the no lymph node metastasis group was lower than that in lymph node metastasis group, with marginal significance (OR=0.65, 95% CI: 0.42–1.01, P=0.05). Additionally, the methylation rate of the MGMT promoter was much lower in patients without distant metastases than in those with metastases (OR=0.27, 95% CI: 0.18–0.40, P<0.001). No significant association of MGMT promoter methylation with Lauren classification, tumor location, tumor invasion, or Helicobacter pylori infection was found. In conclusion, the methylation status of the MGMT promoter was related to gastric cancer risk, distant metastasis, and lymph node metastasis, which indicates that MGMT promoter methylation may play an important role in gastric cancer development.

Keywords: gastric cancer, tumor suppressor gene, cancer risks

Introduction

Gastric cancer is one of the most common cancers worldwide, with an estimated 951,600 new stomach cancer cases and 723,100 deaths in 2012.1 Although diagnostic methods, surgical techniques, and targeted therapy have improved, gastric cancer remains a notable clinical challenge.2 Many studies indicate that epigenetic alterations in tumor suppressor genes, such as cadherin 13,3 Ras association domain family member 1,4 methylation of O6-methylguanine-DNA methyltransferase (MGMT),5 and adenomatosis polyposis coli,6 play an important role in the initiation and progression of human cancer. DNA methylation is one of the most significant processes involved in epigenetic modifications and has an important effect on the development and prognosis of human cancer.7–10 In gastric cancer, hypermethylation of tumor suppressor genes has been frequently found.11

Among these tumor suppressor genes, MGMT in gastric cancer has often been investigated. The MGMT gene, located at chromosome 10q26, includes one noncoding and four...
coding exons.\textsuperscript{12} \textit{MGMT}, a DNA repair enzyme, mainly defends cells against the carcinogenic effects of adducts by eliminating alkyl groups from the O6-position of guanine and then transferring them into its active center.\textsuperscript{13} O6-methylguanine (O6-mG) is the most potent mutagenic lesion that leads to a G-C to A-T transition mutation. \textit{MGMT} can restore this mutagenesis of endogenous DNA damage and play an important role in maintaining normal cell physiology and genomic stability.\textsuperscript{14} Thus, loss of \textit{MGMT} function can cause mutations, leading to human carcinogenesis.\textsuperscript{15} \textit{MGMT} promoter methylation resulting in gene silencing and loss of function was found in many tumors, including colorectal cancer,\textsuperscript{16} non-small-cell lung cancer,\textsuperscript{17} gliomas,\textsuperscript{18} and gastric cancer.\textsuperscript{19} Oue et al\textsuperscript{20} first found that \textit{MGMT} promoter methylation may play a role in carcinogenesis in the stomach. Subsequently, many studies have demonstrated that \textit{MGMT} methylation has been observed more frequently in gastric cancer tissues than in noncancer tissues,\textsuperscript{21–23} suggesting that \textit{MGMT} methylation may be associated with an increased gastric cancer risk. However, contradictory results also existed. Therefore, we performed a meta-analysis to elucidate the associations between \textit{MGMT} promoter methylation and gastric cancer.

Methods

Search strategy

Electronic databases, including Web of Science, and PubMed, were used to systematically search related clinical studies published in English until April 1, 2016. The following terms were used: (methylation or DNA methylation or hypermethylation or demethylation), (gastric cancer or gastric carcinoma or gastric tumor), and (O-6-methylguanine-DNA methyltransferase or \textit{MGMT}).

Inclusion and exclusion criteria

Eligible studies met the following standards: 1) assessed the association between \textit{MGMT} methylation and gastric cancer; 2) case-control or cohort studies; 3) studies with sufficient data for calculating odds ratios (ORs) and their 95% confidence intervals (CIs); 4) at least three case and control groups; 5) patients had a definite diagnosis of gastric cancer by pathological or histological examination. For duplicated data, only the most recent or comprehensive studies were included. Moreover, reviews, meta-analysis, case reports, letters, and animal and cell studies were excluded.

Data extraction

Data from the included studies were extracted independently by two authors. The following data were recorded from each study: First author’s last name, year of publication, ethnicity, the frequency of \textit{MGMT} methylation in case and control groups, detection method, sample type, source of samples, the number of patients with distant metastasis status having a methylated and unmethylated status, lymph node status, sex, Lauren classification and \textit{Helicobacter pylori} infection. Any differences of opinion were discussed till an agreement was reached.

