Minimum Standards of Reporting Checklist

*BioMed Central* advocates full and transparent reporting. Please ensure that your paper provides the information requested below where applicable. On submitting your paper you will be asked to confirm you have included this information, or give reasons for any instances where it is not made available. You will also be asked to upload this file and it should be cited in the Methods section.

**Experimental design and statistics**

The following information should be included in the Methods section and inserted in the table below:

| Question                                                                 | Answer                                                                                                                                 |
|--------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------|
| 1. The exact sample size (n) for each experimental group/condition (as a number, not a range). Include details of a power analysis if done, or any other relevant considerations that determined the choice of sample size. For n < 6, individual data values should be shown rather than summary statistics alone. | Semi-wild huangqi of 1, 2, 3, 4, 5, 8, and 10 years of age was collected separately from Hunyuan County (Table 1) in September 2012. 1-year-old huangqi had 5 samples; 2-year-old huangqi had 8 samples; 3-year-old huangqi had 8 samples; 4-year-old huangqi had 8 samples; 5-year-old huangqi had 5 samples; 8-year-old huangqi had 3 samples and 10-year-old huangqi had 1 sample. |
| 2. A description of sample collection that enables the reader to understand whether the samples represent technical or biological replicates, and an explanation of inclusion/exclusion criteria if samples or organisms were excluded from the analysis. | Semi-wild huangqi of 1, 2, 3, 4, 5, 8, and 10 years of age was collected separately from Hunyuan County (Table 1) in September 2012. The above-ground parts were retained as plant specimens. The roots were dried in the sun for experimental analysis. Sections for a negative control experiment were treated with 70% alcohol for 1 month to remove flavonoids and saponins. |
| 3. How samples/organisms were allocated to experimental groups and processed, and full details of the randomisation procedure used (if relevant). | Segments of the roots of 18-year-old huangqi (length 60 cm, diameter 2-3 cm) were cut transversely (5 mm thick) at 5-mm intervals from the root head to the root tip. These segments were used to prepare positive controls. Huangqi samples of 1, 2, 3, 4, and 5 years of age were used for microscopic examination of growth rings. Root segments were cut at approximately 2 cm from the root head, excluding any rotten tissue. Parts of the segments were cut (5 mm thick), and from these, positive sections were prepared (as described previously) for both microscopic observation. Other parts of the segments were sections at 1-mm thickness by hand with a scalpel. Huangqi samples were classified into three categories based on the extent of rotten tissue: (A) "normal" (B) "small rotten head" (parenchyma, 1/3 of the total diameter), and (C) "large rotten head" (parenchyma to 2/3 of the total diameter). |
| 4. For sample assessment by human investigators, a statement on whether the investigator was blinded to group assignment and outcome assessment, and how this blinding was achieved and evaluated (if relevant). | All samples were authenticated by Professor Huasheng Peng (School of Pharmacy, Anhui University of Chinese Medicine) with reference to the Flora of China [27]. 27. Chinese Academy of Sciences, China flora editorial Committee. Flora of China. Vol 42. Science Press, Beijing,1993:133. |
5. How many times each experiment shown was replicated and an indication of the extent of variation from experiment to experiment.

6. Information on the statistical methods and measures used. It should be clear whether the tests are one-sided or two-sided, whether there are adjustments for multiple comparisons, whether medians or means are being shown, whether error bars are standard deviations (SD), standard error of mean (SEM) or confidence intervals.

7. A justification for the appropriateness of statistical tests used to assess significance. Do the data meet the assumptions of the tests? Is there an estimate of variation within each group of data, and is the variance similar between groups that are being statistically compared?

In addition, information essential to interpreting the data presented should be made available in the figure and table legends.

If the study involves health interventions for human participants, please refer to the relevant reporting guidelines from the EQUATOR Network, and the Biosharing Portal for reporting checklists for biological and biomedical research, where applicable.

Research involving humans

If your research involved humans, please confirm you have adhered to the relevant reporting guideline from the EQUATOR Network, and included the completed checklist as an additional file with your submission:

| Answer (page and line number inserted/Not applicable for my study) |
|---------------------------------------------------------------|
| - I have followed the relevant reporting for my study type, and included a populated checklist with my submission |
| - Not applicable for my study |

Each sample preparation was performed in triplicate.

