# Coal Mine Environment Causes Adverse Effects on Workers

**Huihui Chen**
North China Institute of Science and Technology
**Mengli Zhang**
North China Institute of Science and Technology
**Xinping Ding**
Huaibei Occupational Disease Prevention and Control Institute
**Tianwei Wang**
Weifang Medical University
**Zhiyuan Ma**
Weifang Medical University
**Wenzhong Zhang** ( zhangwz2002@sina.com )
North China Institute of Science and Technology

**Article**

**Keywords:** Coal mine environment, Immune system, Renal function, Apoptosis, Anemia

**Posted Date:** May 10th, 2022

**DOI:** https://doi.org/10.21203/rs.3.rs-1619434/v1

# Abstract

**Purpose:** To study the health effects of coal mine environment on workers, to discover early effective biomarkers.

**Methods:** A cross-sectional survey was conducted with 502 in-service workers, and the population was divided into the coal miner and the auxiliary worker. Clinical examinations were carried out by qualified doctors. Peripheral blood was collected to measure blood biochemistry, hemogram and karyotype apoptosis.

**Results:** All workers were healthy and showed no difference in age, height, weight and BMI between groups. The education level and working years of coal miners were significantly lower than that of auxiliary workers ($P < 0.05$). Compared with auxiliary workers, the concentration and percentage of LYMPH, BASO, EO, RBC and the concentration of HGB of coal miners decreased ($P < 0.05$). The percentage of NEUT of coal miners increased ($P < 0.05$). The coal miner presented higher BUN, lower CRE, and higher BUN/CRE ($P < 0.05$). The proportion of karyotype apoptosis of coal miners was higher ($P < 0.05$).

**Conclusions:** The coal mine environment impacts immune system, induces adverse effects on renal function and enhances the risk of anemia of coal miners. And karyotype apoptosis, LYMPH, BUN/CRE and HGB can be used as early effective biomarkers for coal miners.


# Introduction

In coal mining activities, many occupational hazard factors including coal dust, exhaust gas, high temperature, high humidity, noise, etc., form coal mine environment [1]. It is well known that coal dust exposure leads to pneumoconiosis, which is the priority healthy threat for coal miners [2]. Coal dusts are supervised by government, their concentration is controlled under maximum limit standard ensured no harmful effects. However, many kinds of hazard factors in coal mine environment may generate combination effects which have not been paid enough attention. Such as, coal dust also contains a variety of heavy metals, which pollute the environment during coal mining and transportation [3]. Heavy metals accumulate in multiple organs and tissues both in human and animal [4]. Among them, the kidney is the main target organ for heavy metals accumulation [4]. Batool et al. [5] report that coal dust can cause inflammatory and oxidative damage in coal miners. Oxidative damage contributes to DNA damage, which is an important factor for onset of cancer and chronic diseases [6]. Except heavy metal elements, harmful factors such as exhausted gas formed with multiple carcinogen cause oxidative damage and other health problems [7]. At present, there is no specific effective biomarker used to screen sub-healthy coal miners in coal mine environment.

Thus, the present study aimed to discover the influences of the coal mine environment and to find specific early effective biomarker in traditional healthy coal miners.

# Results

## General information

All workers were male and passed normal clinical physical examinations, found no diseases ($P > 0.05$), there were no differences in age, height, weight and body mass index (BMI) between the groups ($P > 0.05$). The education level of coal miners was significantly lower than that of auxiliary workers ($P < 0.05$). The coal miner's diastolic pressure and pulse frequency were lower than that of auxiliary workers, all parameters were in normal range (Table 1).

Table 1
General information results in two groups (Mean ± SD)

| Parameters | auxiliary worker | coal miner |
|---|---|---|
| Age (year) | 42.63 ± 8.83 | 43.01 ± 6.74 |
| Level of education | 1.56 ± 0.50 | 1.19 ± 0.40* |
| Length of work (year) | 10.93 ± 7.29 | 8.16 ± 7.28* |
| Body mass index (kg/m$^2$) | 24.43 ± 2.90 | 24.16 ± 3.94 |
| Systolic pressure (mmHg) | 130.70 ± 16.02 | 130.61 ± 14.01 |
| Diastolic pressure (mmHg) | 81.65 ± 11.71 | 79.28 ± 10.47* |
| Pulse pressure difference (mmHg) | 46.78 ± 14.31 | 50.84 ± 10.51 |
| Pulse frequency (times/minute) | 79.09 ± 11.36 | 76.96 ± 10.96* |
| Height (cm) | 171.25 ± 5.23 | 171.01 ± 5.11 |
| Weight (kg) | 71.76 ± 9.90 | 70.78 ± 13.04 |
| Note. Auxiliary worker = 279, Coal miner = 223, * Compared with auxiliary worker $P < 0.05$ | | |