Statistical analysis

Stata 12.0 (Stata Corporation, TX, USA) and Review Manager 5.2 (Cochrane Collaboration, Oxford, UK) were used in this meta-analysis. The ORs and 95% CIs were used to evaluate the association between \textit{MGMT} promoter methylation and gastric cancer risk or clinicopathologic features. Heterogeneity between studies was evaluated by the Q-test based on the \textit{χ}\textsuperscript{2} statistic and \textit{I}\textsuperscript{2} statistics.\textsuperscript{24} If substantial heterogeneity existed (\textit{P}<0.05 for the Q statistic or \textit{I}\textsuperscript{2}>50%), a random effect model was applied to pool the ORs; otherwise, a fixed effect model was conducted.\textsuperscript{25} A meta-regression analysis was conducted to explore reasons for statistical heterogeneity. Additionally, subgroup analysis was performed based on sex, ethnicity, and sample size to determine the source of heterogeneity. A sensitivity analysis was conducted to assess the effect of single studies on the overall estimate by omitting one study at a time. A funnel plot, trim and fill method, and Egger’s test were used to assess for publication bias. All tests were two-sided, and \textit{P}<0.05 denoted statistical significance.

Results

Study selection and characteristics

A total of 72 relevant articles were identified from electronic databases. After reading the titles, abstracts, and full text, 56 studies were excluded, because of irrelevant content, duplicated articles, non-English articles, inadequate data, and cell lines research. Finally, a total of 16 studies,\textsuperscript{21–23,26–38} consisting of 1,935 cases and 1,948 controls, were included in the analysis. The study selection process is shown in Figure 1. The methylation rate ranged from 6.9% to 70% in the cancer group and 0% to 44.9% in the control group. Among these studies, 4 studies were conducted on Caucasian, 11 studies on Asian, and 1 study on African individuals. Fourteen studies explored \textit{MGMT} promoter methylation in tissues and two studies explored \textit{MGMT} promoter methylation in blood. The basic characteristics of the included studies are shown in Table 1.

Meta-analysis

\textit{MGMT} promoter methylation and gastric cancer risk

Our results revealed that the frequency of \textit{MGMT} promoter methylation was increased in patients with gastric cancer.
Our results showed that the frequency of MGMT promoter methylation in gastric cancer patients was lower than that in the lymph node metastasis group (OR=0.65, 95% CI: 0.42–1.01, P=0.05, Figure 3) under a random effect model. Additionally, the rate of MGMT promoter methylation was much lower in patients without distant metastases than in patients with metastases (OR=0.27, 95% CI: 0.18–0.40, P<0.001, Figure 4) with a fixed effect model. Results revealed no significant association of MGMT promoter methylation with Lauren classification (OR=0.95, 95% CI: 0.62–1.47, P=0.82), tumor invasion (OR=0.79, 95% CI: 0.60–1.04, P=0.09), tumor location (OR=0.90, 95% CI: 0.68–1.20, P=0.049), or H. pylori infection (OR=1.02, 95% CI: 0.54–1.93, P=0.94) with a fixed effect model. The detailed results are shown in Table 3.

**Publication bias**

The shapes of funnel plots, trim and fill method, and Egger’s linear regression test were used to evaluate the publication bias. Slight asymmetry was observed in funnel plots, indicating that publication bias existed in evaluating the association of MGMT promoter methylation with gastric cancer risk (Figure 5); however, the P-value of Egger’s test was greater at 0.076. Trim and fill analysis was performed, and the pooled OR was 2.05 (95% CI: 1.26–3.33, P<0.001). The results were similar to the crude meta-analysis, suggesting that our analyses were reliable.

**Discussion**

Gastric cancer is still a major clinical challenge with poor prognosis in recent years. A useful detection biomarker for the early diagnosis and prognosis evaluation is needed. Silencing tumor suppressor genes expression by aberrant methylation of the promoter regions has been found in the process of tumors. Thus, by pooling the data from 16 studies, we investigated the associations of MGMT promoter methylation with gastric cancer risk and its clinicopathologic features. According to our meta-analysis, MGMT promoter methylation was significantly correlated with the gastric cancer risk. Our results also suggested that the frequency of MGMT promoter methylation was lower in the lymph node metastasis group than in the lymph node metastasis group, with marginal significance. More importantly, we found that distant metastasis was associated with increased MGMT promoter hypermethylation.

