Differences in anti-inflammatory effects between two specifications of Scutellariae Radix in LPS-induced macrophages in vitro

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[ABSTRACT] Scutellariae Radix (SR), the root of Scutellaria baicalensis Georgi, is used as an antipyretic drug and has been demonstrated to have anti-inflammatory activity. SR is divided into two specifications, “Ku Qin” (KQ) and “Zi Qin” (ZQ), for use against different symptoms (upper energizer heat or lower portion of the triple energizer), according to the theory of traditional Chinese medicine (TCM). However, differences in the efficacies of these two specifications have not been determined. In the present study, we aimed to characterize the differences in the anti-inflammatory activities between KQ and ZQ and to explore how their differences are manifested in lipopolysaccharide (LPS)-induced macrophages. Our results showed that, in RAW264.7 cells (a mouse macrophage cell line derived from ascites), KQ and ZQ displayed anti-inflammatory effects by inhibiting the release of nitric oxide (NO), inducible NOS (iNOS), and nuclear factor-κB (NF-κB) in a dose-dependent manner without distinction. In NR8383 cells (a rat alveolar macrophage cell line), KQ and ZQ displayed similar effects on NO, iNOS, and NF-κB as seen in RAW264.7 cells, but KQ showed a higher inhibition rate for NO and iNOS than that shown by ZQ at the same concentration. These results indicated that there were differences in efficacy between KQ and ZQ in treating lung inflammation. Our findings provided an experimental evidence supporting the different uses of KQ and ZQ in clinic, as noted in ancient herbal records.

[KEY WORDS] Scutellaria baicalensis; “Ku Qin”; “Zi Qin”; Anti-inflammatory activity; Efficacy differences; RAW264.7 cells; NR8383 cells

[CLC Number] R965, R917

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Introduction

Scutellariae Radix (SR), the root of Scutellaria baicalensis Georgi (Lamiaceae), is used to treat upper respiratory tract infection, cough with lung heat, pneumonia, and other lung diseases [1]. The main active constituents of SR are flavonoid compounds, including baicalin, baicalein, wogonin, and wogonoside. SR has been demonstrated to have anti-inflammatory, antioxidant, antibacterial, antiviral, and antitumor activities [2]. SR is divided into two specifications, “Ku Qin” (KQ) and “Zi Qin” (ZQ), according to ancient records of traditional Chinese medicine (TCM) [3]. The perennial and hollow roots with dark cores are designated as KQ, and solid new roots are designated as ZQ. According to TCM theory, KQ is considered to be effective for clearing away upper energizer heat (the part from the throat to the diaphragm with the heart and lung being included), such as lung fire, hyperactive cough, and yellow sputum, whereas ZQ is used for the lower portion of the triple energizer, including the small and large intestines, kidney, and bladder, for symptoms such as hot and humid colonic diarrhea, hot and humid dysentery, and abdominal pain [4]. Currently, the SR preparations sold in most pharmacies in China are ZQ or mixed KQ and ZQ products and are used indistinguishably in clinic. Our previous research has revealed that the antipyretic and anti-inflammatory activities of 4-year-old SR are better than that of 2-year-old SR in yeast-treated mice and in a mouse auricle model established by dimethylbenzene [5]. Thus, KQ and ZQ may have different

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efficacies, as suggested in ancient records. However, previous studies on KQ and ZQ have only compared the differences in the content of their main active constituents but not differences in efficacy. Additionally, because of the complexity of the components in TCM, the pharmacological activities of individual components cannot fully determine the efficacy of medicinal herbs. Therefore, a comparative study on the efficacies and pharmacological activities of KQ and ZQ is particularly important.

