Enhanced Translocation of Particles from Lungs by Jaggery

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Because industrial workers in dusty or smoky environments seemed to experience no discomfort if they consumed the sugar cane product jaggery, experimental studies were undertaken to observe the effects of jaggery on dust-exposed rats. Rats with and without a single intratracheal instillation of coal dust (50 mg/rat) were orally gavaged with jaggery (0.5 g/rat, 5 days/week for 90 days). The enhanced translocation of coal particles from lungs to tracheobronchial lymph nodes was observed in jaggery-treated rats. Moreover, the jaggery reduced the coal-induced histological lesions and hydroxyproline contents of lungs. The lesions induced in omental tissue and regional lymph nodes by a single intraperitoneal injection of 50 mg each of coal and silica dust were modified by jaggery (0.5 g/rat, 5 days/week for 30 days). These findings along with the preventive action of jaggery on smoke-induced lung lesions suggest the potential of jaggery as protective agent for workers in dusty and smoky environments. — Environ Health Perspect 102(Suppl 5):211–214 (1994)

Key words: clearance, translocation, lung, lymph nodes, omentum, silica, coal, jaggery, rat

Introduction

The quality of human life has improved in the last 50 years because of rapid industrialization by exploitation of natural resources for metals and minerals. However, the already existing problems of lung diseases in the mining and related industries and adverse effects of environmental hazards have created a double burden on industrial workers in developing countries. Lung diseases caused by biopersistence of natural minerals and synthetic fibers are difficult both to prevent and to cure. Moreover, total protection of occupationally-exposed workers from inhalation of particles is not yet possible and continued attempts must be made to find effective methods of prevention and possible treatment.

It has been observed that Indian industrial workers who consumed jaggery while working in dusty or smoky environment suffered no discomfort. In the ancient Indian Ayurvedic system of medicine, jaggery was considered to have beneficial effects in certain lung diseases. In recent years there have been a number of reports on the role of carbohydrates in the control of various normal and pathological processes and in the characterization of various antibiotics and antitumour agents (*1,2*). Sugar and carbohydrate are a main source of energy and can be beneficial without producing any adverse effects. Since Indian jaggery contains many important nutritive constituents that may be beneficial, studies were undertaken to observe the effects of jaggery on dust-exposed rats.

Materials and Methods

Sugar cane was introduced in India circa 6000 B.C. (*3*) and the products of sugar cane juice are still consumed in India. Jaggery is a solidified product obtained on boiling and concentrating sugar cane juice (*Saccharum sinense* roxb.) in an open pan. It is produced in all parts of the country and is consumed as such or in confectionery. Jaggery contains all the soluble constituents of sugar cane, which are given in detail in Table 1 (*4*).

Coal and Silica Dust

Coal and silica dust of respirable size (5 μm diameter) and of defined chemical composition, was obtained from the Environmental Monitoring Section of the Industrial Toxicology Research Centre. Coal mine dust was supplemented with silica dust up to 10% of the total mixture to increase fibrogenic potential.

Experiment 1

Four groups of 22 adult male albino rats, body weight 140 to 150 g, were maintained on a standard pellet diet and water *ad libitum*. Group 1, controls, received no treatment whatsoever. Group 2 received an intratracheal injection of 50 mg of coal dust suspended in 1 ml of 0.15 M NaCl solution. Group 3 received 0.5 g of jaggery orally in 1 ml of sterile distilled water 5 days per week, and Group 4 received both the injection of coal dust and the daily administration of jaggery.

Table 1. General composition of Indian jaggery.

Content	Value, range
Carbohydrate, %	83.5–95.0
Sucrose	72.8–80.3
Reducing sugar	6.8–14.2
Minerals, %	0.6–2.6
Calcium	0.2–0.36
Chloride	0.2–0.34
Phosphorus	0.03–0.22
Potassium	0.10–0.16
Sodium	0.006–0.025
Iron	0.005–0.020
Magnesium	0.008–0.105
Copper	0.007–0.010
Cobalt, nickel and molybdenum	0.001–0.008
Protein, %	0.35–0.40
Nonprotein nitrogen (mg/100 g)	19.6–42.9
Protein nitrogen (mg/100 g)	13.7–17.6
Vitamins, mg/100 g	
Thiamin	0.018–0.030
Riboflavin	0.042–0.046
Nicotinic acid	3.92–4.50
Vitamin C	5.20–30.00
Carotene, μg/100 g	155.0–168.0
Phenolics, mg/100 g	280.0–320.0
Fat, wax, pectin and organic acids, %	0.10–0.60
Moisture, %	3.9–7.2

Experiment 2

In the second experiment five groups of 10 rats each were taken. The first group (5) were controls. Group 6 received an intraperitoneal injection of 50 mg coal dust

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Table 2. Experimental protocol.