The current meta-analysis revealed an association between MGMT promoter methylation and gastric cancer risk. This was in line with previous studies in which the frequency of MGMT promoter methylation in tumors was increased compared with control groups, although a
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Jin et al(^21) (2014)</td>
<td>People's Republic of China</td>
<td>90/193</td>
<td>49/234</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>Male</td>
<td>Female</td>
<td>Intestinal</td>
<td>Diffuse</td>
<td>T1-T2</td>
<td>T3-T4</td>
</tr>
<tr>
<td>Xiong et al(^27) (2013)</td>
<td>People's Republic of China</td>
<td>114/299</td>
<td>48/365</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>63/159</td>
<td>51/140</td>
<td>NA</td>
<td>NA</td>
<td>35/143</td>
<td>54/181</td>
</tr>
<tr>
<td>Hiraki et al(^28) (2010)</td>
<td>Japan</td>
<td>26/23</td>
<td>15/34</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>14/16</td>
<td>12/7</td>
<td>13/14</td>
<td>13/9</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Oue et al(^32) (2006)</td>
<td>Japan</td>
<td>20/55</td>
<td>2/33</td>
<td>MSP</td>
<td>Tissue</td>
<td>MIX</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Schilhaus et al(^33) (2005)</td>
<td>Germany</td>
<td>2/5</td>
<td>0/7</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>1/2</td>
<td>0</td>
</tr>
<tr>
<td>Hong et al(^34) (2005)</td>
<td>Korea</td>
<td>25/75</td>
<td>4/234</td>
<td>MSP</td>
<td>Blood</td>
<td>H</td>
<td>H</td>
<td>7/57</td>
<td>18/18</td>
<td>8/22</td>
<td>13/32</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Sabbioni et al(^35) (2003)</td>
<td>Italy</td>
<td>11/10</td>
<td>0/6</td>
<td>MSP</td>
<td>Tissue</td>
<td>H</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Kang et al(^36) (2003)</td>
<td>Korea</td>
<td>11/63</td>
<td>17/63</td>
<td>MSP</td>
<td>Tissue</td>
<td>NT</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Bae et al(^37) (2002)</td>
<td>Korea</td>
<td>21/128</td>
<td>0/149</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>Oue et al(^38) (2001)</td>
<td>Japan</td>
<td>8/42</td>
<td>0/50</td>
<td>MSP</td>
<td>Tissue</td>
<td>AT</td>
<td>CL</td>
<td>NA</td>
<td>NA</td>
<td>7/17</td>
<td>1/25</td>
<td>NA</td>
<td>NA</td>
</tr>
</tbody>
</table>

**Abbreviations:** APETT, archival paraffin embedded tumor tissues; AT, normal gastric adjacent the tumor; CL, cancer lesions; H, healthy controls; H. pylori, Helicobacter pylori; M, methylations; MIX, mixed controls; MSP, methylation-specific; PCR, polymerase chain reaction; NA, not available; NT, normal gastric tissue; U, unmethylations.
study by Kupčinskaitė-Noreikienė et al\textsuperscript{26} revealed no significant association between MGMT promoter methylation and gastric cancer. MGMT promoter methylation could be considered as a risk factor for the development of gastric cancer after large-scale studies are conducted. Silencing MGMT expression by hypermethylation has two consequences for cancer.\textsuperscript{44} First, MGMT can defend cell against alkylating-induced gene mutations, toxicity, and carcinogenicity.\textsuperscript{41} Alkylating agent can result in alkylation of O6 guanine in DNA. O6-methylguanine preferentially pairs with thymine in DNA replication leading to a G-C to A-T transition mutation\textsuperscript{42} and can lead to cross-linking reactions with cytosine on the side chain, resulting in termination of DNA synthesis. These changes may result in K-ras mutation or p53 mutation\textsuperscript{43} leading to the development of cancer.

Unrepaired DNA damage is a major source of potentially mutagenic lesions that may lead to cancer.\textsuperscript{41} Second, MGMT hypermethylation in tumors is sensitive to alkylating drugs used in chemotherapy. MGMT promoter methylation has been associated with chemo-responsiveness with alkylating agent drugs in glioma, Hodgkin’s lymphoma cells and other tumor models.\textsuperscript{44,45} Silencing MGMT gene expression by promoter methylation can increase the sensitivity to temozolomide, which is considered a therapeutic option for some gastric cancers. Additionally, Miura et al indicated that negative expression of the MGMT gene was observed in 45% of the gastroesophageal tumors on the basis of biomarker profiling, suggesting potential sensitivity to temozolomide.\textsuperscript{46}

Therefore, it is necessary to detect the MGMT promoter methylation status in patients with gastric cancer to develop individualized treatment programs when they are treated with temozolomide.