The RAW264.7 cell line, established from a tumor induced by Abelson murine leukemia virus, is a macrophage cell line widely used in inflammation studies. The NR8383 cells were established from normal rat alveolar macrophage cells obtained by lung lavage. NR8383 cells exhibit characteristics of macrophages, such as rat alveolar macrophages, and can be used in vitro to study macrophage-related activities, especially in lung inflammation. SR has been confirmed to have anti-inflammatory properties by inhibiting iNOS, COX-2, PGE2, IL-1β, IL-6, IL-12, and TNF-α expression through the down-regulation of IKKα/β, IkBa, and NF-kB activation via the suppression of c-Raf-1/MEK1/2 and MAP kinase phosphorylation in LPS-induced RAW264.7 cells. It is reported that, in NR8383 cells, wogonin could inhibit the transcription and expression of TNF-α and MCP-1, reduce the concentration of PGE2, PLA2, LTB4, MDA, NO production and the activity of iNOS, and inhibit the nuclear translocation and expression of NF-κB by repressing TLR7-mediated MyD88-dependent signaling pathway to reduce the inflammatory response in influenza infection. Based on the characteristics of SR, we considered that a model with NR8383 cells was a more appropriate system to evaluate the differences in efficacy between KQ and ZQ and used RAW264.7 cells as a control, which are not organ specific.

In the present study, we hypothesized that KQ and ZQ would have different efficacies in NR8383 cells, which are located only in the lungs. To test this hypothesis, the anti-inflammatory activities of KQ and ZQ were assessed in LPS-induced NR8383 and RAW264.7 cells, in order to provide a preliminary explanation for the different clinical uses of KQ and ZQ.

**Materials and Methods**

**Reagents**

Ham's F12K medium and lipopolysaccharide were purchased from Sigma-Aldrich (Saint Louis, MO, USA). Fetal bovine serum (FBS) was purchased from BI (Beit Haemek, Israel). Dulbecco's modified Eagle's medium (DMEM), phosphate-buffered saline (PBS), the Bicinchoninic Acid (BCA) Protein Assay Kit, and other cell culture reagents were purchased from Solarbio (Beijing, China). Griess reagent and NP-40 lysis buffer were purchased from Beyotime (Shanghai, China). Antibodies specific for iNOS, IkBa, p65, GAPDH, and β-tubulin were purchased from Santa Cruz Biotechnology (Santa Cruz, CA, USA). Anti-mouse and anti-rabbit immunoglobulin G (IgG)-conjugated horseradish peroxidase (HRP) were purchased from EarthOx (San Francisco, CA, USA).

**Crude drugs**

The crude drugs of KQ and ZQ (Table 1) were confirmed to be the roots of *Scutellaria baicalensis* by Professor CAI Shao-Qing of Peking University (Beijing, China). All of the drugs were screened according to the characteristics of KQ and ZQ (Fig. 1).

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Specification</th>
<th>Collection time</th>
<th>Collection location</th>
<th>Producing area</th>
<th>Wild/Cultured</th>
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<td>Anguo, Hebei</td>
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<td>6630</td>
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<td>7486</td>
<td>“Ku Qin”</td>
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<tr>
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<td>7711</td>
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**Cell culture**

The NR8383 cells and RAW264.7 cells used in the present study were obtained from the Cell Bank of the Chinese Academy of Sciences (Shanghai, China). The NR8383 cells were cultured in Ham’s F12K medium supplemented with 20% FBS and the RAW264.7 cells were cultured in DMEM with 10% FBS, in a humidified atmosphere of 5% CO2 at 37 °C. The appropriate amount of the crude drugs was ground into powder and sieved through a 40-mesh sieve. Precisely weighed powders were extracted using 100-fold boiled distilled water at 100 °C for 30 min. The extracted solution was filtered through
filter paper, evaporated to dryness, and then freeze-dried. The yields of the water extracts obtained are shown in Table 2.