Group	Number of rats used	Route of administration of dust particles	Treatment: Dust particle	Treatment: Jaggery[a]	Period of killing
Experiment 1					
1	22	–	–	–	30, 60, and 90 days
2	22	IT	Coal[b]	–	
3	22	–	–	+	
4	22	IT	Coal[b]	+	
Experiment 2					
5	10	–	–	–	30 days
6	10	IP	Coal[b]	–	
7	10	IP	Coal[b]	+	
8	10	IP	Silica[c]	–	
9	10	IP	Silica[c]	+	

[a]Jaggery, 0.5 g/1 ml in sterile distilled water given orally 5 days/week. [b]Coal dust, 50 mg/1 ml of 0.15 M NaCl solution, sterile suspension was injected intratracheally (IT) or intraperitoneally (IP). [c]Silica dust, 50 mg/1 ml of 0.15 M NaCl solution, sterile suspension was injected (IP).

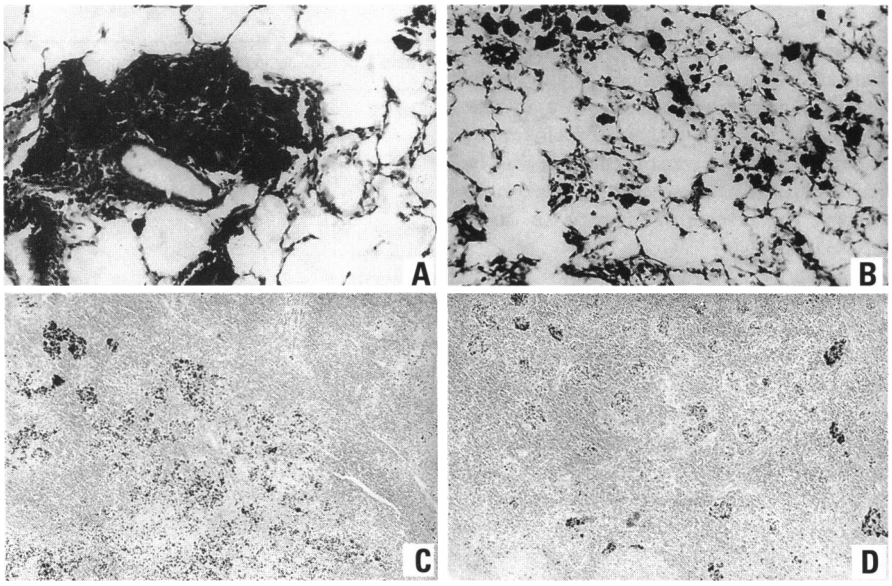

Figure 1. The effect of jaggery on coal-induced lesions in lungs and lymph nodes at 90 days. (*A*),Typical coal collection in lung (Group 2); (*B*), diffuse pattern of particles with less fibroblastic effects in Group 4 (coal and jaggery); (*C*), translocation of coal in lymph node, Group 2; (*D*), focal collection of dust in node, Group 4. All Hematoxylin and eosin stained; magnification: A, ×140, B, ×128, C and D, ×70.