In the subgroup analysis based on ethnicity, a significant association between MGMT promoter methylation and gastric cancer risk was observed in Asians, but no significant difference was found in Caucasians. It suggested that a combination of differences in gene backgrounds and the environment may have certain impact on the prevalence of MGMT methylation. Furthermore, limited studies were conducted on Caucasians, which may bring about a false-negative result. Further study is warranted to investigate the association between MGMT promoter methylation and gastric cancer in Caucasians. In the subgroup analysis of sample size, statistical associations were found for all

**Table 2** Stratified analysis of the frequency of MGMT promoter methylation in gastric cancers compared with noncancer controls

<table>
<thead>
<tr>
<th>Study group</th>
<th>N</th>
<th>OR (95% CI)</th>
<th>χ² (%)</th>
<th>Pexpo</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total</td>
<td>16</td>
<td>3.46 (2.13, 5.61)</td>
<td>75</td>
<td>&lt;0.001</td>
<td>0.076</td>
</tr>
<tr>
<td>Ethnicity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caucasians</td>
<td>4</td>
<td>2.70 (0.61, 12.06)</td>
<td>69</td>
<td>0.02</td>
<td>0.126</td>
</tr>
<tr>
<td>Asians</td>
<td>11</td>
<td>3.29 (1.99, 5.44)</td>
<td>73</td>
<td>&lt;0.001</td>
<td>0.063</td>
</tr>
<tr>
<td>Case sample</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>size &lt;60</td>
<td>5</td>
<td>4.14 (2.22, 7.73)</td>
<td>0</td>
<td>0.52</td>
<td>0.021</td>
</tr>
<tr>
<td>size &gt;60</td>
<td>11</td>
<td>3.25 (1.83, 5.77)</td>
<td>82</td>
<td>&lt;0.001</td>
<td>0.120</td>
</tr>
</tbody>
</table>

Note: Values in bold indicate statistical significance. Abbreviations: CI, confidence interval; MGMT, O6-methylguanine-DNA methyltransferase; N, total number of eligible studies; NA, not available; OR, odds ratio; Pexp, P-value of Egger linear regression test for evaluating publication bias; Pvalue of Q test for heterogeneity among studies.
subgroups. Additionally, the results of subgroup analysis based on sex revealed that MGMT promoter methylation has no relationship with sex in patients with gastric cancer patients.

The associations between MGMT promoter methylation and Lauren classification, H. pylori infection, tumor location, distant metastasis, and lymph node status were also investigated. Our results showed that MGMT promoter methylation was related to lymph node metastasis and distant metastasis was observed in our study, indicating that MGMT promoter methylation may be involved in the metastasis of gastric cancer. The tumor microenvironment plays an important role in tumor progression and may exert an influence on the epigenetic status of micrometastatic colonies in the lymph nodes. Therefore, the frequency of methylation differs depending on whether lymph node metastasis has occurred. Therefore, it was hypothesized that MGMT promoter methylation may serve as a biomarker for monitoring gastric cancer metastasis. Furthermore, Li et al. investigated the role of MGMT in gastric cancer cell migration, invasion, and metastatic potential. They demonstrated that loss of MGMT expression induced increases in gastric cancer cell metastasis.

![Forest plot for evaluating the association between MGMT promoter methylation and lymph node metastasis.](image1)

**Figure 3** Forest plot for evaluating the association between MGMT promoter methylation and lymph node metastasis. Random-effect model was used for the analysis. **Notes:** The pooled OR from 9 studies included 692 gastric cancer patients with no lymph node metastasis and 638 patients with lymph node metastasis (OR=0.65, 95% CI: 0.42–0.47, P=0.05). Negative: patients with no lymph node metastasis, positive: patients with lymph node metastasis. **Abbreviations:** MGMT, O6-methylguanine-DNA methyltransferase; CI, confidence interval; M–H, Mantel–Haenszel; OR, odds ratio.

