THE JOURNAL OF
SEXUAL MEDICINE

ORIGINAL RESEARCH

BASIC SCIENCE

Formaldehyde Inhibits Sexual Behavior and Expression of Steroidogenic Enzymes in the Testes of Mice

Zhi-Jun Zang, MD, PhD,[1,*] You-Qiang Fang, MD, PhD,[2,*] Su-Yun Ji, MD, PhD,[3] Yong Gao, MD, PhD,[4] Yuan-Qiang Zhu, MSc,[1] Ting-Ting Xia, MSc,[1] Mei-Hua Jiang, PhD,[5] and Ya-Nan Zhang, MSc[1]

ABSTRACT

Background: Formaldehyde, a ubiquitous environmental pollutant, is used extensively and has been proved to impair male reproduction in mammals. However, no trials have explored whether formaldehyde affects sexual function.

Aim: To evaluate the effect of long-term formaldehyde exposure on sexual behavior and to investigate the potential mechanism.

Methods: Forty C57BL/6 male mice were randomly allocated to four equally sized groups. Mice were exposed to formaldehyde at a dose of 0 (control), 0.5, 5.0, or 10.0 mg/m^3 by inhalation for 60 days.

Outcomes: Sexual behavior, body and reproductive organ weights, testosterone concentration in serum and testicular tissue, expression of steroidogenic enzymes, quality of sperm, and testicular structure were measured.

Results: Formaldehyde inhibited sexual behavior and decreased reproductive organ weights in mice. Serum testosterone levels and intratesticular testosterone concentrations were decreased in the formaldehyde-treated groups. Expression levels of steroidogenic enzymes, including steroidogenic acute regulatory protein, cytochrome P450 cholesterol side-chain cleavage enzyme, and 3β-hydroxysteroid dehydrogenase (3β-HSD), also were decreased in the testes of mice exposed to formaldehyde. Moreover, the structure of seminiferous tubules was destroyed and sperm quality decreased after formaldehyde exposure. In addition, the results indicated that the effects of formaldehyde were dose dependent.

Clinical Implications: Efforts should be undertaken to decrease impairment of sexual function caused by formaldehyde exposure.

Strengths and Limitations: The relatively small sample might have affected the outcomes. Further experiments are needed to study the mechanism of action of formaldehyde.

Conclusion: Exposure to formaldehyde gas inhibited sexual behavior, caused reproductive organ atrophy, and impaired spermatogenesis in male mice, which might have been induced by suppressed expression of steroidogenic enzymes in Leydig cells and decreased testosterone synthesis. **Zang Z-J, Fang Y-Q, Ji S-Y, et al. Formaldehyde Inhibits Sexual Behavior and Expression of Steroidogenic Enzymes in the Testes of Mice. J Sex Med 2017;14:1297–1306.**

Key Words: Formaldehyde; Sexual Behavior; Steroidogenic Enzymes; Testosterone; Spermatogenesis; Mice

Received April 20, 2017. Accepted September 1, 2017.

[1]Department of Infertility and Sexual Medicine, The Third Affiliated Hospital, Sun Yat-sen University, Guangzhou, People's Republic of China;

[2]Department of Urology, The Third Affiliated Hospital, Sun Yat-sen University, Guangzhou, People's Republic of China;

[3]Department of Dermatology, Guangdong Provincial Dermatology Hospital, Guangzhou, People's Republic of China;

[4]Reproductive Medicine Centre and Guangdong Provincial Key Laboratory of Reproductive Medicine, The First Affiliated Hospital, Sun Yat-sen University, Guangzhou, People's Republic of China;

[5]Department of Anatomy, Zhongshan School of Medicine, Sun Yat-sen University, Guangzhou, People's Republic of China

*Equivalent contribution.