## Hemogram

Compared with auxiliary workers, the concentration and percentage of lymphocytes (LYMPH), basophils (BASO), eosinophils (EO), red blood cells (RBC) and the concentration of hemoglobin (HGB) of coal miners decreased ($P < 0.05$). The concentration of neutrophils (NEUT) elevated without statistical difference ($P > 0.05$), however, the percentage of NEUT increased significantly ($P < 0.05$). Parameters of white blood cells (WBC), mean corpuscular volume (MCV), platelet (PLT), mean platelet volume (MPV), platelet hematocrit (PCT), mean corpuscular hemoglobin (MCH) and monocyte macrophage (MONO) presented no difference between two groups ($P > 0.05$) (Table 2).

Table 2
General information results in two groups (Mean ± SD)

| Parameters | auxiliary worker | coal miner |
|---|---|---|
| Age (year) | 42.63 ± 8.83 | 43.01 ± 6.74 |
| Level of education | 1.56 ± 0.50 | 1.19 ± 0.40* |
| Length of work (year) | 10.93 ± 7.29 | 8.16 ± 7.28* |
| Body mass index (kg/m$^2$) | 24.43 ± 2.90 | 24.16 ± 3.94 |
| Systolic pressure (mmHg) | 130.70 ± 16.02 | 130.61 ± 14.01 |
| Diastolic pressure (mmHg) | 81.65 ± 11.71 | 79.28 ± 10.47* |
| Pulse pressure difference (mmHg) | 46.78 ± 14.31 | 50.84 ± 10.51 |
| Pulse frequency (times/minute) | 79.09 ± 11.36 | 76.96 ± 10.96* |
| Height (cm) | 171.25 ± 5.23 | 171.01 ± 5.11 |
| Weight (kg) | 71.76 ± 9.90 | 70.78 ± 13.04 |
| Note. Auxiliary worker = 279, Coal miner = 223, * Compared with auxiliary worker $P < 0.05$ | | |

# Blood Biochemistry Parameters

Compared with auxiliary workers, blood urea nitrogen (BUN) and ratio of blood urea nitrogen to creatinine (BUN/CRE) increased, creatinine (CRE) decreased in coal miners ($P < 0.05$) (Fig. 1). Parameters of alanine aminotransferase (ALT), aspartate aminotransferase (AST), albumin (ALB), total bilirubin (TBIL), alkaline phosphatase (ALP), fasting blood glucose (GLU), cholesterol (CHO), triglyceride (TG), high density lipoprotein cholesterol (HDL-C) and low density lipoprotein cholesterol (LDL-C) presented no difference between two groups ($P > 0.05$) (Table 3).

Table 3
Renal function indicators results in two groups (Mean ± SD)

| Parameters | auxiliary worker | coal miner |
|---|---|---|
| ALT (U/L) | 29.63 ± 18.92 | 26.36 ± 18.14 |
| AST (U/L) | 24.69 ± 14.16 | 24.30 ± 9.95 |
| ALB (g/L) | 45.83 ± 2.20 | 45.67 ± 2.32 |
| TBIL (umol/L) | 12.61 ± 5.28 | 11.88 ± 5.18 |
| ALP (U/L) | 86.38 ± 20.10 | 85.87 ± 18.22 |
| GLU (mmol/L) | 5.25 ± 1.03 | 5.64 ± 3.09 |
| CHO (mmol/L) | 3.97 ± 1.69 | 3.28 ± 1.87 |
| TG (mmol/L) | 1.56 ± 1.24 | 1.59 ± 1.52 |
| HDL-C (mmol/L) | 1.59 ± 0.66 | 1.79 ± 0.70 |
| LDL-C (mmol/L) | 2.62 ± 0.61 | 2.56 ± 0.68 |
| Note. Auxiliary worker = 279, Coal miner = 223, *Compared with auxiliary worker $P < 0.05$ | | |

# Karyocyte Apoptosis In Peripheral Blood

Compared with auxiliary workers, the percentage of apoptotic cells in peripheral blood increased significantly in the coal miner ($P < 0.05$) (Fig. 2).

