Respiratory Effects and Cytogenetic Changes Associated with Occupational Exposure to Thinners Compound

By

Amany Ahmed Khairy Kazem

MB, B.Ch, Faculty of Medicine – Ain Shams University 1986

M.Sc, Faculty of Medicine – Ain Shams University 1995

A Thesis submitted in partial fulfillment
of
the requirements for the Doctor of Philosophy
in
Environmental Science

Under the supervision of:

Prof. Dr. Mohamed Sayed Mansour El-Gewaily
Professor of Occupational Medicine,
Occupational Health and Environmental Health
Faculty of Medicine – Ain Shams University

Prof. Dr. Mahmoud Serry El-Bokhary
Professor of Chest Diseases and
Head of Medical Environmental Science Department
Institute of Environmental Studies and Research
Ain Shams University

Prof. Dr. Maha Roushdy El-Kady
Professor of Medical Research
National Center of Radiation Research and Technology

Prof. Dr. Hassan Abd El Sattar El-Dawy
Professor of Cytogenetic
National Center of Radiation Research and Technology

2011

Acknowledgements

Words are not enough to express my sincerest gratitude and my deepest thanks and grateful appreciation to **Professor Dr. Mohamed Sayed Mansour El-Gewaily**, Professor of Occupational Medicine, Occupational Health and Environmental Health, Faculty of Medicine – Ain Shams University, for supporting me throughout my thesis with his constructive suggestions, valuable advice and superior guidance throughout this study.

I'm deeply indebted to **Professor Dr. Mahmoud Serry El-Bokhary**, Professor in Medical Science Department, Institute of Environmental Studies & Research, Ain Shams University, for his kind help, sincere advice and co-operation through the preparation of this work. His constructive criticism, devoted effort, and faithful supervision will always be sincerely remembered.

I owe a special word of thanks to **Prof. Dr. Maha Roushdy El-Kady**, Professor of Medical Research, National Center of Radiation Research and Technology, for her co-operation, encouragement and for the chance she willingly gave me to carry this work forward. I would like also to thank her for her great support, valuable time, and careful supervision

I wish also to express my heartfelt thanks to **Prof. Dr. Hassan Abd El Sattar El-Dawy**, Professor of Cytogenetic, National Center of Radiation Research and Technology, for his valuable opinions, fruitful discussions and continuous support. Throughout all stages of my research, he saved no efforts in providing detailed guidance, illuminating suggestions and constant advice.

Last but not least, I would like to thank those who were working behind the scene, however they played the most effective role to finish this thesis and for facilities they offered me to make this work come to light, to them I am so grateful.

Dedication

This work is dedicated to my **family**: my **parents**, my **husband** and my **children** who supported, encouraged and motivated me when I badly needed that.

Abstract

The present study was carried out in order to evaluate the respiratory effects and the cytogenetic changes associated with occupational exposure to thinners compounds (mixture of organic solvents). This study included 40 male subjects working as painters. Four groups of studied individuals were chosen according to the type of handling thinners as follows: ten outdoor painters (Group II), ten cars in air painters (Group III) and ten cabinet painters (Group IV). All groups were compared to ten healthy normal unexposed subjects as controls (Group I).

Measurement of pulmonary function tests (FVC), (FEV_1), (FEV_1/FVC) and (FEF_{25-75}) were estimated to each group. Also plasma IL-5 and plasma MDA concentration were investigated.

Additionally, cytogenetic analysis was applied on lymphocyte cells to evaluate the genotoxic risk association. In addition, total and differential white blood cell counts were examined for all exposed groups. The obtained results were statistically analyzed and compared to the values of the healthy control group. A statistical significant decrease was recorded in pulmonary function tests among cabinet painters group only.

Regarding biochemical tests there were variable changes in them but the maximum increase in plasma MDA and IL-5 were observed in cabinet painters group. Also cytogenetic changes as chromosomal aberrations and MN frequency in lymphocytes were highly found especially in cabinet painters. Total and differential white blood cell count shows that all individuals have clinically normal percentage of white blood cells as compared to the controls.

In conclusion, those exposed workers may face very serious health problems, therefore we recommend for them periodical biological examinations and the use of personal protective devices aiming at decreasing genetic damage and risk of respiratory and further diseases.

Table of Contents

❋ ❋ ❋ ❋ ❋

List of Abbreviations

Ab:	aberration
ACD:	acid citrated dextrose
AM:	alveolar macrophage
ATS:	American Thoracic Society
B:	breaks
Chd:	chromatid
Chs:	chromosome
COPD:	Chronic obstructive pulmonary disease
Dic:	dicentric
DNA:	Deoxyribonucleic acid
dt:	Duncan's multiple range test
EDTA:	ethylene diamine tetra-acetic acid
EPA:	Environmental Protection Agency
F:	fragment
FEF_{25-75}%	Forced Expiratory Flow with average flow rate between 25% - 75% of FVC
FEV_1:	Forced Expiratory Volume in one second
Frag:	fragment
FVC:	Forced vital capacity
GINA:	Global Initiative for asthma
GM-CSF:	Granulocyte-macrophage colony-stimulating factor

IARC:	International Agency for Research on Cancer
IL-5:	Interleukin-5
Lpo:	Lipid peroxidation
LRT:	lower respiratory tract
MEK:	Methyl ethyl ketones
MN:	Micronuclei
MRL:	Minimal risk level
NADP:	Nicotinamide Adenine Dinucleotide Phosphate
NADPH:	Reduced form of NADP
NAL:	Nasal lavage
NIH:	National Institute of Health
NIOSH:	National Institute for Occupational Safety and Health
No.:	Number
NTP:	National Toxicity Porgram
OHS:	Occupational Health Services
%:	percent of aberrent cells
ppm:	part per million
PUFA:	poly unsaturated fatty acids
RNA:	Ribonucleic acid
ROS:	Reactive oxygen substances
Rpm:	revolution per minute
S.D.	Standard Deviation
SAS:	Statistical Analysis System

