

Histopathologic Changes of Rat Tracheal Mucosa Following Formaldehyde Exposure

Cambios Histopatológicos de la Mucosa Traqueal de la Rata Seguidos a la Exposición de Formaldehído

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SUMMARY: Formaldehyde is a chemical, which is used traditionally for fixing the cadaver. It is vaporized during dissection and practical studying on cadaver. Studies show that this vapor can cause some clinical symptoms such as throat, eye, skin and nasal irritation.

This study was designed to determine the histopathologic changes of rat tracheal mucosa while all of the experiments were exposed to formaldehyde for 18 weeks. This study was performed on 28, 6-7 weeks postnatal albino Wistar rats. The rats were divided into 3 case groups (E1: 4h/d, 4d/w; E2: 2h/d, 4d/w; E3: 2h/d, 2d/w) and 1 control group. The tracheal specimens were sectioned and stained with H&E technique for histopathologic study.

An epithelial disorganization, cilia disappearance, slight dysplastic changes and slight subepithelial lymphocytic infiltration were observed in the case of E1. Epithelial disorganization, irregular cilia and slight subepithelial infiltration were seen in E2 and E3 groups. The results of this study show that "the more exposure to formaldehyde vapor, the more intense epithelial changes".

KEY WORDS: Formaldehyde exposure; Rat; Tracheal mucosa.

INTRODUCTION

Formaldehyde (CH₂O) is a flammable, colorless reactive, readily polymerized gas at normal room temperature and pressure, with a relative molecular mass of 30.03, and a pungent odor. Formaldehyde is soluble in water, ethanol and diethyl ether. Also, it is used in polymerized form (Paraformaldehyde) (World Health Organization, 1989).

Under atmospheric conditions, formaldehyde is readily photo-oxidized by sunlight to carbon dioxide. In the absence of nitrogen dioxide, the half-life of formaldehyde is approximately 50 minutes during the daytime; in the presence of nitrogen dioxide, this drops to 35 minutes (World Health Organization).

There are various sources of formaldehyde, but the major anthropogenic sources which affecting humans are in the indoor environments. Other anthropogenic sources include direct emissions; especially from the production and use of formaldehyde (World Health Organization).

Its potential to act as an electrophile and act with macromolecules such as DNA, RNA and protein to form reversible adducts or irreversible cross-links (International Agency for Research on Cancer, Lyon, 1995) makes it as a conventional tissue fixative (particularly in cadaver's fixation).

Acute formaldehyde exposure produces mainly mucosal irritation of the eye and upper respiratory tract in humans (Zwart *et al.*, 1988) and a long-term exposure leads to the production of nasal tumors in rodents (Monticello *et al.*, 1996). Formaldehyde also causes pulmonary function impairment (Berbstein *et al.*, 1984) and asthmatics reactions in sensitized individuals (Burge *et al.*, 1985; Gorski *et al.*, 1991).

In the dissection lab, and during cadaver's dissection, instructors of Anatomy and medical students are exposed to formaldehyde vapor derived from cadaver's fixative.

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In order to study the histopathologic changes in the tracheal respiratory mucosa, due to formaldehyde exposure in the dissection lab, and determining its relationship with the duration of exposure, this study was carried out on 28 Albino Wistar rats.

MATERIAL AND METHOD

This study was done on 28, 6-7 weeks postnatal Albino Wistar rats (bought from Iranian Pastor Institute), randomly divided into three equal case groups, based on the differences between exposure periods: E1 (4h/d, 4d/w), E2 (2h/d,4h/w), E3(2h/d,2d/w) and a control group without any exposure.

Using a digital scale, the mean weights for each group were 252g (E1), 209g (E2) and 222g (E3) and 195g (control group). The concentration of formaldehyde vapor was measured at the beginning, during and at the end of the study by means of Detector Tube and Dragger Pump (model 31, made in Germany) after the covers of the cadavers were removed. The vapor concentration of dissection room was 1-1.5 ppm when the air-conditioner was ON and 1.5-1.9 ppm when it was OFF. The temperature of dissection room was 20-26 °C and the air pressure was 760-763 atm.

At non-exposure times, all groups were kept in laboratory animal house, which was far from the place of exposure with no formaldehyde detection. The animal house was ventilated and its temperature was kept around 21°C with air conditioner system and adequate light was prepared. All groups were fed with a standard similar diet (bought from Iranian Pastor Institute) two times a day (morning and afternoon); but water was available all the time (*ad libitum*). The cages of the case groups were placed at a height the same as cadaver's height with a distance of 15cm apart from them for 18 weeks, corresponding time protocols mentioned above. During each period of exposure, the control group was kept in the animal house.

When the exposure period was expired, each of the rats of both experiments and control groups were anaesthetized with chloroform. After cervical delocalization, the thorax, was dissected and tracheal specimens with 4mm length were taken about 5mm above the "Carina Angle ". These specimens were fixed in "Clark solution", for 48h. After tissue processing and paraffin embedding, 10 sections from each specimens were cut at 4µm and stained with Hematoxylin and Eosin (H&E). All of the sections were studied by Olympus light microscope, with multiple magnifications (40x, 100x, 400x).

