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# The monitoring of some heavy metals in oriental mineral medicines

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# 광물성 생약 중 중금속의 모니터링

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**Abstract:** This paper reports on the monitoring results of some heavy metals (Pb, Cd, As and Hg) in oriental mineral medicines. Levels of Pb, Cd and As were determined using the acid digestion method and ICP-MS. Hg levels were determined using the gold amalgamation method and Automatic mercury analyzer (AMA). The results indicated that, in the case of Pb, 25.81% (24 cases) of the samples were over MRL (5 mg/kg). Pb levels in Calamina, Pumex, and Cinnabaris exceeded MRL (5 mg/kg). In the case of Cd, 9.68% (17cases) of the samples were over MRL (0.3 mg/kg). Cd levels in Calamina, Pumex and Cinnabaris were exceeded MRL (0.3 mg/kg). In case of As, 24.73% (23 cases) of samples exceeded MRL (3 mg/kg). As levels in Calamina, Lithargyrum, Vermilionum, Cinnabaris and Chalcocitum exceeded MRL (3 mg/kg). In the case of Hg, it was shown that the 76.34% (78 cases) of the samples exceeded MRL (0.2 mg/kg). Hg levels in almost all the samples exceeded MRL (0.2 mg/kg). These results led us to recommend that the domestic commercial law to be legislated to ensure the quality estimation and safety of oriental mineral medicines.

**요 약:** 본 연구에서는 시중에 유통중인 광물성 생약에서의 중금속(Pb, Cd, As, Hg)에 대해 모니터링 하였다. Pb, Cd, As의 경우 시료 전처리는 습식분해법으로 하였으며, ICP-MS를 이용하여 검출하였고 Hg의 경우 AMA를 이용하여 모니터링 하였다. 모니터링의 결과 전체 시료(92)의 25.81%(24)가 Pb를 함유하고 있었으며 능자광(85.5 mg/kg), 부석(41.2 mg/kg), 진사(20.4 mg/kg)에서 기준치(0.5 mg/kg) 이상으로 검출되었다. Cd의 경우 전체 시료(92)의 9.68%(17)가 함유되어있었으며, 능자광(345 mg/kg), 부석(0.44 mg/kg), 진사(2.78 mg/kg)에서 기준치(0.3 mg/kg) 이상으로 검출되었고, 전체 시료(92)의 24.73%(23)가 As

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를 함유하고 있었으며, 능자광(5.72 mg/kg), 조산화납(22.2 mg/kg), 황화제이수은(10.0 mg/kg), 진사(12.5 mg/kg), 흑운모(14.6 mg/kg)에서 기준치(3 mg/kg)이상으로 검출되었다. Hg의 경우는 전체 시료(92)의 76.34%(78)가 함유되어 있었으며, 전체 13 종 광물 중 10종에서 기준치(0.2 mg/kg) 이상으로 검출되었다. 따라서, 국내에 유통되고 있는 광물성 생약재의 품질 평가와 안전관리를 보완하여 광물성 생약에 대한 신뢰성과 안전성을 확보할 필요성이 있는 것으로 판단된다.

**Key words :** monitoring; four heavy metals (Pb, Cd, As, Hg); oriental mineral medicines; inductively coupled plasma-mass spectrometry; automatic mercury analyzer

## 1. Introduction

The use of medicinal plants in therapeutics or as dietary supplements goes back to beyond the recorded history and has increased substantially in the last few decades.[1-3] Medicinal plants play an important role in the traditional medicines and are widely consumed as home remedies. The past decade has seen a significant increase in the use of mineral medicines because of their relatively minor side effect, availability and acceptability to the majority of consumers. Environmental pollution, atmosphere, soil, harvesting and handling are some of the factors, which play an important role in contaminating medicinal plants by metals and microbial growth.[4-5]

However, the safety of their use has recently been questioned because of the reports of illness and fatalities.[6-7] Especially, heavy metals turned out to be detrimental to human beings. For instance, Pb and Hg cause adverse effects on the renal and nervous systems with potential toxic effects in the fetus.[8] Likewise, Cd causes nephrotoxicity in humans, mainly due to abnormalities of tubular re-absorption.[9] Although heavy metal poisonings associated with the presence of toxic metals in medicinal plants were reported in Asia, Europe, and the United States, very little work is on record regarding heavy metal accumulation in herbal medicines of medicinal plants, and now it has gained the attention of researchers worldwide because of a developing faith in herbal drugs owing to various ill effects of synthetic drugs.[10-13] Thus, there is an urgent need to establish the identity, purity and quality assurance of herbal drugs to have full efficacy and safety of the mineral products.