SCE:	Sister chromatid exchange
SIR:	standardized incidence ratio
SMR:	standardized morbidity ratio
O_2^{--}	Superoxide anion
TBA:	Thiobarbituric acid
TBARS:	Thiobarbituric acid reactive substances
TEAM:	Total Exposure Assessment Methodology
TH cell:	T helper cells
Tr. dic.:	Tri-radial dicentric
URT:	upper respiratory tract
VOCs:	Volatile organic compounds
WBCs:	white blood cells
WHO:	World Health Organization

List of Tables

List of Figures

Introduction

Spray painting contaminates workplace air with inhalable paint ingredients in the form of particles and vapours. Spray painters, therefore, work in a potentially hazardous environment where the level of risk depends on the intensity of exposure and the toxicities of the materials used ***(Henry et al., 2004)***.

Exposure to harmful pollutants in work place can cause various diseases with short or long latent period for manifestations. Exposure to solvents and paints mainly affects the respiratory and nervous systems **(*Lundberg et al., 1994*)**. Occupational exposures make an important contribution to the burden of obstructive airway diseases for example, asthma and chronic obstructive pulmonary diseases (COPD) ***(Hammond et al., 2005)*.**

There is growing concern about possible mutagenic and carcinogenic effects of environmental agents in occupationally exposed workers. Painters are exposed to an extensive variety of hazardous substances like organic solvents, some of which have shown clastogenic activity **(*Carrnao et al., 1988*)**.

Paint thinner is commonly used in industry. The toxic effect of paint thinner is caused by formation of reactive oxygen species (ROS). There are several studies reporting that paint thinner causes cellular damage via formation of ROS (***Ulakoglu et al., 1998*** *and* ***Mattia et al., 1991****)*. ROS are believed to cause lipid peroxidation resulting in damage to biological membranes. Examination of the pharmacology and epidemiology of inhalation of thinner and other organic solvents, especially the ones used in the paint industry, requires great attention (***Carbabez et al., 1998***). The main components of thinner used in industry are toluene (63 percent), acetone (13 percent), isobutyl acetate (10 percent), isobutanol (7.5 percent) and butyl glycol (6.5 percent)

(***Ulakoglu et al., 1998***). Abuse of this mixture as a narcotic agent by young people is a very important health problem. Thinner taken by inhalation shows its effects on the intestines, liver, kidneys, adrenal gland and the central nervous system (***Carabez et al., 1998*****)**. Toluene unlike other components of thinner can diffuse into body fluids and causes formation of (**ROS) *(Nakajima and Wang, 1996) and (Kato et al., 1990*)** which are the main agents responsible for cellular damage. Superoxide anions, ferryl ions and hydroxyl ions are the common reactive compounds that cause lipid peroxidation (***Halliwell et al., 1986)***. The present study aimed to examine the effects of thinner inhalation on painters who have to deal with thinner in the course of their work. These people constantly and involuntarily inhale thinner during their working hours. One of the products of lipid peroxidation is plasma malondialdehyde (MDA) which was measured in these workers and in control group composed of healthy subjects.

Many industrial solvents possess mutagenic or carcinogenic potential; there is also evidence that cytogenetic damage is associated with occupational exposure to organic solvents. Increased frequencies of micronuclei (MN) in lymphocytes and in buccal epithelial cells were observed among paint industry workers, which were attributed to working conditions mostly to the organic solvents present in the working areas ***(Bender et al., 1989*****).** Similar considerations were made by ***Silva and Santos-Mello (1996*****)** who observed increased proportions of aneuploid lymphocytes and chromosome deletions in car painters and by ***Fuchs et al. (1996)*** who reported a transient increase in DNA strand breaks in car spray painters.

Genetic damage can have serious effects on human health, including a wide range of hereditary diseases, cancer, congenital anomalies and even reduced life expectancy. The induction of damage to the germ line and to the mechanisms that control cellular division in living organisms has therefore, substantial consequences on health and environment. So, the evaluation of

genetic hazards in a population (environmentally and occupationally) due to certain mutagenic exposure is necessary ***(Brusick et al., 1992)***.

Knowledge about the health risk can be derived from biomarker-based, population-monitoring studies; one of these biomarkers is the cytogenetic assays, and one of these assays is the chromosomal aberration analysis which has a major advantage in biomarker-based studies which can provide advance warning signals for the development of health effects ***(AU, 1991)***.

Oxidative stress, which can be defined as an increased exposure to oxidants and/or decreased antioxidant capacities is widely recognized as a central feature of many diseases and considerable evidence now links COPD and asthma with increased oxidative stress. There are technical difficulties in measuring specific markers of Oxidative injury. Instead, investigators rely on indirect measurements of free radical activity in biological fluids such as measurements which assess oxidative damage to lipids, proteins or DNA. Levels of lipid peroxides in plasma measured as lipid peroxidation products assessed as thiobarbituric acid reactive substances (TBARS) are significantly increased in patients with acute exacerbation of COPD and asthma ***(Rahman and MacNee, 1996)***. Oxygen radicals can also produce DNA nicking chromosomal aberrations causing mutations ***(Repine et al., 1997).***

Chronic alcohol exposure (as ethanol which a component of thinner compound) alters leucocytes count and percentage of total nucleated cells ***(Livant et al., 1997).***