ORIGINAL ARTICLE

THE EFFECTS OF SUBCHRONICAL EXPOSURE TO SO_2 ON BIOCHEMICAL AND HEMATOLOGICAL PARAMETERS IN GUINEA PIGS

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Summary: The effects of subchronical exposure to SO_2 (400ppm, 3 hours daily, 28 days) on biochemical and hematological parameters were investigated in guinea pigs. Mostly no significant changes in the values of biochemical parameters and no significant changes in hematological parameters were found. The levels of investigated ions (K^+, Na^+, Cl^-, Ca^{++}, Mg^{++} and phosphates), proteins (albumines, globulines, total proteins), enzymes (LD, ALT, AST, CK) and other biochemical parameters (urea, creatinine, bilirubin) were not significantly different between groups, with the exception of a significantly higher ALP concentration in the exposed group as compared with controls (2,17 μkat and 1,85 μkat, respectively). It can be concluded that a subchronical exposure to sulphur dioxide mostly did not induce any definite changes in biochemical and hematological parameters in guinea pigs.

Key words: *Sulphur dioxide; Subchronical exposure; Biochemical parameters; Hematological parameters*

Introduction

Environmental pollution is one of the most overwhelming contemporary problems and sulphur dioxide is one of the most common atmospheric pollutants in the Middle Europe region. Sulphur dioxide and various particles are emitted into the atmosphere by the burning of fossil fuels and smelting of metals. Many of these particles can promote the conversion of SO_2 to the more irritant sulphuric acid (1,10). Inversion, fog, and cold temperatures also support the negative consequences of exposure to SO_2 (2).

The influence of exposure to SO_2 on the respiratory tract has been demonstrated in various studies (4, 11, 13). Sulphur dioxide is a soluble gas which is readily absorbed in the nose and upper respiratory tract. SO_2 dissolves in the fluid lining the airway with the production of sulphite and bisulphite ions. These may react with low molecular weight disulphide groups in proteins. Excretion of sulphur absorbed as SO_2 occurs via sulphate, sulphate is produced by the conversion of sulphite being catalyzed by oxidative enzymes (5). Until now, only limited informations about the effects of SO_2 on the other body systems are available (genotoxical effects of SO_2 and increase of lung cancer mortality have been described - 6, 14). The aim of the study was to investigate the effects of the subchronical exposure to SO_2 on the biochemical and hematological parameters in guinea pigs.

Materials and methods

Male guinea pigs (BFA) with an average weight of 500g were used. The standard laboratory conditions were respected. The handling of experimental animals was under the supervision of the Ethics Committee of the Medical faculty, Charles University, Hradec Králové.

Two groups of animals were used. The experimental group (n=12) of male guinea pigs with an average weight of

500g was subchronically exposed to SO_2 (400ppm, 3 hours daily, 5 days in a week) for 28 days. Exposure to SO_2 was realized in special chambers, in each one 8 animals could be exposed together. The control group (n=11) was „sham" exposed to the air in the same time.

Exposures were conducted in 0,278 m^{-3} stainless steel and glass exposure chambers designed by Drew (3). The supply air was filtred and controlled for temperature and humidity, and flow rate 100 L/min was established by the main exhaust pump. Sulphur dioxide for calibration (Linde Technoplyn) was used for exposures. Nominal and analytical concentrations were determined daily. Analytical concentrations of sulphur dioxide were determined by spectrophotometric method described in unificated methodologies for determination of harmful compounds in air (7).

The noninvasive polygraphic cardiac recordings of systolic time intervals were measured using a polygraph Biomedica C6b (Italy) in ketamine anaesthesia (150 mg/kg i.p., Narkamon 5%, Léčiva, Czech Republic) at the beginning of the experiment (7 days before the exposure) and then weekly in the 7th, 14th, 21st and 28th day of the exposure (3 hours after the end of the exposure to SO_2 or after the „sham" exposure). Index PEP:LVET was calculated on the basis of simultaneous recordings of the electrocardiogram, phonocardiogram and carotid pulse waveforms (these results are not demonstrated in this paper).

At the end of the experiment (i.e., after the last polygraphic measurement, on the day 28 of the exposure), anesthesia was enhanced by urethan (20% solution, 0,65 ml/100g i.p.) and blood samples were taken from *v. cava caudalis* for biochemical and hematological analysis. After the sacrifice of the animal, the tissues (heart, muscle, lungs, liver) were taken for histological examination. Biochemical parameters (in plasma/serum) were determined with standard biochemical methods using an automatic analyzer Hitachi 717, Japan. Hematological parameters were determined using analyzer Coulter T890, U.S.A.