The standard curves of the HPLC method, the correlation between growth rings and age, and correlations between the concentrations of the six examined bioactive constituents (calycosin, calycosin-7-glucoside, formononetin, ononin, astraglaside A, and astraglaside II) and age were analyzed using SPSS 19.0 software. The T test we used was two-sided. P-value <0.05 indicated that there was significant difference, whereas P ≥ 0.05 indicated that there was no significant difference. Data of the concentration of six compounds are expressed as the mean ± standard deviations.

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SPSS 19.0 software was used to analyze the correlation between growth rings (X) and age (Y). The result showed a significant correlation (Y = X, r = 1.000, P = 0.000).

The correlations between years of growth and the four flavonoids were as follows: calycosin-7-glucoside (P = 0.88, r = 0.08), ononin (P = 0.848, r = 0.102), calycosin (P = 0.859, r = 0.095), and formononetin (P = 0.657, r = -0.233). There were no significant correlations between these four flavonoids and years of growth.

The SPSS analysis results for the relationship between years of growth and astraglaside A revealed a high correlation (P = 0.029, r = -0.233). The relationship between years of growth and astraglaside II also showed high correlation (P = 0.012, r = -0.858).

Table 1

| TRR: the number of growth rings observed with the naked eye after staining of transverse sections with phloroglucinol–HCl reagent |
|---------------------------------------------------------------|
| TRM: the number of growth rings observed using light microscopy |

- The yellow arrows represent growth rings.
## Resources

A description of all resources used should be included in the Methods section, with enough information to allow them to be uniquely identified. The table below should be completed with confirmation that this was done (i.e. included in the Methods section) or is not applicable. If this has not been completed, but is applicable, you should contact the journal editorial staff before proceeding.

| Resource Category | Answer (page and line number inserted/Not applicable for my study) |
|-------------------|---------------------------------------------------------------|
| Antibodies        | Not applicable for my study                                  |
| Cell lines        | Not applicable for my study                                  |
| Organisms         | Not applicable for my study                                  |
| Tools             | Page 9, line 185.                                            |

- **Antibodies**: report source, catalogue code, characteristics, dilutions and how they were validated for the system under study.

- **Cell lines**: report source, whether identity has been authenticated and whether tested for mycoplasma contamination. We encourage researchers to check the NCBI database for contamination of cell lines.

- **Organisms**: report source, species, strain, sex, age, husbandry, inbred and strain characteristics of transgenic and mutant animals.

- **Tools (software, databases and services)**: report standard tool name, provider and version number, if available. For antibodies, model organisms (mice, zebrafish and flies) and tools, authors are strongly encouraged to cite Research Resource Identifiers (RRIDs). To do so, please go to the Resource Identification Portal to search for your research resource and insert the reference text into your Methods section.
**Availability of data and materials**

The table below should be completed with confirmation that this was done (i.e. included in the Methods section) or is not applicable.

| Description                                                                 | Answer                                                                 |
|-----------------------------------------------------------------------------|------------------------------------------------------------------------|
| All datasets on which the conclusions of the paper rely must be either deposited in publicly available repositories (where available and ethically appropriate) or presented in the main paper or additional supporting files, in machine-readable format whenever possible. If authors are unable to fulfil this requirement, they should contact journal editorial staff, after checking our list of Recommended Repositories. | Page 17, line 356.                                                     |
| Links to deposited datasets, or datasets in additional files, should be explicitly referenced in a section entitled “Availability of Data and Materials”. Guidance on where to deposit your data can be found on the Availability of Data and Materials policy page. | Page 18, line 370.                                                     |
| If computer code was used to generate results that are central to the paper’s conclusions, include a statement in the “Availability of data and materials” section to indicate how the code can be accessed. Include version information and any restrictions on availability. For deposited data and published code, a full reference with an accession number, doi or other unique identifier should be included in the reference list. | Not applicable for my study                                             |
| If reproducible materials are generated as a result of the research (for example new animal mutants), a statement on their availability should be included. | Not applicable for my study                                             |