**Fig. 1** Photographs of the SR samples used in the present study. (A) 6590-KQ; (B) 6596-ZQ; (C) 6628-KQ; (D) 6630-ZQ; (E) 7486-KQ; (F) 7487-ZQ; (G) 7711-KQ; (H) 7712-ZQ; (I) 7591-KQ; (J) 7593-ZQ

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>6590</th>
<th>6596</th>
<th>6628</th>
<th>6630</th>
<th>7486</th>
<th>7487</th>
<th>7711</th>
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<tr>
<td>Yield (%)</td>
<td>41.62</td>
<td>48.90</td>
<td>43.79</td>
<td>54.03</td>
<td>45.94</td>
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<td>47.25</td>
<td>56.19</td>
<td>46.52</td>
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</table>

**Cytotoxicity assay**

The NR8383 cells were seeded at $1 \times 10^5$ cells/well in a 96-well plate and incubated with various concentrations of KQ and ZQ (0.5, 1, 2, and 5 mg·mL$^{-1}$, dissolved in cell culture medium) for 6, 12, 24, and 48 h. The RAW264.7 cells were seeded at $1 \times 10^4$ cells/well in a 96-well plate and incubated with various concentrations of KQ and ZQ (50, 100, 200, and 500 µg·mL$^{-1}$) for 6, 12, 24, and 48 h. CCK-8 solu-
Effects of KQ and ZQ on cell viability in macrophages

Concentrations of KQ and ZQ (50, 100, and 200 µg·mL⁻¹) for 6, 12, 24, and 48 h in the presence of various concentrations of KQ and ZQ (0.5, 1, and 2 mg·mL⁻¹). The RAW264.7 cells were seeded at a density of 2.5 × 10⁵ cells/well in a 48-well plate and incubated with lipopolysaccharide (LPS; 1 µg·mL⁻¹) for 6, 12, 24, and 48 h in the presence of various concentrations of KQ and ZQ (50, 100, and 200 µg·mL⁻¹). The NO concentration in the cultured medium was determined via the Griess reaction. Specifically, 50 µL of supernatant from each well was mixed with 100 µL of Griess reagent (1% sulfanilamide in 5% phosphoric acid and 0.1% naphthylenediamine dihydrochloride in water) in a separate 96-well plate. After an incubation of 15 min at room temperature, the optical density was determined at 540 nm using a microplate reader.

Measurement of nitric oxide (NO) production

The NR8383 cells were seeded at a density of 2.5 × 10⁵ cells/well in a 48-well plate and incubated with lipopolysaccharide (LPS; 1 µg·mL⁻¹) for 6, 12, 24, and 48 h in the presence of various concentrations of KQ and ZQ (0.5, 1, and 2 mg·mL⁻¹). The RAW264.7 cells were seeded at a density of 2.5 × 10⁵ cells/well in a 48-well plate and incubated with LPS (1 µg·mL⁻¹) for 6, 12, 24, and 48 h in the presence of various concentrations of KQ and ZQ (50, 100, and 200 µg·mL⁻¹). The NO concentration in the cultured medium was determined via the Griess reaction. Specifically, 50 µL of supernatant from each well was mixed with 100 µL of Griess reagent (1% sulfanilamide in 5% phosphoric acid and 0.1% naphthylenediamine dihydrochloride in water) in a separate 96-well plate. After an incubation of 15 min at room temperature, the optical density was determined at 540 nm using a microplate reader.

Western blotting analysis

The NR8383 cells and RAW264.7 cells were cultured at 2 × 10⁵ cells/well in 6-well plates and incubated with LPS (1 µg·mL⁻¹) in the presence of various concentrations of KQ and ZQ (as previously described) for 6 or 24 h. After incubation, cells were collected by centrifuging at 1 000 × g for 5 min and then washed with cold PBS three times. To each tube of cells, NP-40 lysis buffer (mixed with protease inhibitor and PMSF) was added followed by vortexing for 1 min. The mixture was centrifuged at 12 000 × g for 10 min at 4 °C, and the supernatants (cytosolic protein) were collected. The protein concentration in the samples was determined by BCA protein concentration assay. The protein samples (40 µg) were subjected to SDS-PAGE, separately transferred to polyvinylidene difluoride membranes, which were treated with 5% fat-free milk powder to block nonspecific IgGs and incubated overnight at 4 °C with specific antibodies to iNOS, IkBα, p65, GAPDH, and β-tubulin. The blots were subsequently incubated with anti-mouse or anti-rabbit IgG HRP for 1 h. The immunoreactivity was detected using enhanced chemiluminescence. Quantitative data were obtained using computed densitometry with the Quantity One image analysis software system (Bio-Rad, Hercules, CA, USA).

