Available online at www.sciencedirect.com
ScienceDirect
Toxicology and Applied Pharmacology 225 (2007) 229–237
Toxicology and Applied Pharmacology
www.elsevier.com/locate/ytaap

Contemporary Issues

Toxicologic significance of histologic change in the larynx of the rat following inhalation exposure: A critical review

Thomas G. Osimitz [a,*], Wiebke Droege [a], John M. Finch [b]

[a] *Science Strategies, LLC, 600 E. Water Street, Suite C, Charlottesville, VA 22902, USA*
[b] *Charles River Laboratories, Preclinical Services Tranent, Edinburgh, EH33 2NE, UK*

Received 18 May 2007; revised 20 August 2007; accepted 31 August 2007
Available online 12 September 2007

Abstract

The larynx is a site in the respiratory tract of animals that often shows a response to inhaled substances. In many cases, the most sensitive endpoint in repeated dose inhalation studies is squamous metaplasia (often of minimal severity) of the larynx. The U.S. Environmental Protection Agency has speculated that squamous metaplasia in the rodent larynx might be a pre-neoplastic lesion or a precursor to other serious effects and has proposed to use the effect of squamous metaplasia occurring in subchronic inhalation toxicology studies as a toxicologic endpoint for use in quantitative risk assessment [U.S. Environmental Protection Agency, 2006a. Reregistration Eligibility Decision for MGK-264, U.S. Environmental Protection Agency, 2006b,Reregistration Eligibility Decision for Piperonyl Butoxide, U.S. Environmental Protection Agency, 2006c. Reregistration Eligibility Decision for Pyrethrins]. To reach a conclusion as to its significance, we sought to establish the nature of this effect in the relevant context of rodent inhalation studies.

A comprehensive review of the literature shows that laryngeal metaplasia can be produced by a wide range of chemically dissimilar substances, and even by "non-chemical" means such as irritation by aerosols and particles, and dehydration by alcohols or low humidity air. There is no published evidence that this effect is pre-neoplastic and it is clearly and repeatedly characterized as an adaptive response. Moreover, the well-differentiated character of laryngeal squamous metaplasia, the reversibility of incidence and severity of it during recovery periods, combined with no significant clinical observations and the lack of progression over time indicates that this response is adaptive and should not be considered to be indicative of significant human risk. We therefore conclude that squamous metaplasia of the rodent larynx is not a relevant toxicologic endpoint.

Keywords: Metaplasia; Larynx; Rat; Respiratory; Inhalation; Human risk; Toxicologic endpoint

Contents

* Corresponding author. Fax: +1 434 817 1425.
E-mail address: tom@sciencestrategies.com (T.G. Osimitz).

0041-008X/$ - see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.taap.2007.08.027

Introduction

The larynx is a site in the respiratory tract of animals that often shows a response to inhaled substances. In many cases, the most sensitive endpoint in repeated dose inhalation studies is squamous metaplasia (often of minimal severity) of the larynx. The U.S. Environmental Protection Agency (EPA) has proposed to use the effect of squamous metaplasia occurring in subchronic inhalation toxicology studies as a toxicologic endpoint for use in quantitative risk assessment (USEPA, 2006a,b,c). The EPA has speculated that squamous metaplasia in the rodent larynx might be a pre-neoplastic lesion or a precursor to other serious effects. It is therefore essential to establish the nature of this effect, in the relevant context of rodent inhalation studies, in order to reach a conclusion as to its significance.

The purpose of this review was to search and analyze the relevant literature to address the question of whether squamous metaplasia of the rodent larynx should be considered as a toxicologic, potentially pre-neoplastic effect, or as an adaptive response. On the basis of our review, there is no published evidence that this effect is pre-neoplastic and it is clearly and repeatedly characterized as an adaptive response. We therefore conclude that squamous metaplasia of the rodent larynx is not a relevant toxicologic endpoint and should not be used as a toxicologic endpoint for quantitative risk assessment.

Background

Comparative morphology of the larynx

Rodents (rats, mice, and hamsters) are generally regarded as good models for humans and are the most common animals used for inhalation toxicology studies (Renne et al., 1993). Although notable morphological differences exist between different rodent species, upon comparison of Sprague–Dawley rats with Syrian Golden Hamsters, and to some extent with Fischer 344 rats, the authors concluded that rodent larynges offer a better animal model than other mammals (e.g. dogs) for the human respiratory tract.

Detailed reviews of the micro-anatomy of the rodent larynx have been provided by various authors (Renne et al., 1993, 2007; Lewis, 1991; Renne and Gideon, 2006) and in this review we wish only to highlight those aspects relevant to the safety assessment of cellular changes. An important similarity between human and rodent larynges is that both feature a similar transitional area from squamous to pseudostratified ciliated columnar (respiratory) epithelium. It is this region that is predisposed to developing cellular changes upon inhalation exposure to xenobiotics with airflow characteristics and epithelial sensitivities playing a role (Gopinath et al., 1987; Lewis, 1991; Renne and Gideon, 2006; Renne et al., 2007). That being said, differences between the morphology of the human and rodent laryngeal epithelium do exist with regard to exact location within the larynx, thickness, and type of cells. The main relevant difference between rats and humans in this respect is that this area of transition occurs at the base of the epiglottis in rats whereas in humans it occurs at the vocal folds. However, in each species it is this area of transition that is the most susceptible to toxicologic effects. In dogs and monkeys stratified squamous epithelium on the epiglottis is thicker and more extensive than in rodents giving better protection from inhaled materials.

Cellular responses in the respiratory tract

The following changes, benign and malignant, can occur to normal cells:

1) Hyperplasia is an increase in the number of cells in a tissue and this results from an increase in cell division. It can occur as the result of a physiologic, pathologic or compensatory process. It is defined as a non-neoplastic proliferation of cells;
2) Metaplasia is the replacement of one differentiated adult cell by a differentiated adult cell of another kind. It may appear in concurrence with hyperplasia or dysplasia. It is a reversible, non-neoplastic, adaptive proliferation of cells;
3) Dysplasia is an atypical cellular proliferation. A characteristic is partial or complete loss of specialized structures. Dysplasia is not considered a neoplastic lesion but, clinically, can be a precursor to malignant neoplastic changes;
4) Neoplasia is defined by cellular proliferation and growth without causal, external stimulation. Benign and malignant neoplasms exist, with the latter having the ability to invade surrounding tissue and to metastasize.

In practice, metaplasia is limited in adult mammals to the replacement of one epithelial tissue by another or by the replacement of one mesenchymal tissue by another. Although some rare examples of direct metaplasia (where one cell actually changes its own phenotype to become a second cell type) are believed to occur; generally metaplasia involves the loss of the first cell type and its replacement by proliferation of reprogrammed precursors into a new cell type that is better adapted to the new circumstances (indirect metaplasia). Laryngeal squamous metaplasia is a classic example of this indirect metaplasia. Inhalation of an irritant damages sensitive respiratory or transitional epithelium, so the cells that proliferate to replace the lost cells produce a replacement epithelium that is better adapted to the new environment.

The important features of metaplasia that link it with neoplasia are that metaplasia is a form of cell proliferation and as such usually occurs in response to chemical and mechanical