# Regression Analysis Of Risk Factors

A multiple stepwise regression was developed and independent variables were used to determine the risk indicators of the coal mine environment to the coal miner (Table 4). The karyocyte apoptosis variable explained 36.0% ($P < 0.001$) of the risk index, while the absolute value of LYMPH explained 34.0% ($P < 0.001$), the BUN/CRE variable explained 2.0% ($P < 0.001$) and the HGB variable explained 0.4% ($P < 0.001$) of the risk index in the coal mine environment. The regression model created for the variables were explained 72% by the 4 parameters.

Table 4
Multiple regression calculations

| Dependent variable | n | Independent variables | r | $R^2$ | $R^2$ variation | F value | *P* |
|---|---|---|---|---|---|---|---|
| Work Type (auxiliary worker = 1, coal miner = 2) | 502 | Apoptosis cell ratio | 0.361 | 0.130 | 0.129 | 75.842 | 0.000 |
| | 502 | Absolute value of lymphocytes | 0.706 | 0.498 | 0.496 | 250.583 | 0.000 |
| | 502 | Ratio CRE/BUN | 0.721 | 0.519 | 0.516 | 181.519 | 0.000 |
| | 502 | Hemoglobin | 0.725 | 0.526 | 0.522 | 139.353 | 0.000 |

# Discussion

A 10-year cohort study conducted in Henan province of China, with 12,000 male coal miners as the research object, showed that the risk of hypertension in coal miners related with the level of coal dust exposure [8]. Additionally, epidemiological study has found that the prevalence of cardiovascular diseases in coal miners is high and gradually increasing, in which obesity is the main risk factor [9]. The present study showed no obesity and cardiovascular diseases in coal miners, even more coal miners' diastolic pressure and heart rate were better than that of auxiliary workers, which suggested physical work benefiting cardiac function [10]. Also, studies have shown that the poor living habits and stressful working environment of coal miners can easily lead to dyslipidemia [11, 12]. Additionally, excessive caloric intake and insufficient exercise result in abnormal blood lipid metabolism [10]. The present study showed no abnormal lipid metabolism. Our results indicate that the coal mine environment selected in present study has no adverse effects on cardiovascular system and lipid metabolism.

Bhuiyan et al. [13] report that the water nearby the coal mine is polluted with heavy metals. Heavy metals mainly accumulate in the kidney of fish living in water polluted by copper, lead, chromium and cadmium [4]. The results are confirmed with a population study, which shows that the risk of kidney disease is higher for people who living in coal mining areas [14]. Additionally, studies have found that exposure to silica dust increases the risk of chronic kidney disease, and there is a direct relationship between air pollution and renal disease [15, 16]. BUN and BUN/CRE are effective biomarkers for renal function, and their increasing associated with renal impairment [17, 18]. BUN and BUN/CRE of coal miners increasing in present study, suggested that the coal mine environment induced adverse effects on renal function. Except causing renal function damage, air pollution also correlated with prevalence of anemia, characterized with a reduction of RBC and HGB [19, 20]. RBC and HGB are biomarkers of anemia, the present study showed lower RBC and HGB of coal miners, which indicated that the coal mine environment may enhance the risk of anemia.

As the main indicator of the immunity function, LYMPH reducing means immune functional impairment [21]. In addition, EO and BASO are involved in innate and adaptive immune regulation, contributing to homeostasis in the body [22, 23]. In the present study, LYMPH, BASO and EO of coal miners decreased, which suggested that the coal mine environment may impair immune system. When immune function is weak, it is susceptible to infection. Epidemic study and animal experiment show that coal dusts can induce inflammatory response in lung and cardiovascular system [9, 24]. Except coal dusts, exhausted gas also induces inflammatory response [25]. NEUT, as the main immune cells, increase significantly in inflammatory status [26]. Higher NEUT of coal miners in the present study indicated that the coal mine environment induced inflammatory.