Statistical analysis

The data were analyzed using one-way analysis of variance (ANOVA) and Student's t-test with GraphPad InStat 6.0 software (GraphPad Software, Inc., La Jolla, CA, USA). The data are presented as mean ± SEM. P < 0.05 was considered statistically significant.

Results

Effects of KQ and ZQ on cell viability in macrophages

The viabilities of NR8383 and RAW264.7 cells in the presence of KQ and ZQ are shown in Fig. 2. The cytotoxicity of KQ and ZQ (up to a concentration of 5 mg·mL⁻¹) in NR8383 cells and that of SR (up to a concentration of 200 µg·mL⁻¹) in RAW264.7 cells were not obvious after 6, 12, and 24 h of incubation. Based on this result, concentrations of up to 2 mg·mL⁻¹ and 200 µg·mL⁻¹ for KQ and ZQ, respectively, were chosen for subsequent experiments.

Effects of KQ and ZQ on NO release in LPS-induced RAW264.7 cells

The effects of five groups of drugs on NO production in LPS-induced RAW264.7 cells were examined to verify their anti-inflammatory activities. Based on preliminary observations, 1 µg·mL⁻¹ LPS was chosen to stimulate cells to trigger an inflammatory response (Fig. 3A-B). As shown in Fig. 4A, the most obvious inhibitory effects of KQ and ZQ on NO occurred at 48 h.

After incubating RAW264.7 cells with the five groups of drugs at a concentration of 200 µg·mL⁻¹ for 48 h, all of the drugs were found to have significant inhibitory effects on NO production (Fig. 4B). In addition, KQ and ZQ significantly suppressed LPS-induced NO release in a concentration-dependent manner compared to the LPS-treated cells without SR (Fig. 4C). These results confirmed that KQ and ZQ could inhibit the release of NO in RAW264.7 cells without any differences between them, indicating that KQ and ZQ had equivalent anti-inflammatory activities in the RAW264.7 cells.

Effects of KQ and ZQ on NO release in LPS-induced NR8383 cells with different intensities

The effects of the five groups of drugs on NO production in LPS-induced NR8383 cells were then examined. Based on preliminary observations, 1 µg·mL⁻¹ of LPS was chosen to trigger an inflammatory response in the cells (Fig. 3C). As shown in Fig. 5A, there was more than a 30% difference between KQ and ZQ when the cells were exposed to them for 12 h and 24 h, and the effect of KQ was more obvious at 24 h.

The NR8383 cells were incubated with the five groups of drugs at a concentration of 2 mg·mL⁻¹ for 24 h, and four groups of the drugs had significant inhibitory effects on NO production (Fig. 5B). Interestingly, for three of these groups, there were significant differences between KQ and ZQ, and the difference was most obvious between 7591-KQ and 7593-ZQ. KQ and ZQ also significantly suppressed LPS-induced NO release in a concentration-dependent manner as in the RAW264.7 cells, but KQ had a higher inhibition rate (up to 28%) than ZQ at the three concentrations (Fig. 5C). These results demonstrated that KQ and ZQ also had inhibitory effects on NO in the NR8383 cells and that there were differences in efficacy between them, indicating that the varying anti-inflammatory activities of KQ and ZQ could be observed in the cells but not in the RAW264.7 cells.