In the work, we studied the monitoring of Pb, Cd, As and Hg in the mineral medicines. We analyzed 92 mineral medicines of 13 kinds purchased in Korean markets that are made in Korea and China.

## 2. Experimental

### 2.1. Samples collected

The 92 samples from 13 mineral medicine species were collected from different sources. The samples were kept in plastic bags until their analysis. The species and numbers of collected samples are listed in *Table* 1.

### 2.2. Apparatus

ICP-MS model Elan 6100 DRC plus (Perkin Elmer Sciex, Ontario, Canada) was applied for determination

*Table* 1. The list of species and numbers of collected samples

| Name | No. of collected Samples |
|---|---|
| Calamina | 6 |
| Lithargyrum | 5 |
| Tremolitum | 10 |
| Vermilionum | 4 |
| Muscovitum | 10 |
| Cinnabaris | 3 |
| Halloysitum Rubrum | 10 |
| Chalcocitum | 4 |
| Glauberitum | 10 |
| Pumex | 8 |
| Calcitum | 10 |
| Stalactitum | 2 |
| Fluoritum | 10 |

*Table* 2. Operating conditions for ICP-MS

| Descriptions | Conditions |
|---|---|
| R.F. generator | Free-running type, 40 MHz |
| R.F. power | 1400 W |
| Coolant gas flow rate | 17.0 L/min. |
| Auxiliary gas flow rate | 2.00 L/min. |
| Nebulizer gas flow rate | 1.05 L/min. |
| Sample uptake flow | 1.00 mL/min. |
| Nebulizer | Concentric type |
| Spray chamber | Cychronic type |
| Torch | Demountable |
| Interface cones | Platinum |
| Quadrupole chamber | $1\times10^{-6}$ torr |
| Dwell time | 600 ms |
| Pb/Mass | 208, 206 |
| Cd/Mass | 114, 112 |
| As/Mass | 75 |

*Table* 3. Operating conditions for AMA

| Descriptions | Conditions |
|---|---|
| Wavelength | 253.65 nm |
| Interface filter | 254 nm |
| Bandwidth | 9 nm |
| Detector | Silicon UV photo detector |
| Working range | 0.02~600 ng Hg |
| Drying Temp. | 300°C |
| Drying Time | 80 sec. |
| Decomp. Temp. | 850°C |
| Decomp. Time | 150 sec. |
| Waiting Time | 60 sec. |
| Amalgam-heating Time | 12 sec. |
| Recording Time | 30 sec. |
| Sampling mode | Auto |

of Pb, Cd and As in mineral medicines. Automatic mercury analyzer model AMA 254 (Milestone, Connecticut, US) was applied for determination of Hg. The instrumental operating conditions for determination of the heavy metals are summarized in *Table* 2 and 3.

### 2.3. Reagents and materials

All reagents used in this study were of analytical grade. Also nitric acid, hydrochloric acid, sulfuric acid (Dong Woo Fine-Chem, Kyeong gi, Korea) and hydrofluoric acid (J.T. Baker, NZ, USA) were used in the acid digestion method. Multi element stock solution (PE sciex Concord, Ontario, Canada) was used for calibration curve. The hot plate was used (MTOPS, Kyeong gi, Korea) for acid digestion method.

### 2.4. Analytical procedure

Before experiment, the major components in mineral medicines were checked and are shown in *Table* 4. Heavy metals in mineral medicines were individually analyzed according to the different main components, separately.