![Forest plot for evaluating the association between MGMT promoter methylation and distant metastasis.](image2)

**Figure 4** Forest plot for evaluating the association between MGMT promoter methylation and distant metastasis. Fixed-effect model was used for the analysis. **Notes:** The pooled OR from 6 studies included 1,154 gastric cancer patients with no distant metastasis and 107 patients with distant metastasis (OR=0.27, 95% CI: 0.18–0.40, P=0.0001). Negative: patients with no distant metastasis, positive: patients with distant metastasis. **Abbreviations:** MGMT, O6-methylguanine-DNA methyltransferase; CI, confidence interval; M–H, Mantel–Haenszel; OR, odds ratio.
Association of MGMT promoter methylation with clinical-pathologic features in gastric cancer

<table>
<thead>
<tr>
<th>Clinicopathological characteristics</th>
<th>N</th>
<th>OR (95% CI)</th>
<th>I²</th>
<th>P_h</th>
<th>P_bias</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>10</td>
<td>0.75 (0.51, 1.11)</td>
<td>52</td>
<td>0.03</td>
<td>0.170</td>
<td>0.15</td>
</tr>
<tr>
<td>Lauren classification</td>
<td>6</td>
<td>0.95 (0.62, 1.47)</td>
<td>38</td>
<td>0.15</td>
<td>0.079</td>
<td>0.82</td>
</tr>
<tr>
<td>Tumor invasion</td>
<td>7</td>
<td>0.79 (0.60, 1.04)</td>
<td>0</td>
<td>0.86</td>
<td>0.945</td>
<td>0.09</td>
</tr>
<tr>
<td>Distant metastasis</td>
<td>6</td>
<td>0.27 (0.18, 0.40)</td>
<td>1</td>
<td>0.41</td>
<td>0.435</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Lymph node status</td>
<td>9</td>
<td>0.65 (0.42, 1.01)</td>
<td>55</td>
<td>0.03</td>
<td>0.674</td>
<td>0.05</td>
</tr>
<tr>
<td>H. pylori infection</td>
<td>3</td>
<td>1.02 (0.54, 1.93)</td>
<td>0</td>
<td>0.68</td>
<td>0.881</td>
<td>0.94</td>
</tr>
<tr>
<td>Tumor location</td>
<td>3</td>
<td>0.9 (0.68, 1.20)</td>
<td>0</td>
<td>0.80</td>
<td>0.470</td>
<td>0.49</td>
</tr>
</tbody>
</table>

Note: Values in bold indicate statistical significance.

Abbreviations: H. pylori, Helicobacter pylori; MGMT, O6-methylguanine-DNA methyltransferase; N, total number of eligible studies; P_h, P-value of Egger linear regression test for evaluating publication bias; P_bias, P-value of Q test for heterogeneity among studies.

Association of MGMT promoter methylation with gastric cancer risk.

Several potential limitations were noted in this meta-analysis. First, significant heterogeneity between studies existed, but no sources of heterogeneity were found by meta-regression and subgroup analysis. The control group consisted of both normal gastric tissue adjacent to the tumor and normal gastric tissue, thus, the diversity of control groups may have affected on our research results. Thus, the nonuniform definition of control groups may lead to some heterogeneity. Second, many other factors, such as age as a risk factor for gastric cancer, MGMT mRNA expression, tumor grade, 10-year disease-free survival, disease-specific survival, and general demographic information, could not be assessed because of inadequate data. Large detailed studies should be included in further research. Third, we chose the only studies published in English, and this may contribute to potential selection bias, which may not be possible to avoid entirely. Finally, all included studies were retrospective; hence, it is impossible to determine whether MGMT promoter methylation is an early cancer-causing aberration or an influence of cancer progression. In the future, multiple prospective studies should be conducted to clarify this.

Although this study does have some limitations, it also has some strengths. Most importantly, our study showed a strong association of MGMT promoter methylation with risk of gastric cancer, which is consistent with previous findings that MGMT promoter methylation could be a risk factor for other types of cancer, such as colon adenocarcinoma, breast cancer, and non-small-cell lung cancer. Moreover, we found that MGMT promoter methylation may serve as a biomarker for monitoring gastric cancer metastasis, although many future studies are recommended to repeat these findings. MGMT promoter methylation has been associated with chemo-responsiveness with alkylating agent drugs; therefore, it is essential to detect the MGMT promoter methylation status in patients if they need treatment with alkylating agent drugs.

In summary, MGMT promoter hypermethylation is associated with gastric risk, distant metastasis and lymph node metastasis, which indicates that MGMT promoter methylation may play an important role in gastric cancer.
large-scale multicenter and well-matched cohort research studies are warranted to confirm our results and elucidate the exact mechanisms involved.

**Acknowledgment**

This work was supported by the National Natural Science Foundation of China (81171669).

**Disclosure**

The authors report no conflicts of interest in this work.

**References**