Effects of KQ and ZQ on iNOS expression via the NF-xB pathway in LPS-induced NR8383 cells

The iNOS expression and NF-xB pathway-related proteins were evaluated to explore the reasons for the efficacy differences between KQ and ZQ in the NR8383 cells. The LPS-induced NR8383 cells were exposed to KQ and ZQ for 12 h, and the results indicated that three concentrations (0.5, 1,
Fig. 2  (A) Effects of KQ and ZQ on NR8383 cell viability. Macrophages were exposed to 0.5, 1, 2, and 5 mg·mL$^{-1}$ KQ or ZQ for 6, 12, 24, and 48 h. (B) Effects of KQ and ZQ on RAW264.7 cell viability. Macrophages were exposed to 50, 100, 200, and 500 µg·mL$^{-1}$ KQ or ZQ for 6, 12, 24, and 48 h. Data are presented as the mean ± SEM for $n = 3$. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$ vs control

Fig. 3  Effects of LPS on NO production in macrophages. (A) RAW264.7 cells were stimulated with LPS (1 µg·mL$^{-1}$) for 1, 6, 12, and 24 h. (B) RAW264.7 cells were stimulated with LPS (1, 5, and 10 µg·mL$^{-1}$) for 24 h. (C) NR8383 cells were stimulated with LPS (1, 5, and 10 µg·mL$^{-1}$) for 1, 6, 12, and 24 h. Data are presented as the mean ± SEM for $n = 3$. **$P < 0.01$, ***$P < 0.001$ vs control
Fig. 4  Effects of KQ and ZQ on NO production in LPS-induced RAW264.7 cells. (A) Macrophages were exposed to KQ and ZQ (200 µg·mL\(^{-1}\)) with LPS (1 µg·mL\(^{-1}\)) for 6, 12, 24, and 48 h. (B) Macrophages were exposed to five pairs of KQ and ZQ (200 µg·mL\(^{-1}\)) with LPS (1 µg·mL\(^{-1}\)) for 48 h. (C) Macrophages were exposed to 50, 100, and 200 µg·mL\(^{-1}\) KQ and ZQ with LPS (1 µg·mL\(^{-1}\)) for 48 h. Data are presented as the mean ± SEM for \(n = 3\). * \(P < 0.05\), *** \(P < 0.001\) vs the LPS group.

Fig. 5  Effects of KQ and ZQ on NO production in LPS-induced NR8383 cells. (A) Macrophages were exposed to KQ and ZQ (2 mg·mL\(^{-1}\)) with LPS (1 µg·mL\(^{-1}\)) for 6, 12, 24, and 48 h. (B) Macrophages were exposed to five pairs of KQ and ZQ (2 mg·mL\(^{-1}\)) with LPS (1 µg·mL\(^{-1}\)) for 24 h. (C) Macrophages were exposed to 0.5, 1, and 2 mg·mL\(^{-1}\) KQ and ZQ with LPS (1 µg·mL\(^{-1}\)) for 24 h. Data are presented as the mean ± SEM for \(n = 3\). * \(P < 0.05\), ** \(P < 0.01\), *** \(P < 0.001\) vs the LPS group; # \(P < 0.05\), ## \(P < 0.01\), ### \(P < 0.001\) KQ vs ZQ for the same concentration.
and 2 mg·mL$^{-1}$) of KQ inhibited the expression of iNOS, whereas ZQ inhibited it only at 2 mg·mL$^{-1}$ (Fig. 6A). At the same concentration, KQ showed higher inhibitory effects than ZQ, especially at 1 mg·mL$^{-1}$. The up-regulation effect on p65 by the KQ and ZQ treatments increased with increasing concentrations, and the difference increased with increasing concentrations (up to 47% at 2 mg·mL$^{-1}$), as the up-regulation effect of KQ was more obvious (Fig. 6B). Similarly, their effect on IκBα differed by as much as 100% at 2 mg·mL$^{-1}$ (Fig. 6C). These results suggested that the efficacy differences between KQ and ZQ might result from effects on upstream target proteins, which were eventually reflected in the downstream effector molecules.