*Table* 4. Main components in mineral medicines

| Main compound | Sample name | Chemical formulas of main components |
|---|---|---|
| Si | Tremolitum | $Ca_2(Mg, Fe)_5Si_8O_{22}(OH, F)_2$ |
| | Muscovitum | $KAl_2(AlSi_3)O_{10}(OH, F)_2$ |
| | Fluoritum | $SiO_2$ |
| | Halloysitum Rubrum | $Al_4(HSiO_4)H_2O$, F, Ca, Mg, Mn |
| | Chalcocitum | Silicicate salt |
| | Pumex | $SiO_2$, $Al_2O_3$ |
| Carbonate | Calcitum | $Na_2Ca(SO_4)_2$ |
| | Glauberitum | $CaSO_42H_2O$ |
| | Stalactitum | $CaCO_3$ |
| Hg | Vermilionum | HgS > 98.0% |
| | Cinnabaris | HgS > 96.0% |
| Other | Calamina | $ZnCO_3$ > 40.0% |
| | Lithargyrum | PbO > 95.0% |

In the case of mineral medicines containing carbonate, (Calcitum, Glauberitum and Stalactitum), the acid digestion method was used to mix acid ($HNO_3$ and $H_2SO_4$). The samples containing silicate (Tremolitum, Muscovitum, Fluoritum, Halloysitum Rubrum, Chalcocitum and Pumex) were digested to use a mixed acid ($HNO_3$, HF, and $H_2SO_4$).The samples containing Hg (Vermilionum and Cinnabaris) were digested to use a mixed acid (aqua regia and HF). The prepared samples were analyzed on ICP-MS and AMA for determination of Pb, Cd and As and Hg in mineral medicines, respectively.

2.4.1. Procedure for the analysis of mineral medicines containing carbonate

0.5 g sample was added to a mixed acid of nitric acid (10 mL) and sulfuric acid (1 mL) and allowed to stay overnight at room temperature. Then, it was heated on a hot plate at 150°C for acid digestion. After cooling down, nitric acid (10 mL) was added two or three times. $CH_3COOH$ + $CH_3COONH_3$ (2-3 drops) were added and heated. Next, distilled water (30 mL) was added and heated again until a solution (10 mL) reminded. After cooling down, it was filtered through Whatman filter paper NO.5C into a volumetric flask. The final volume was made up to 25 mL with distilled water. The prepared samples were analyzed on ICP-MS for determination of Pb, Cd and As. For the Hg analysis, 0.01 g of dried samples was put in AMA to use the gold amalgamation method.

2.4.2. Procedure for the analysis of mineral medicines containing Hg

0.5 g of the sample with 20 mL nitric acid and hydrochloric acid (1:3) was transferred into teflon 250 mL beakers and left overnight at room temperature. It was heated on a hot plate at 150°C for acid digestion. And HF (10 mL) was added and heated until a clear solution (2 mL) reminded. Then, distilled water (30 mL) was added and heated again until the volume of the solution reached 10 mL. After cooling down, the solution was filtered through Whatman filter paper NO.5C into a polypropylene (PP) volumetric flask. The final volume was diluted to 25 mL with distilled water. The prepared samples were analyzed on ICP-MS for determination of Pb, Cd and As. We did not analyze Hg in these mineral medicines, because the main compound of these mineral medicines is HgS.

2.4.3. Procedure for the analysis of mineral containing silicate

0.5 g of sample with 5 mL nitric acid and 10 mL hydrofluoric acid (1:2) was transferred into in teflon 250 mL beakers and left overnight at room temperature. It was heated on a hot plate at 150°C for acid digestion. Then, $HNO_3$ (5 mL) and HF (10 mL) were added two or three times and $H_2SO_4$ (3 mL) was added. It was heated again, followed by addition $CH_3COOH$ + $CH_3COONH_3$ (2-3 drops). After then, it was added distilled water (30 mL) and heated again until a solution (10 mL) reminded. After cooling down, it was filtered through Whatman filter paper NO.5C into a polypropylene (PP) volumetric flask. The final volume was increased 25 mL with distilled water. The prepared samples were analyzed on ICP-MS for determination of Pb, Cd and As. For the Hg analysis, 0.01 g of dried samples was analyzed in AMA.