Statistical evaluation of values was performed using an unpaired t-test (comparison of different groups) for the level of significance ($p \leq 0.05$). Values are expressed as mean ± S.E.M.

Results

Biochemical parameters:

Almost no differences in biochemical parameters (Tab. 1) between the SO_2 exposed and control group were found, although in most of them the tendency towards an elevation could be observed in the former group. The levels of investigated ions, proteins, enzymes and other biochemical parameters were not significantly different, with the exception of a significantly higher ALP concentration in the exposed group as compared with controls (2,17 μkat and 1,85 μkat, respectively).

Table 1: Biochemical parameters

Parameter	SO_2	Sham
glucose (mmol/l)	9,31±0,37	8,13±0,79
sodium (mmol/l)	136,6±21,04	134,5±60,69
potassium (mmol/l)	9,65±0,84	9,13±1,00
chloride (mmol/l)	104,77±0,76	102,89±1,02
calcium (mmol/l)	2,53±0,05	2,40±0,03
magnesium (mmol/l)	1,36±0,06	1,35±0,03
phosphate (mmol/l)	2,77±0,15	2,44±0,13
urea (mmol/l)	13,07±0,67	12,06±0,56
creatinine (μmol/l)	54,15±2,41	55,67±2,58
uric acid (μmol/l)	108,31±7,38	99,33±7,76
bilirubin (μmol/l)	0,00±0,00	0,00±0,00
LD (μkat/l)	6,31±1,09	7,53±1,07
ALT (μkat/l)	1,22±0,08	1,11±0,10
AST (μkat/l)	2,31±0,32	2,32±0,46
CK (μkat/l)	32,68±11,38	26,96±8,82
ALP (μkat/l)	2,17±0,10*	1,85±0,09
cholesterol (mmol/l)	1,01±0,073	0,99±0,09
triglycerides (mmol/l)	0,95±0,22	0,75±0,05
protein (g/l)	46,41±1,05	45,86±0,86
albumin (%)	30,92±0,63	31,13±0,52
ealb	0,59±0,01	0,59±0,01
α1 globulin	0,03±0,00	0,02±0,00
α2 globulin	0,26±0,01	0,27±0,01
βglobulin	0,07±0,00	0,14±0,07
γglobulin	0,05±0,01	0,04±0,00
a/g quotient	1,45±0,04	1,37±0,11

LD lactate dehydrogenase
ALT alanine aminotransferase
AST aspartate aminotransferase
CK creatine kinase
ALP alkaline phosphatase
a/g quotient = albumin/globulin quotient
* statistical significant difference ($p \leq 0.05$) between groups

Hematological parameters:

No significant differences were found in hematological parameters (white blood cells count and white blood picture, red blood cells count, hemoglobin, hematocrit and thrombocytes count) between the guinea pigs exposed to sulphur dioxide and the control group and the observed non-significant changes did not exhibit consistent trends (Tab. 2).

Table 2: Hematological parameters

Parameter	SO_2	Sham
leucocytes (10^9/l)	4,45±0,36	4,16±0,21
erythrocytes (10^{12}/l)	5,38±0,16	5,53±0,09
hemoglobin (g/l)	140,36±5,27	145,90±3,73
hematocrit (ratio)	0,43±0,01	0,43±0,01
MCV (fl)	79,10±0,82	79,00±0,79
trombocytes (10^9/l)	573,9±129,70	560,70±45,57
eosinophils (%)	0,27±0,14	0,50±0,31
basophils (%)	0,09±0,09	0,10±0,10
monocytes (%)	5,36±1,42	5,90±0,86
lymphocytes (%)	57,5±44,26	62,60±4,91

MCV mean cellular volume

Discussion

Almost no significant differences in the values of biochemical parameters and no significant differences in hematological parameters were found after the exposure of guinea pigs to SO_2 (400ppm, 3 hrs daily, 28 days). Therefore, it can be concluded that a subchronical exposure to sulphur dioxide mostly did not induce any definite changes of parameters studied in guinea pigs. These results are in accordance with the conclusions of the investigation of the subchronical effects of SO_2 on the cardiac function (where only mild changes were found, 8). Previous experiments (investigating the effects of an acute exposure to SO_2) revealed, on the other hand, more frequent and - though mild - significant changes in the followed-up biochemical and hematological parameters as well as in parameters of cardiac function. The influence on the respiratory system following acute exposure to SO_2 (disposition to artificially induced cough and airway reactivity to histamin were significantly enhanced - 12) has also been demonstrated. On the base of mentioned differences between subchronical and acute exposure to SO_2 an adaptation of the organism to some effects of SO_2 exposure in guinea pigs can not be excluded.

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