**Results**

**Fig. 6** Effects of KQ and ZQ (0.5, 1, and 2 mg·mL$^{-1}$) on iNOS, p65, and IκBα in LPS-induced NR8383 cells. (A) Effects of KQ and ZQ on iNOS. (B) Effects of KQ and ZQ on p65. (C) Effects of KQ and ZQ on IκBα. Data are presented as the mean ± SEM for $n = 3$. *$P < 0.05$, **$P < 0.01$, vs the LPS group; *$P < 0.05$, KQ vs ZQ at the same concentration.

**Effects of KQ and ZQ on iNOS expression via the NF-κB pathway in LPS-induced RAW264.7 cells**

To determine if there would be any differences between the effects of KQ and ZQ on iNOS and NF-κB in the RAW264.7 cells as seen with NO production, the LPS-induced RAW264.7 cells were exposed to KQ and ZQ for 24 h. Our results showed that there were no differences in the inhibitory effect on iNOS or the up-regulation effect on p65 and IκBα between the KQ and ZQ treatments (Fig. 7), indicating that the efficacy differences between KQ and ZQ in the NR8383 cells were not observed in the RAW264.7 cells.

**Discussion**

In the present study, we investigated the anti-inflammatory activities of KQ and ZQ in LPS-induced macrophages to prove that there were differences in their efficacies and to explore the mechanisms for the differences. The differences in the efficacies of KQ and ZQ were reflected in their effects on NO, iNOS, and NF-κB pathways in the NR8383 cells but not in the RAW264.7 cells (Table 3). The LPS-induced macrophage inflammatory model was used in the present study to evaluate the anti-inflammatory activity of KQ and ZQ. Previous studies have reported that SR inhibits iNOS expression, NO release, and inflammation-related cytokines through the NF-κB pathway$^{[11,14]}$. In the present study, the concentration of nitrite in the cell culture medium after the KQ and ZQ treatments revealed that KQ and ZQ began to inhibit NO release after 48 h administration in the RAW264.7 cells and that all five groups of drug samples collected from different areas were able to inhibit NO release, confirming that KQ and ZQ had anti-inflammatory activities. Further analysis showed that their effects on NO occurred in a concentration-dependent manner. However, there was no difference between KQ and ZQ at the same concentrations.
In the NR8383 cells, KQ and ZQ started to inhibit NO release after 24 h, and the efficacy difference appeared at 12 h and 24 h. Three groups of drugs showed efficacy differences in the four active groups. Compared with the same concentration of ZQ, KQ displayed significantly higher effects on NO at 1 and 2 mg·mL⁻¹. Our previous work found that the content ratios of aglycones and glycosides (baicalein and baicalin, wogonin and wogonoside, oroxylin A and oroxylin A 7-O-glucuronide) were significantly higher in KQ than in ZQ in 30 samples collected from different areas [15]. These results indicated that the content of flavonoid glycosides was lower in KQ and that the content of flavonoid aglycones was higher in KQ compared with ZQ. Moreover, according to our study data, the contents of the glycosides in the group of 6590-6596 and 6628-6630 were higher than those in the group of 7486-7487 and 7591-7593 (Table 4), while the efficacy difference was only observed in group 7486-7487 and 7591-7593. Baicalin can only be absorbed after hydrolysis by intestinal bacteria, and baicalein can be directly absorbed by the small intestine, indicating that baicalein can be absorbed faster than baicalin [16]. Therefore, KQ may play a more effective anti-inflammatory role in alveolar macrophages because of its higher content of aglycones compared with ZQ. Further studies are needed to characterize the relationship between quality and efficacy.