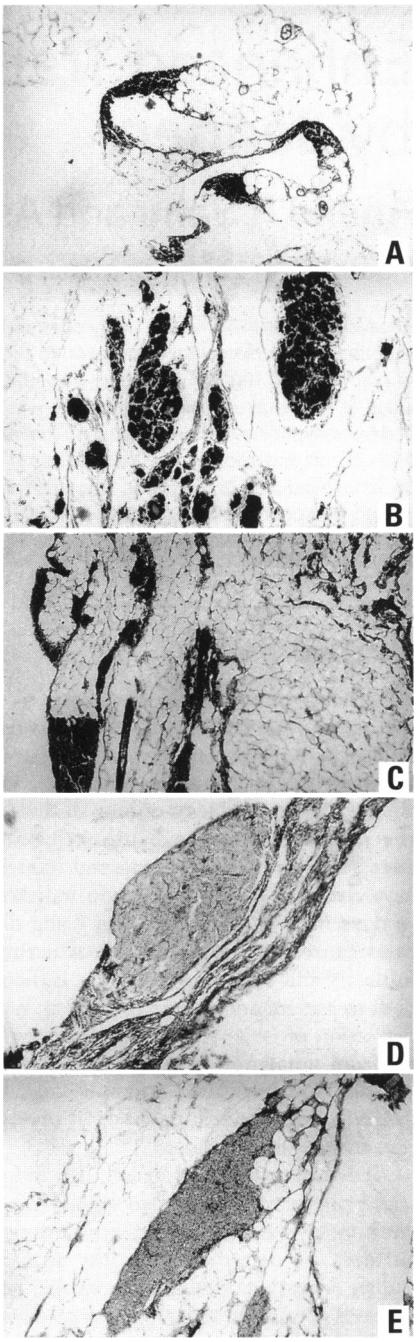

Figure 2. Omentum at 30 days after intraperitoneal injection of dusts. (*A*), Normal control (Group 5); (*B*), collection of coal dust (Group 6); (*C*), change pattern of effect in Group 7 (coal and jaggery); (*D*), a typical nodule induced by silica, Group 8; E, reduced effect in omentum, Group 9 (silica and jaggery). All H & E stained and magnified ×70.

alone; and Group 7 received coal dust and jaggery. Group 8 received silica dust alone and Group 9, silica dust and jaggery. The two protocols are summarized in Table 2. The dose of jaggery per rat was calculated on the basis of an average consumption of jaggery by workers.

Following treatment, the animals were killed at 30, 60, and 90 days in experiment 1 and at 30 days in experiment 2; autopsies were performed. The omentum and tracheobronchial lymph nodes (TLN) were excised carefully and fixed in Bouin's fluid. The lungs were inflated with 10% formal saline before being fixed. Normal histopathological techniques were used for tissues studied in these experiments. Lung collagen content was estimated by measuring hydroxyproline (HP) levels as described earlier (*5,6*).

Results

Experiment 1

The histopathological findings in Group 1 (controls), showed no significant changes. Group 2 (coal alone) 30 days after intratracheal injection showed a mild reaction in the form of phagocytosis with diffuse distribution of coal particles in the lung. Coal-laden macrophages and particles were widely observed in the alveoli. After 60 days, cytotoxic and fibrogenic reactions were observed in the form of proliferation and collection of dust cells and thickening