RESULTS

In control group, no histopathologic changes were seen (Fig 1). Disorganization of epithelium, as well as disappearance of epithelial cilia was found within all tissue sections of group E1. Epithelial cells showed mild dysplastic changes and their nuclei were also hyperchromatic. Slight subepithelial lymphocytic infiltration were also seen (Fig.2).

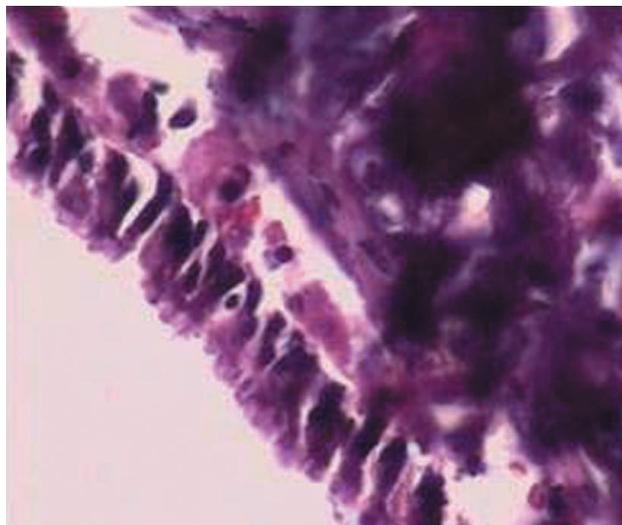


Fig. 1. Shows no histopathologic changes in control groups (SM). H. E. 400x.

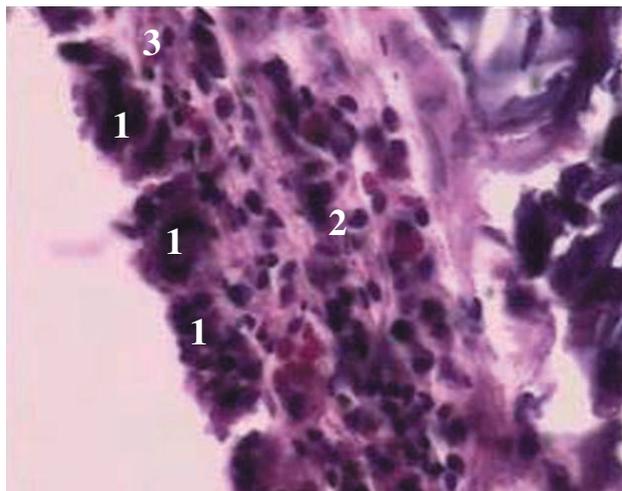


Fig. 2. Shows epithelial disorganization (1), mononuclear cell infiltration (2) and intense nuclear hyperchromasis (3) in group E1. H. E. 400x.

The histopathologic changes in group E2 and E3 were less intensive, when these changes were compared with control group. Epithelium was disorganized and the cilia were irregular. Also slight subepithelial lymphocytic infiltration was seen (Figs. 3 and 4).

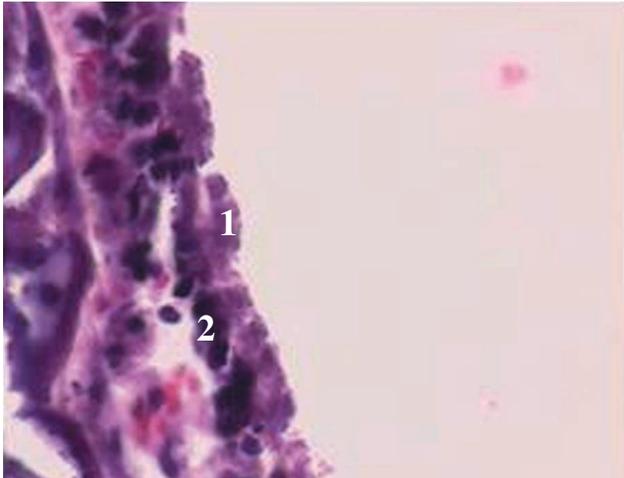


Fig. 3. Shows epithelial disorganization (1) and mononuclear cell infiltration (2) in group E2. H. E. 400x.

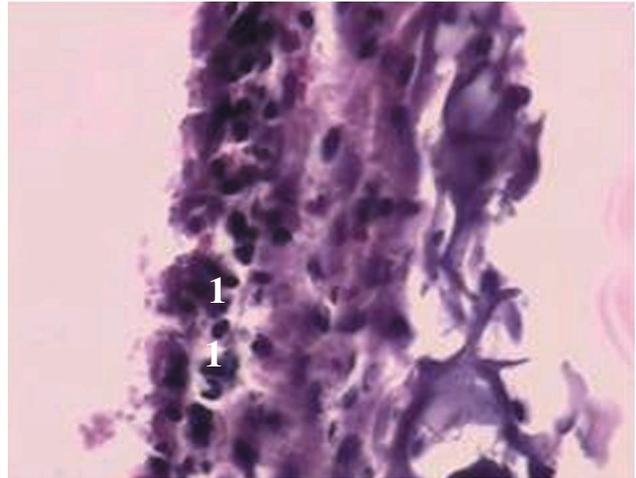


Fig. 4. Shows epithelial disorganization (1) in group E3. H. E. 400x.