Macrophages are essential components of the downstream effects of the immune system. Monocytes derived from embryonic precursors in the bone marrow turn into macrophages when tissues are damaged. Under homeostatic conditions, macrophages located in different tissues acquire distinct morphological and functional features for various
requirements [17]. The absence of difference in efficacy between KQ and ZQ in RAW264.7 cells suggested that using lung derived cells may provide a better indication of the effects of the drugs on lung diseases.

| Table 4 | Peak areas of baicalin, wogonoside and oroxylin A 7-O-glucuronide in the four groups of KQ and ZQ |
|------------------|----------------------------------------------------------|-----------------|-----------------|
|                  | Peak area (mAU*s)                                         | Baicalin       | Wogonoside      | Oroxylin A 7-O-glucuronide |
| 6590             | KQ                                                       | 32 543.8       | 8 314.7         | 4 987.3                    |
| 6596             | ZQ                                                       | 37 619.2       | 11 075.7        | 5 086.7                    |
| 6628             | KQ                                                       | 48 692.4       | 12 034.3        | 2 461.8                    |
| 6630             | ZQ                                                       | 45 560.4       | 11 964.1        | 2 571.3                    |
| 7486             | KQ                                                       | 25 418         | 6 871.2         | 2 256.2                    |
| 7487             | ZQ                                                       | 36 839.8       | 9 103.3         | 3 335.8                    |
| 7591             | KQ                                                       | 33 175         | 9 371.4         | 3 571.9                    |
| 7593             | ZQ                                                       | 30 935.1       | 7 240.4         | 2 489.1                    |

The Western blotting results showed that KQ and ZQ significantly inhibited the expression of iNOS and had effects on the NF-κB pathway by regulating the expression of iNOS (Fig. 8). In the inflammatory response, the NF-κB pathway is activated, p65 is translocated into the nucleus after the phosphorylation of IκBα to activate inflammation-related genes, and inflammatory mediators are then highly expressed, such as IL-6, TNF-α, and iNOS, which promote the formation of NO and its release outside of cells [18-19]. In the present study, KQ displayed larger effects on p65, and IκBα and significantly larger effects on NO, and iNOS compared to ZQ in the NR8383 cells, suggesting that the efficacy differences between KQ and ZQ might result from effects on the upstream target proteins, which are eventually reflected in the downstream effector proteins and molecules. However, the mechanism of responsible for the differences between KQ and ZQ still requires clarification, including their effects on other inflammation-related pathways and inflammatory factors in vitro and in vivo.

In conclusion, the present study confirmed that KQ and ZQ had anti-inflammatory activities in the RAW264.7 and NR8383 cells and demonstrated that there were significant differences in efficacy between KQ and ZQ in the NR8383 cells. These results provided experimental evidence that supports the different uses of KQ and ZQ in the clinic, as noted in ancient herbal records. However, further experiments are needed to fully understand the mechanisms of action and the specific targets of KQ and ZQ.

![Fig. 8](image_url)  Proposed mechanisms of action for the effect of KQ and ZQ on the NF-κB pathway in LPS-induced macrophages. *P < 0.05
required to elucidate the relationship between quality and efficacy, which will provide a more scientific basis for clinical use of the two different specifications of SR, KQ and ZQ.

References


Dr. WANG Xuan, Ph.D. & Professor, Peking University

Dr. Wang has been working on activity-oriented analysis of Traditional Chinese Medicine (TCM) in vitro and in vivo. While TCM has been used effectively in the clinic, there are few analytical approaches in therapeutic efficacy-oriented quality control of TCM. These problems prompted her and her team to focus on bioactive constituents with multiple combination of pharmaceutical analysis and biology methods. They have developed a mitochondria-based centrifugal ultrafiltration/liquid chromatography/mass spectrometry method for screening mitochondria-targeted bioactive constituents from TCM and established an Eastern Blotting Technique for the visual detection of Aristolochic Acids by using a monoclonal antibody.

Dr. Wang’s research interest also include Quality Control of TCM. They have developed and reported analysis methods for the identification of Panax notoginseng and Scutellariae baicalensis with different growth year, area and specifications and so on.

Dr. Wang and her team have published multiple articles in influential journals in related fields and her current projects are funded by National Natural Science Foundation of China, Pharmacopoeia, etc.