## 3. Results and Discussion

### 3.1. The recovery results of the acid digestion method

Before the monitoring of heavy metals in the mineral medicines, we studied the recovery of the acid digestion method. Recoveries in mineral medicines were achieved by monitoring of standard 200 ppb spiked sample (n=10) using the acid digestion method. Recoveries for Pb, Cd and As were 92.60%, 90.91% and 97.31%, respectively (*Table* 5). Relative

*Table* 5. Recovery of the acid digestion method (n=10)

| Elements | Average | RSD (%) |
|---|---|---|
| Pb | 92.60 | 5.10 |
| Cd | 90.91 | 1.87 |
| As | 97.31 | 3.86 |

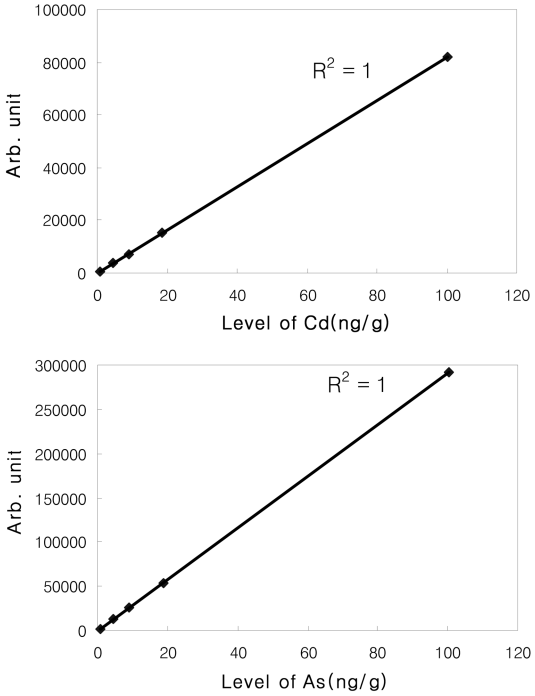


*Fig.* 1. Calibration curves of Pb, Cd and As.

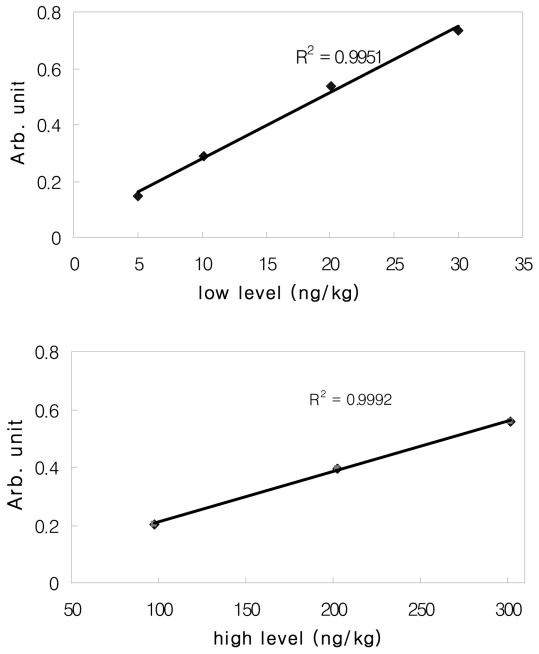


*Fig.* 2. Calibration curves of Hg.

standard deviation (RSD) of Pb, Cd and As were 5.10, 1.87, and 3.86, respectively.

