# Histological changes in the lung of Wistar albino rats (*Rattus norvegicus*) after exposure to Arabian incense (Genus *Boswellia*)

Majed S. Alokail, MSc, PhD; Saud A. Alarifi, PhD

ncense (Bokhour) is an oleoresin that oozes from incisions in the trunks and leaves of trees of the genus *Boswellia* (*B. carterii* and *B. papyrifera*, native to Arabia, Africa and India).<sup>1</sup> In Arabian cultures the incense is traditionally burnt for several domestic purposes. airborne pigmented and depigmented dermatitis have been reported as being caused by incense.<sup>2</sup>

Several investigators in Asia and North America have investigated possible links between incense stick exposure and health problems.<sup>3-5</sup> Extensive indoor incense exposure has been reported as a risk factor for obstructive lung disease in non-smoking Saudi women.<sup>6</sup> A significant association between lung cancer risk factors and incense stick exposure has been reported.<sup>7,8</sup> In addition, Arabian incense exposure was an important precipitating factor in about 18% of 414 asthmatic children ranging in age from 7 months to 12 years in Qatar.9 As far as we know, these reports are the only studies related to the effect of Arabian incense (Bokhour) on lung morphology. The association between exposure to Arabian incense (Bokhour) and the effect on human health has not been investigated deeply. In this investigation, our interest was to study the histological characteristics of two kinds of popular, commercially available incense.

## Methods

Wistar albino male rats (*Rattus norvegicus*) of approximately the same age, weighing 95  $\pm$ 10 grams (King Saud University colony) were kept in the laboratory under constant conditions of temperature (25°C) for the period of study, and maintained on a standard diet. Fresh chow and water were available ad libitum. One group of rats was exposed to expensive Bokhour incense smoke (n=16), another to inexpensive Bokhour incense smoke (n=16), and a third unexposed group served as controls (n=16). Rats in the first two groups were exposed to 420 grams of incense for 14 weeks at the rate of 4 grams per day in an exposure chamber.

After completion of the treatment regimen, the rats were anesthetized by diethyl ether, the lungs were removed and submitted for histological studies. The inflated lungs were fixed unless noted otherwise. The choice of fixative depended on the histological procedure to be performed. When morphometrical studies are involved, shrinkage of tissue is minimized by choosing a fixative with the correct osmolarity, usually 2% glutaraldehyde. After fixation, lung or lobe volume was determined by water displacement. Five From the Department of Biochemistry, College of Science, King Saud University, Riyadh, Saudi Arabia

Correspondence to: Dr. Majed S. Alokail Department of Biochemistry (5) College of Science King Saud University P.O. Box 2455 Riyadh 11451 Saudi Arabia

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micron thick paraffin sections of the lung were stained with hematoxylin and eosin, Sirius red (collagen) or Verhoeff-Van Gieson (elastic tissue) stains. All of the sections were reviewed for both the extent and severity of tissue injury. After careful examination, the selected slides were photographed on colored films.

#### Results

Control rats showed normal lung architecture without alteration in the components of the bronchioles, alveolar ducts, alveolar sacs, alveoli or psneumocytes (Figure 1). Incense produced alterations in the interstitial septa and the vascular tissues in the lungs of the treated rats. Marked inflammatory infiltrates consisting mainly of lymphocytes and plasma cells were seen. These inflammatory infiltrates were mainly localized in the perivascular connective tissue and in the lamina propria of some bronchioles (Figure 2). Some bronchioles of incense-treated rats contained neutrophilic exudates and cell debris (Figure 3), but there were no exudates in the alveolar ducts, alveolar sacs and alveoli. Alveolar septa surrounding the bronchioles were thickened and infiltrated by lymphocytes, histocytes and neutrophils (Figure 4). Moreover, incense exposure produced lung lesions with increased numbers of pigmented alveolar macrophages distributed throughout the parenchyma and focal collections of enlarged alveolar macrophages with concomitant alveolar epithelial hyperplasia.