Oxidative stress induced by coal dusts shows time dependent increasing in coal miners, furthermore it can damage DNA [5, 27]. In addition to oxidative stress, DNA damage can also be induced by water-soluble heavy metals including copper, lead, chromium and cadmium [28, 29]. Studies have demonstrated that DNA damage results in karyocyte apoptosis [30, 31]. Besides DNA damage, many factors including inflammatory cytokines, coal dusts and high temperature also can induce apoptosis [32, 33, 34]. Additionally, karyocyte apoptosis contributes to a reduction of LYMPH in population [32]. The present study showed that apoptotic cells elevated and LYMPH decreased in peripheral blood of coal miners, which indicates that LYMPH reducing may attribute to apoptosis.

In the present study, regression analysis reveals four effective biomarkers sequenced with karyocyte apoptosis, LYMPH, BUN/CRE and HGB that are sensitive to coal mine environment.

## Conclusions

In summary, the coal mine environment impacts immune system, induces adverse effects on renal function and enhances the risk of anemia of coal miners. Therefore, karyocyte apoptosis, LYMPH, BUN/CRE and HGB can be used as early effective biomarkers to screen sub-healthy coal miners. The present study based on a cross-sectional survey focusing upon one region cannot represent the whole country, it is necessary to carry out cohorts and more wide regional study to verify coal mine environment impactions and biomarkers.

## Methods

# Cross-sectional study

The study was approved by the Ethics Committee of Huaibei Occupational Disease Prevention and Control Institute (approval number: 20180603). The study confirmed that all experiments were performed in accordance with relevant guidelines and regulations and informed consent was obtained from all participants.Five hundred and two in-service coal miners recruited from Huaibei Mining Group, to carry out a molecular epidemiological study. Face-to-face survey conducted by trained investigators using a

unified questionnaire. The general conditions including eating habits, history of smoking, drinking, family and occupation, etc., were collected. In the present study, we recognized the tunneling workers and drilling workers as coal miners, and the other types of workers as auxiliary workers.

# Clinical Examination

Physical examinations for workers including medical examination, surgical examination, respiratory function, imaging examination (including electrocardiogram, chest X-ray and abdominal ultrasound) was carried out by qualified doctors.

# Hematology And Blood Biochemistry

Based on the informed consent of the study subjects, 5 ml of peripheral venous blood was collected for the determination of biomarkers. Hematological parameters of whole blood were measured with an automatic hemocytometer (Nihon Kohden MEK-6813K). Blood biochemistry parameters were analyzed with an automated biochemical analyzer (AU680, Beckman Coulter Ltd.).

# Apoptosis Or Necrosis Analysis By Alkaline Comet Assay

Karyocyte apoptosis or necrosis (referred to as apoptosis) was analyzed with the alkaline comet assay. Cells with tail DNA percent more than 95% were considered as apoptosis and then the apoptosis rate was determined. Alkaline comet assay kit was purchased from Huaxing Innovation Biotechnology Co., Ltd, China (HIB). Comets were visualized using a light microscope with 510 excitation and 590 emission filters. The images were analyzed using the software Comet A1.0 created by HIB.

# Statistical analysis

All parameters are expressed as the mean ± standard deviation (SD). Homogeneity of variance was examined by Levene's test. If Levene's test demonstrated no significant deviations, group means were compared by one-way analysis of variance (ANOVA) followed by post hoc Fisher's Least Significant Difference tests for pairwise comparisons. A multiple stepwise regression was performed, all variables were entered into the model initially, with the least significant variables associated with values of $P \leq 0.05$ remained. All statistical tests were two-tailed. P value less than 0.05 is as the significant difference.

# Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

# Declarations


### Acknowledgements

The authors would like to acknowledge Huaibei Occupational Disease Prevention and Control Institute for their help in completing this study. We thank International Science Editing (http://www.internationalscienceediting.com) for editing this manuscript.


### Author Contributions

Wenzhong Zhang designed study. Wenzhong Zhang, Huihui Chen, Mengli Zhang, Xinping Ding, Tianwei Wang and Zhiyuan Ma performed the study. Wenzhong Zhang and Huihui Chen analyzed the data, interpreted the data. All authors approved of the final manuscript.


### Funding

This work was supported by Funding for Basic Scientific Research Operation of Central Universities (3142019002).


### Competing interests

The authors declare no competing interests.