### 3.2. Calibration curves

Calibration curves of Pb, Cd and As are shown in *Fig.* 1. The calibration curve levels of Pb, Cd and As were 1, 5, 10, 20 and 100 ng/kg. Response factors ($R^2$) of Pb, Cd and As were 1 to target heavy metals. In the case of Hg, we created two different calibration curves for low and high levels. The calibration curves for low and high levels of Hg were 5, 10, 20, 30 and 100, 200, 300 ng/kg, respectively. As shown in *Fig.* 2, the response factors

*Table* 6. Average levels of Pb, Cd, As and Hg in mineral medicines

| Name | Pb (mg/kg) | Cd (mg/kg) | As (mg/kg) | Hg (mg/kg) |
|---|---|---|---|---|
| Calamina | 85.5 | 345 | 5.72±3.02 | 11.9 |
| Lithargyrum | N.D[1)] | 0.19 | 22.2±17.0 | 1.05 |
| Tremolitum | 0.75 | 0.16 | 0.20 | 6.12 |
| Vermilionum | 2.43 | 0.02 | 10.0 | N.A[2)] |
| Muscovitum | 4.07 | 0.003 | 0.50 | 3.88 |
| Cinnabaris | 20.4 | 2.78 | 12.5 | N.A[2)] |
| Halloysitum Rubrum | 4.79 | 0.02 | 0.44 | 0.41 |
| Chalcocitum | 0.45 | N.D[1)] | 14.6 | 0.39 |
| Glauberitum | 0.10 | N.D[1)] | 0.31 | 0.41 |
| Pumex | 41.2 | 0.44 | 1.41 | 0.48 |
| Calcitum | N.D[1)] | 0.03 | 0.54 | 0.67 |
| Stalactitum | N.D[1)] | N.D[1)] | 0.23 | 0.04 |
| Fluoritum | 3.24 | 0.03 | 0.14 | 0.93 |

[1)]N.D = Not Detected.
[2)]N.A = Not Analyzed.

*Table* 7. Level ranges of Pb, Cd, As and Hg in mineral medicines

| Name | Pb (mg/kg) | Cd (mg/kg) | As (mg/kg) | Hg (mg/kg) |
|---|---|---|---|---|
| Calamina | 46.8~128 | 250~437 | 3.26~11.5 | 7.40~20.9 |
| Lithargyrum | N.D[1)] | N.D~0.76 | 0.32~41.7 | 0.34~1.80 |
| Tremolitum | 0.46~1.26 | 0.11~0.23 | 0.08~0.40 | 1.15~35.5 |
| Vermilionum | N.D~4.17 | N.D~0.05 | 0.26~17.9 | N.A[2)] |
| Muscovitum | 2.41~8.46 | N.D~0.03 | 0.05~12.3 | 0.38~19.0 |
| Cinnabaris | 20.3~35.0 | 3.52~3.81 | 11.3~19.4 | N.A[2)] |
| Halloysitum Rubrum | 2.28~16.6 | N.D~0.07 | N.D~4.31 | 0.12~2.14 |
| Chalcocitum | 0.55~8.29 | N.D~0.02 | 16.7~22.6 | 0.24~0.72 |
| Glauberitum | N.D~0.99 | N.D[1)] | 0.08~0.60 | 0.19~0.69 |
| Pumex | 35.4~46.1 | 0.29~1.34 | N.D~5.68 | 0.35~1.07 |
| Calcitum | N.D[1)] | N.D~0.15 | 0.05~1.05 | 0.04~4.93 |
| Stalactitum | N.D[1)] | N.D[1)] | 0.14~0.33 | 0.04 |
| Fluoritum | 1.54~4.80 | 0.02~0.06 | 0.02~0.30 | 0.31~2.30 |

[1)]N.D = Not Detected.
[2)]N.A = Not Analyzed.

($R^2$) of Hg were 0.9951 and 0.9992 for low and high levels, respectively.

### 3.3. The monitoring results of Pb, Cd, As and Hg in mineral medicines

A comparison of the average levels and level ranges of Pb, Cd, As and Hg in 13 different species of 92 samples is summarized in *Tables* 6 and 7.

Korea food and drug agency (KFDA) has regulated the Maximum Residue Limits (MRL) of Pb, Cd, As and Hg to be 5 mg/kg, 0.3 mg/kg, 3 mg/kg, and 0.2 mg/kg, respectively, in foods and drugs.