## Discussion

This study examined the influence of the Arabian incense (Bokhour) on the risk of histological alteration of lung tis-

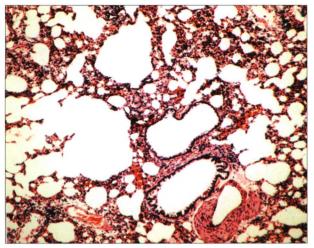


Figure 1. Light micrograph of control rat lung showing normal bronchioles, alveolar ducts and alveolar sacs (H&E, X80).

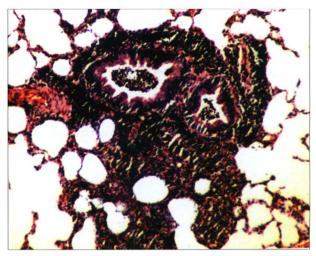


Figure 2. Light micrograph of incense exposed rat lung showing inflammatory infiltrates in the perivascular connective tissue and in the lamina propria of some bronchioles (H&E, X80).



Figure 3. Light micrograph of incense exposed rat lung showing bronchioles containing neutrophilic exudates and cell debris (H&E, X80).

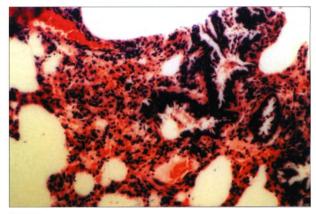


Figure 4. Light microscope of incense exposed rat lung showing alveolar septa surrounding that the bronchioles are thickened and infiltrated by a number of inflammatory cells (H&E, X80).

sue. The widespread use of incense in the Arabian tradition for several domestic purposes has prompted the need for investigation on whether incense smoke exposure might carry hazards to human health. There is little information on the relationship between the Arabian incense and human health. Fumes of incense as a major indoor hazard were recently investigated in asthmatic children in Qatar and Kuwait,<sup>9,10</sup> and the use of burnt incense has been reported as a risk factor for obstructive lung disease in non-smoking Saudi women.<sup>6</sup> Our data showed for the first time that Arabian incense smoke exposure has histological effects in the interstitial septa and in the vascular tissues in the rat lung.

The results of this study clearly demonstrate that Arabian incense exposure produced sustained pulmonary inflammation and the appearance of lymphocyte aggregates at sites of inflammation. As a general rule, alveolar lymphocytes are not activated in normal lung. Similar data has been reported previously with cigarette smoking in humans and laboratory animals,<sup>11</sup> Our data confirms that smoke exposure produced lung lesions that included increased numbers of pigmented alveolar macrophages distributed throughout the parenchyma, and focal collections of enlarged alveolar macrophages with concomitant alveolar epithelial hyperplasia and neutrophilic alveolitis. In addition, our data showed that the bronchioles were thickened and infiltrated (Figure 4), with a marked increase in the number of inflammatory cells such as lymphocytes, histiocytes and neutrophils surrounding the alveolar walls. The thickness of alveolar walls deformed by the accumulation of inflammatory cells in the interstitium, interstitial and the lager alveoli was paralleled by large increases in the volume of collagen, which relates to stress-bearing in both the alveolar septa and alveolar ducts.<sup>12</sup> Moreover, the increase in the number of inflammatory cells has been reported as alveolitis, which refers to the cell types that are most abundant. Alveolitis results in the derangement of the alveolar-capillary units, resulting in their dysfunction and eventual loss as gas exchange units. This effect has been reported as an effect of the reduction of monocyte and macrophage mitotic activities against the background of intraalveolar lymphocytes in alveolitis.<sup>13,14</sup> In conclusion, our data confirm that exposure to Arabian incense (Bokhour) decreases the ability of the lungs to clear inhaled materials. We suggest that the Wistar albino rat constitutes a useful model for studies of the effects of Arabian incense smoke on lung histopathology.

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