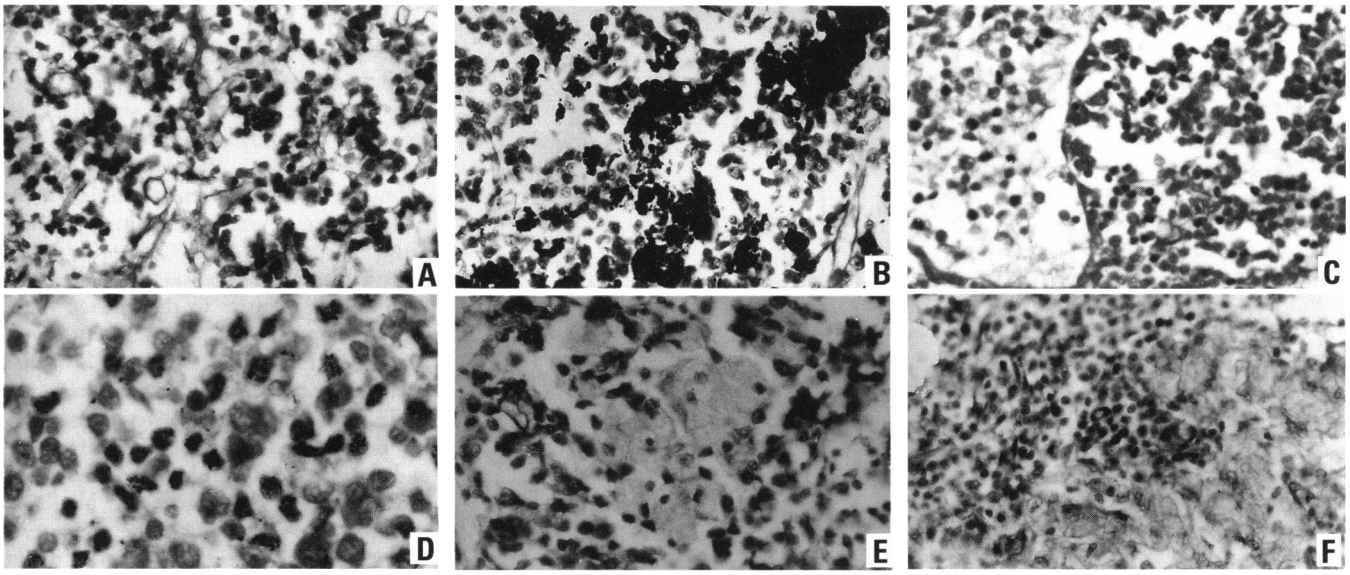

Figure 3. Tracheobronchial lymph node at 30 days. (*A*), Normal control (Group 5); (*B*), collection of coal particles, Group 6; (*C*), translocation of coal particles by phagocytosis along with cell necrosis, Group 7; (*D*), higher view of 3C; (*E*), deposition of silica along with fibroblastic reaction, Group 8; (*F*), reduced fibroblastic reaction in Group 9, All H & E stained; magnification A, B, C, E and F, × 560 and D × 1,400.

of alveolar septa. By 90 days the typical coal-induced focal areas of fibrosis along with dust particles were seen (Figure 1A). Upon silver impregnation, thick reticulin fibers were observed. Lungs of Group 3 (jaggery alone) did not reveal any histopathological changes at 30, 60, or 90 days. In Group 4 (coal and jaggery) the initial reaction was similar to that observed in Group 2 at 30 days except that proliferation of macrophages and the dust cell reaction were not seen. At 60 days, the lumen of respiratory and terminal bronchioles were filled with small to large aggregates of coal particles. This indicated possible physiological clearance through the mucociliary pathway. In peripheral alveoli, moderate phagocytosis of coal particles by macrophages were observed. At 90 days the widely scattered and partly unphagocytized coal particles with minimal cellular reaction and fibrogenic response were seen (Figure 1B).

TLN of Group 1 did not show any histopathological changes. In Group 2 at 30 days the TLN were enlarged with marked cellular reaction including hyperplasia of reticular cells. The intensity of coal accumulation was further increased by 60 days. Enhanced fibroblastic activity in the areas of coal dust localization and degeneration and necrosis of cells were observed. At 90 days, the TLN were enlarged with diffuse distribution of coal particles predominantly in the paracortical region with little focal collection in the cortex and medulla (Figure 1C). Marked fibroblastic reaction was seen in these areas. The TLN of Group 3 (jaggery alone) showed hyperplasia of germinal centers in the cortical region with increased mitotic activity and increased population of plasma cell series in the medullary cords. In Group 4 (coal dust and jaggery) the intensity of coal particles transported from lungs to lymph nodes increased progressively from 30 to 60 to 90 days. Minimal reaction was provoked by coal aggregates that were focally distributed (Figure 1D). Focal areas of coal aggregates did not reveal any significant fibroblastic reaction.

Experiment 2

The omental tissue of control rats (Group 5) did not show pathological changes (Figure 2A). The coal-induced lesions in the omentum (Group 6) revealed large dust foci having a central area of coal particles surrounded by the dust-laden macrophages along with lymphocytes. The fibroblastic activity continued at the periphery of the dust mass (Figure 2B). In the jaggery-treated group (Group 7), the clearance of coal particles from the peritoneal cavity to regional lymph nodes and in TLN was observed. The fibroblastic reaction to omental tissue was not as marked in Group 6, treated with coal alone (Figure 2C). The effect of silica (Group 8) on omental tissue showed a very severe fibroblastic reaction around the dust mass together with marked accumulation of lymphocytes at the periphery (Figure 2D). The omental tissue of the silica and jaggery treated group (Group 9) showed less fibroblastic reaction than to silica alone (Group 8) (Figure 2E).