#### 3.3.1. Lead

The results showed that 25.81% (24 cases) of the samples exceeded 5 mg/kg of the Maximum Residue Limits (MRL) in foods and drugs set by KFDA. Pb level range was N.D to 127 mg/kg. The levels of Pb in Calamina (85.5 mg/kg), Pumex (41.2 mg/kg) and Cinnabaris (20.4 mg/kg) exceeded MRL (5 mg/kg). In Muscovite (4.79 mg/kg), 4 cases of 10 samples exceeded MRL (5 mg/kg). Therefore, they need to be monitored consistently. Lead was not detected in Lithargyrum, Calcitum and Stalactitum.

#### 3.3.2. Cadmium

The monitoring results showed that 18.28% (17 cases) of samples exceeded the MRL (0.3 mg/kg) in foods and drugs set by KFDA. The Cd level ranged from N.D to 437 mg/kg. The Cd levels in Calamina (345 mg/kg), Pumex (0.44±0.41 mg/kg) and Cinnabaris (2.78 mg/kg) exceeded MRL (0.3 mg/kg). Calamina contained the highest Cd level. Cd levels in Lithargyrum (0.19 mg/kg), Tremolitum (0.16 mg/kg), Muscovitum (0.003 mg/kg), Halloysitum Rubrum (0.02 mg/kg), Pumex (0.44 mg/kg), Calcitum (0.03 mg/kg) and Fluoritum (0.03 mg/kg) were within the MRL (0.2 mg/kg) by KFDA. Chalcocitum, Glauberitum and Stalacitum were not detected in Cd.

#### 3.3.3. Arsenic

The monitoring results indicated that 24.73% (23 cases) of the samples exceeded the MRL (3 mg/kg) in foods and drugs set by KFDA.

The As level ranged from N.D to 22.6 mg/kg. The As levels in Calamina (5.72 mg/kg), Lithargyrum (22.2 mg/kg), Vermilionum (10.0 mg/kg), Cinnabaris (12.5 mg/kg), and Chalcocitum (14.6 mg/kg) exceeded MRL (3 mg/kg). As levels in Tremolitum (0.20±0.13 mg/kg), Muscovitum (0.50 mg/kg), Halloysitum Rubrum (0.44±1.38 mg/kg), Glauberitum (0.31 mg/kg), Pumex (1.41 mg/kg), Calcitum (0.54 mg/kg), Stalacitum (0.23 mg/kg) and Fluoritum (0.14 mg/kg) were within the MRL (3.0 mg/kg).

#### 3.3.4. Mercury

Our test results showed that 76.34% (78cases) of samples exceeded the MRL (0.2 mg/kg) set by KFDA. The Hg level ranged from 0.04 to 35.5 mg/kg. The Hg levels in Calamina (11.9 mg/kg), Lithargyrum (1.05 mg/kg), Tremolitum (6.12 mg/kg), Muscovitum (3.88 mg/kg), Halloysitum Rubrum (0.41 mg/kg), Chalcocitum (0.39 mg/kg), Glauberitum (0.41 mg/kg), Pumex (0.48 mg/kg), Calcitum (0.67 mg/kg) and Fluoritum (0.93 mg/kg) exceeded the MRL (0.2 mg/kg), which were found in almost all the monitored mineral medicines. Only one sample, Stalactitum (0.04 mg/kg), was within the MRL (0.2 mg/kg).

## 4. Conclusions

Ninety-two 92 samples collected from 13 different species of mineral medicines were monitored by the acid digestion method to obtain levels of Pb, Cd, As and Hg. The results showed that, in the case of Pb, 25.81% (24 cases) of samples exceeded MRL (5 mg/kg). In the cases of Cd and As, 18.28% (17 cases) and 24.73% (23 cases) of samples exceeded the MRL (0.3 mg/kg and 3 mg/kg, respectively). In the case of Hg, 76.34% (78 cases) of the samples exceeded the MRL (0.2 mg/kg). Almost all the mineral medicines contained high level of heavy metals that exceeded the MRL.

These results led us to conclude that the domestic commercial law of oriental mineral medicines needs to be legislated to ensure the quality estimation. Therefore, we need continuously monitoring to ensure confidence and safety for these mineral medicines. And these results are used for the establishment of policy for regulations and controls to these oriental mineral medicines to be based of hazardous indexes and proposed safety standards.

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