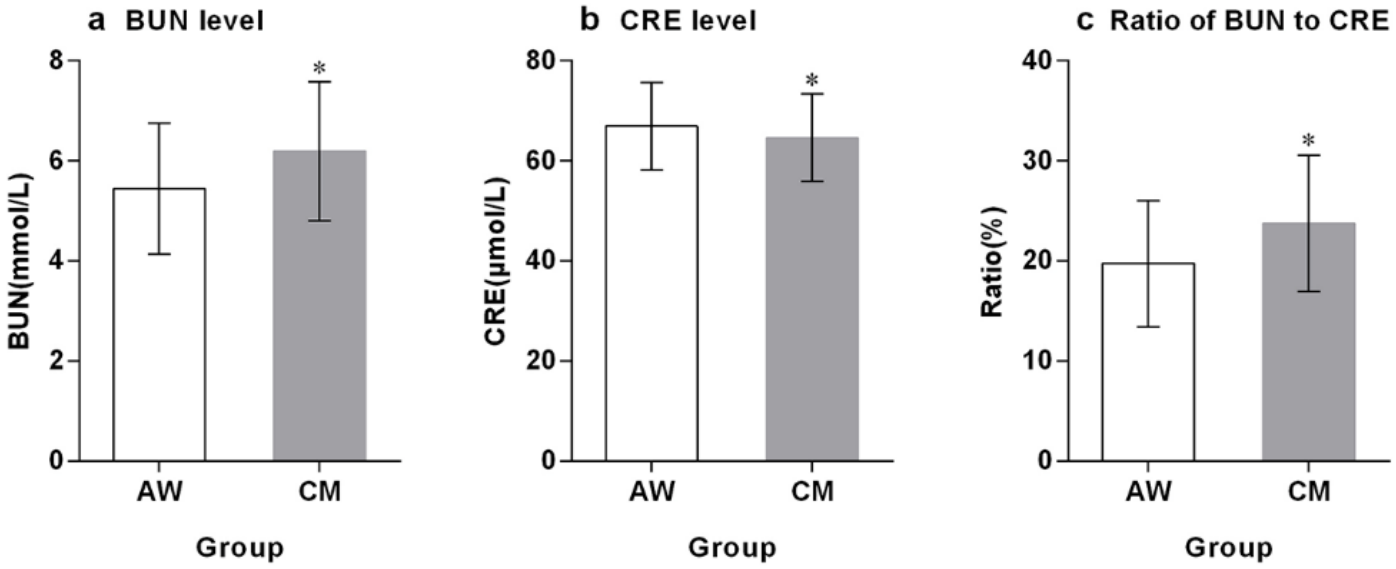
# References


1. Strzemecka, J., M. Goździewska, J. Skrodziuk, E.M. Galińska and S. Lachowski Factors of work environment hazardous for health in opinions of employees working underground in the 'Bogdanka' coal mine. Ann Agric Environ Med **26**, 409–414 (2019).
2. Hall, N.B., D.J. Blackley, C.N. Halldin and A.S. Laney Current Review of Pneumoconiosis Among US Coal Miners. Curr Environ Health Rep **6**, 137–147 (2019).
3. Zhang, H., et al. Pollutant source, ecological and human health risks assessment of heavy metals in soils from coal mining areas in Xinjiang, China. Environ Res **202**, 111702 (2021).
4. Lakra, K.C., B. Lal and T.K. Banerjee Coal mine effluent-led bioaccumulation of heavy metals and histopathological changes in some tissues of the catfish Clarias batrachus. Environ Monit Assess **191**, 136 (2019).
5. Batool, A.I., N.H. Naveed, M. Aslam, J. da Silva and M.F.U. Rehman Coal Dust-Induced Systematic Hypoxia and Redox Imbalance among Coal Mine Workers. ACS Omega **5**, 28204–28211 (2020).