http://dx.doi.org/10.1016/j.jsxm.2017.09.001

INTRODUCTION

Formaldehyde is a high-volume chemical that is ubiquitously found in the environment. Building materials, consumer products, and combustion processes are the major indoor air sources of formaldehyde.[1] Because formaldehyde is widely used in industry and households, people are increasingly in contact with it indoors, outdoors, at work, in residences, and through medicine and food, mainly by inhalation.[2] Common symptoms of acute inhalation of formaldehyde include headache, streaming runny eyes, sniffling, coughing, sneezing, nausea, and dyspnea.[3] Humans will feel uncomfortable when the concentration of formaldehyde in the air exceeds 0.1 mg/m^3, have teary eyes at concentrations higher than 0.5 mg/m^3, develop a sore throat at concentrations higher than 0.6 mg/m^3, develop dyspnea, coughing, chest tightness, and headache at concentrations of 12 to 24 mg/m^3, and develop pneumonia, emphysema, and even death at concentrations higher than 60 mg/m^3.[4] The occupational limit of formaldehyde exposure in China is 0.5 mg/m^3.[5] However, it has been reported that formaldehyde concentrations in some occupational environments are much higher than this standard. For example, Jie et al[6] observed that the concentration of formaldehyde in the indoor air of public places was 0 to 1.12 mg/m^3. Fan et al[7] reported that formaldehyde concentrations in several hospital pathology laboratories were as high as 5.84 mg/m^3. Zhang et al[8] found that formaldehyde concentrations in some anatomy laboratories were higher than 10 mg/m^3. Long-term exposure to formaldehyde can induce multisystem diseases. The International Agency for Research on Cancer has classified formaldehyde as a human leukemogen.[9,10] Epidemiologic studies have reported that exposure to formaldehyde can lead to diseases such as leukemia,[11] Hodgkin lymphoma,[11] asthma,[12] bone marrow toxicity,[13] and chromosomal aberrations.[14] It has been reported that S-phase sensing of DNA-protein crosslinks triggering DNA topoisomerase 2-binding protein-1—independent ataxia telangiectasia mutated and Rad3-related activation and p53-mediated cell death induced by formaldehyde might be important molecular mechanisms of formaldehyde-induced toxicity.[15]

A substantial body of evidence from humans and animals has indicated negative effects of formaldehyde on male reproduction. A meta-analysis by Duong et al[16] addressed reproductive and developmental toxicities in formaldehyde-exposed humans, including a high risk of spontaneous abortion, birth defects, low birth weight, and malformations. Majumder and Kumar[17] reported that formaldehyde could decrease sperm motility, viability, and counts and even decrease the DNA content in the testes and prostates of rats. Zhou et al[18] found that Fas-mediated apoptosis might be one of the basic mechanisms involved in the reproductive toxicity of formaldehyde. Han et al[19] indicated that formaldehyde exposure triggered autophagy, which might be a critical factor responsible for male reproductive impairment induced by formaldehyde.

Formaldehyde also can affect cognitive function and behavior. A previous study reported that formaldehyde could alter the metabolism of brain cells, induce neurodegeneration, and impair cognitive performance.[20] A moderate concentration of inhaled formaldehyde can negatively affect learning and memory in mice.[21] Exposure of male mice to formaldehyde vapor can affect their locomotor and explorative activity in the open field.[22] However, to the best of our knowledge, the effect of formaldehyde on sexual behavior has not been identified.

We hypothesized that long-term exposure to formaldehyde might inhibit sexual behavior by disrupting testosterone biosynthesis. To test this hypothesis, we evaluated sexual behavior and testosterone levels in serum and testis tissue, examined the expression of steroidogenic enzymes, including steroidogenic acute regulatory protein (StAR), cytochrome P450 cholesterol side-chain cleavage (P450scc) enzyme, and 3β-hydroxysteroid dehydrogenase (3β-HSD), in Leydig cells, and evaluated sperm quality in the epididymis.

METHODS

Animals and Gaseous Formaldehyde Exposure

C57BL/6 mice (10 weeks old, 40 male and 30 female) were purchased from the Animal Center of Sun Yat-sen University (Guangzhou, China) and were housed five per cage under standard conditions (12-hour light-dark cycle [lights on from 7:00 PM to 7:00 AM], temperature = 22 ± 1°C, humidity = 50—70%). All male mice were randomly assigned to four equal-size groups (10 mice per group). The mice in each experimental group were exposed to formaldehyde (0.5, 5.0, or 10.0 mg/m^3, 4 hours daily) through inhalation from 8 to 12 AM for 60 consecutive days. Control animals inhaled distilled vapor. Gaseous formaldehyde was prepared from 10% formalin. Formaldehyde vapor was generated and administered to mice using HOPE-MED 8052 inhalation equipment (HOPE-MED Company, Tianjin, China). During exposure, the concentration of formaldehyde was monitored using a gaseous formaldehyde analyzer (4160-2, Interscan, Simi Valley, CA, USA). The female mice, which were used as sexual stimuli, underwent double ovary resection 1 month before sexual behavior testing. Before behavioral testing, each female mouse was pretreated with estradiol benzoate (50 μg dissolved in peanut oil 50 μL 48 hours before testing) and progesterone (500 μg dissolved in peanut oil 50 μL 5 hours before testing) to reach a state of estrous. The animals were handled in accordance with the guidelines of the Sun Yat-sen University institutional animal care and use committee. All efforts were made to minimize the number of mice used and to decrease animal suffering.

Sexual Behavior Testing

The experimental procedure was adapted from that described previously.[23,24] The apparatus for the sexual behavior tests was a clean cage (40 × 26 × 21 cm) in a quiet environment. Before