DISCUSSION

The results of our study showed that exposure to formaldehyde vapor, cause histopathologic changes. such as: subepithelial lymphocytic infiltration, dysplastic changes, hyperchromasis of nuclei, epithelial disorganization and cilia disappearance of rat tracheal epithelium as well.

The data obtained in the experiment of Kamata *et al.*, (1997) showed inflammatory cell infiltration in the nasal mucosa of all groups exposed to 0, 0.3, 2 and 15 ppm. Also Ohtsuka *et al.*, (1998) reported mild inflammation of nasal septal mucosa in F-344 rats and BN rats exposed to aerosol inhalation of 1% formaldehyde.

Cellular disarrangement and hyperchromatic nuclei of rat respiratory mucosa accompanied by mild dysplasia found in our experiment resemble the study done by Javedan *et al.*, (1999) on rat nasal mucosa, exposed to 2 and 5 ppm.

Using an electron microscope, Monterio-Riviere & Popp (1966), reported abnormal cilia in rat nasal epithelium exposed to 0.5 ppm. Their findings, reinforces our findings about the absence of cilia in groups E1.

In contrast to our findings, exposure of rats to 2 ppm in the study of Wilmer *et al.*, (1989) and exposure to 1 ppm in the study of Zwart *et al.* showed no histopathologic effects in nasal mucosa.

When using "duration" as a factor, histopathologic changes found in groups E1 (4h/d, 4d/w) were more intensive than in groups E2 (2h/d, 4d/w) and E3 (2h/d, 2d/w).

In a similar study of Kerns *et al.*, (1983) on rat nasal mucosa exposed to 2, 5, 6 and 14.3 ppm, showed non-neoplastic dysplasia in 14.3 ppm group, after 6 months of exposure while neoplastic dysplasia were seen in the same group after 18 months of exposure.

Feron *et al.*, (1988) exposed three groups of rats to 10 ppm for 4, 8 and 13 weeks of duration and reported mild squamous metaplasia only in 13-week exposed group.

Javedan *et al.*, divided Albino Wistar rats into two groups of acute and subacute exposure and exposed them to 2 and 5ppm. They found that histopathologic changes were more intensive in subacute exposure group in comparison to acute exposure group.

In contrast to above-motioned studies, we omitted the effects of aging and gradual recovery that may be affecting the findings of others, by letting all groups to expose to the formaldehyde vapor from beginning to the end.

The results obtain in our study, pertaining to the relationship between histopathologic changes and exposure duration, are in agreement with those obtained by Kerns *et al.*; Feron *et al.* and Javedan *et al.* Consequently it seems that "the histopathologic changes have direct relationship with formaldehyde vapor exposure duration."

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DAVARIAN, A.; FAZELI, S. A.; AZARHOUSH, R. & GOLALIPOUR, M. J. Cambios histopatológicos de la mucosa traqueal de la rata seguidos a la exposición de formaldehído. *Int. J. Morphol.*, 23(4):369-372, 2005.

RESUMEN: El formaldehído es un producto químico que se usa tradicionalmente para la fijación de cadáveres. Éste se vaporiza durante la disección y los estudios prácticos en el cadáver. Investigaciones han mostrado que este vapor puede causar algunos síntomas clínicos como la irritación de garganta, ojos, piel y mucosa nasal.

Este estudio tuvo como objetivo determinar los cambios histopatológicos en la mucosa de la tráquea de la rata para lo cual todos los animales fueron expuestos al formaldehído, durante 18 semanas. El estudio fue realizado en 28 ratas albinas Wistar, con 6-7 semanas de vida. Las ratas fueron divididas en 3 grupos (E1: 4h/día, 4días/semana; E2: 2h/día, 4días/semana; E3: 2h/día, 2días/semana) y un grupo control. Fueron seccionadas las tráqueas de los especímenes y teñidas con H&E para su estudio histopatológico.

Una desorganización epitelial, desaparición de cilios, leves cambios dispásticos y leve infiltración linfocítica subepitelial fueron observados en el grupo E1. Desorganización epitelial, cilios irregulares y leve infiltración subepitelial fueron observados en los grupos E2 y E3. Los resultados de este estudio muestran que a mayor exposición al vapor del formaldehído, más intensos son los cambios epiteliales de la mucosa traqueal.

PALABRAS CLAVE: Exposición al formaldehído; Rata; Mucosa traqueal.

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