6. Eckhardt, C.M. and H. Wu Environmental Exposures and Lung Aging: Molecular Mechanisms and Implications for Improving Respiratory Health. Curr Environ Health Rep **8**, 281–293 (2021).
7. Usemann, J., et al. Gasoline particle filter reduces oxidative DNA damage in bronchial epithelial cells after whole gasoline exhaust exposure in vitro. Sci Rep **8**, 2297 (2018).
8. Zhi, C.X., et al. [Association between dust exposure and the risk of hypertension of male coal miners in Henan Province]. Zhonghua Yu Fang Yi Xue Za Zhi **53**, 597–602 (2019).
9. Casey, M.L., et al. Evaluation of high blood pressure and obesity among US coal miners participating in the Enhanced Coal Workers' Health Surveillance Program. J Am Soc Hypertens **11**, 541–545 (2017).
10. Seravalle, G. and G. Grassi Obesity and hypertension. Pharmacol Res **122**, 1–7 (2017).
11. Fan, Y., et al. Prevalence of dyslipidaemia and risk factors in Chinese coal miners: a cross-sectional survey study. Lipids Health Dis **16**, 161 (2017).
12. Fu, Z.D., et al. [Correlative factors on prevalence rate of dislipidemia among 1 337 coal miners in Shanxi province]. Zhonghua Liu Xing Bing Xue Za Zhi **38**, 163–167 (2017).
13. Bhuiyan, M.A.H., M. Bodrud-Doza, M.A. Rakib, B.B. Saha and S.M.D. Islam Appraisal of pollution scenario, sources and public health risk of harmful metals in mine water of Barapukuria coal mine industry in Bangladesh. Environ Sci Pollut Res Int **28**, 22105–22122 (2021).
14. Hendryx, M. Mortality from heart, respiratory, and kidney disease in coal mining areas of Appalachia. Int Arch Occup Environ Health **82**, 243–9 (2009).
15. Möhner, M. An approach to adjust standardized mortality ratios for competing cause of death in cohort studies. Int Arch Occup Environ Health **89**, 593–8 (2016).
16. Afsar, B., et al. Air pollution and kidney disease: review of current evidence. Clin Kidney J **12**, 19–32 (2019).
17. Hoseinynejad, K., M. Radan, M. Dianat and F. Nejaddehbashi Adipose-derived mesenchymal stem cells protects renal function in a rat model of emphysema. Tissue Cell **73**, 101613 (2021).
18. Shen, S., X. Yan and B. Xu The blood urea nitrogen/creatinine (BUN/cre) ratio was U-shaped associated with all-cause mortality in general population. Ren Fail **44**, 184–190 (2022).
19. Diab, H.M., et al. Coexistence of diverse heavy metal pollution magnitudes: Health risk assessment of affected cattle and human population in some rural regions, Qena, Egypt. J Adv Vet Anim Res **7**, 345–359 (2020).
20. Elbarbary, M., et al. Ambient Air Pollution Exposure Association with Anaemia Prevalence and Haemoglobin Levels in Chinese Older Adults. Int J Environ Res Public Health **17**, (2020).
21. Chen, X., S. Yuan and J. Zhang Correlation study between blood cytokines and lymphocytes in early postoperative critical patients with compromised immune function. Medicine (Baltimore) **99**, e22459 (2020).
22. Arock, M. [The basophil: From control of immunity to control of leukemias]. Ann Pharm Fr **80**, 9–25 (2022).

23. Magrone, T., M. Magrone and E. Jirillo Eosinophils, a Jack of All Trades in Immunity: Therapeutic Approaches for Correcting Their Functional Disorders. Endocr Metab Immune Disord Drug Targets **20**, 1166–1181 (2020).
24. Sunil, V.R., et al. World Trade Center (WTC) dust exposure in mice is associated with inflammation, oxidative stress and epigenetic changes in the lung. Exp Mol Pathol **102**, 50–58 (2017).
25. Tang, M., et al. Seasonal and areal variability in PM(2.5) poses differential degranulation and pro-inflammatory effects on RBL-2H3 cells. Chemosphere **279**, 130919 (2021).
26. Bongers, S.H., et al. Kinetics of Neutrophil Subsets in Acute, Subacute, and Chronic Inflammation. Front Immunol **12**, 674079 (2021).
27. Peters, A., T.S. Nawrot and A.A. Baccarelli Hallmarks of environmental insults. Cell **184**, 1455–1468 (2021).
28. Feng, X., et al. Oxidative potential and water-soluble heavy metals of size-segregated airborne particles in haze and non-haze episodes: Impact of the "Comprehensive Action Plan" in China. Sci Total Environ **814**, 152774 (2022).
29. Feng, X., et al. Particle-induced oxidative damage by indoor size-segregated particulate matter from coal-burning homes in the Xuanwei lung cancer epidemic area, Yunnan Province, China. Chemosphere **256**, 127058 (2020).
30. Li, C., et al. DNA damage-triggered activation of cGAS-STING pathway induces apoptosis in human keratinocyte HaCaT cells. Mol Immunol **131**, 180–190 (2021).
31. Liu, P., et al. TRESK Regulates Gm11874 to Induce Apoptosis of Spinal Cord Neurons via ATP5i Mediated Oxidative Stress and DNA Damage. Neurochem Res **46**, 1970–1980 (2021).
32. Cizmecioglu, A., et al. Apoptosis-induced T-cell lymphopenia is related to COVID-19 severity. J Med Virol **93**, 2867–2874 (2021).
33. Mu, M., et al. Coal dust exposure triggers heterogeneity of transcriptional profiles in mouse pneumoconiosis and Vitamin D remedies. Part Fibre Toxicol **19**, 7 (2022).
34. Rahman, M.S. and M.S. Rahman Elevated seasonal temperature disrupts prooxidant-antioxidant homeostasis and promotes cellular apoptosis in the American oyster, Crassostrea virginica, in the Gulf of Mexico: a field study. Cell Stress Chaperones **26**, 917–936 (2021).

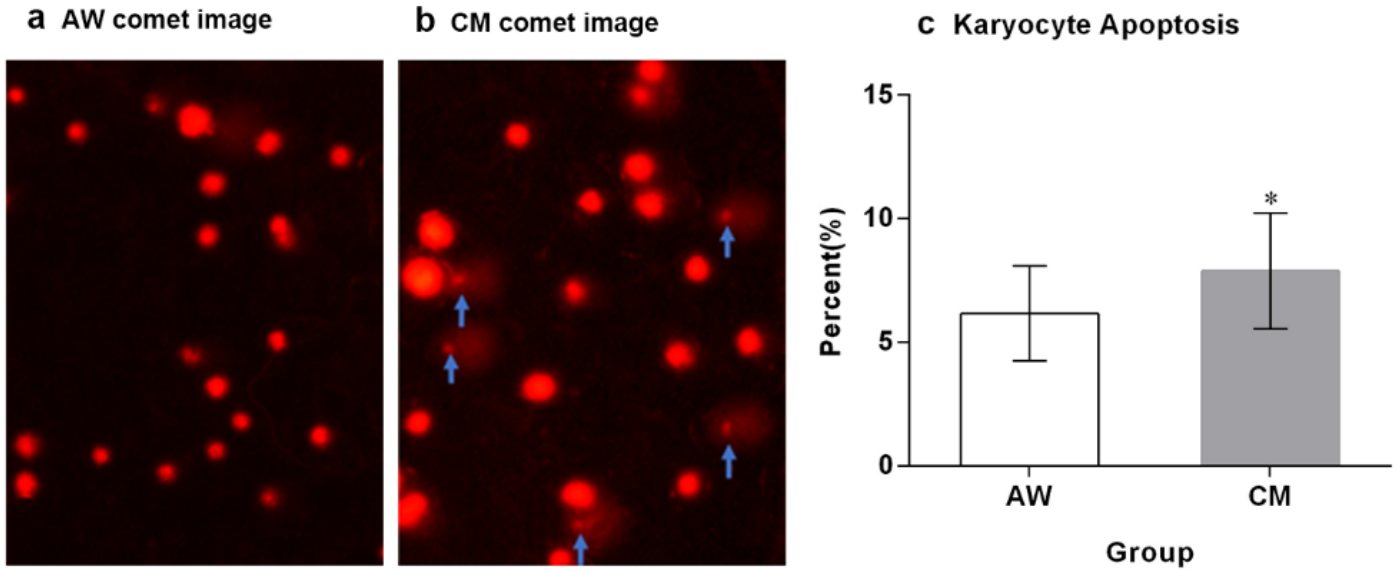
# Figures




## Figure 1

Renal function indexes of coal miner and auxiliary worker

***Note.*** Auxiliary worker (AW) = 279, Coal miner (CM) = 223, *Compared with auxiliary worker $P < 0.05$




## Figure 2

Apoptotic cells in peripheral blood of coal miner and auxiliary worker

***Note.*** Auxiliary worker (AW) = 279, Coal miner (CM) = 223, *Compared with auxiliary worker $P < 0.05$, the apoptotic cells marked with the arrow