The TLN in Group 5 (control rats) did not reveal any change of significance (Figure 3A). In Group 6 (coal alone) TLN showed heavy collection of coal dust in the paracortical region and phagocytized cells were seen around the dust mass. In Group 7 (coal and jaggery) TLN showed the presence of coal particles in the phagocytized cells together with cell necrosis. No dust mass was observed in the lymph nodes (Figure 3C,D). The well marked fibroblastic reaction was observed in the TLN in Group 8 (silica alone; Figure 3E), while less fibroblastic reaction was observed in Group 9 (silica and jaggery) (Figure 3F).

Lung Collagen

In experiment 1 the HP content of lung was measured at all the time periods (30, 60, and 90 days) (Figure 4). In Group 2 the increase in the HP content was from 16% at 30 days ($p < 0.001$) to 71% at 90 days ($p < 0.001$) in comparison to controls (Group 1). The HP content in the lungs of Group 3 did not show any change. Group 4 (coal and jaggery) showed a slight increase in HP content at 30 days, with nearly 10% increase ($p < 0.01$) at 60 and 90 days.

Discussion

The principal defense mechanisms that prevent the lung disease caused by deposition of inorganic dust particles are clearance by mucociliary processes in conducting airways and transport of particles

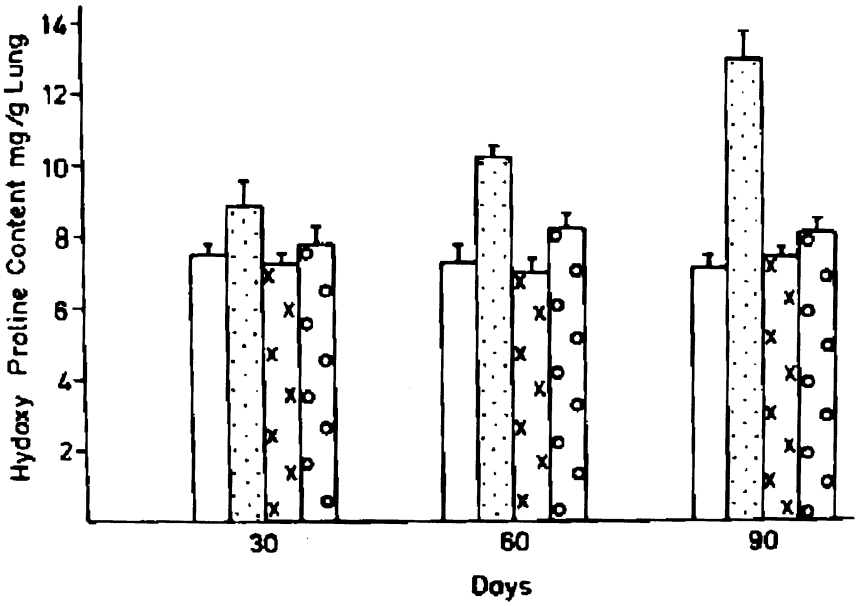

Figure 4. Hydroxyproline content of rat lung in Groups 1 to 4. Plain bar, control (Group 1); dotted bar, coal (Group 2); "X" bar, jaggery (Group 3), and circle bar, coal and jaggery (Group 4).

from lung to regional tracheobronchial lymph nodes (TLN). The mucociliary clearance from the tracheobronchial region usually is completed within 24 hr (*7*). The second step of clearance from alveolar region is through phagocytosis by alveolar macrophages and then removal by mucociliary clearance. Particle translocation from the alveolar region also involves the lymphatic system (*8*). Particles from the alveolar region are moved either by alveolar macrophages or by themselves via an unknown mechanism to the lymphatic channel in the alveolar wall. These particles are transported away from the alveolar region within the lymphatic channel and eventually are trapped in lymph nodes. The fate of particles trapped in lymph nodes is unknown, although some particles can move from a lymph node to another organ (*9*).

In the present studies, treatment with jaggery activated both defense mechanisms of the lungs. In mucociliary clearance of dust particles, the deposition of coal particles was observed in the conducting airways from terminal bronchioles to bronchi in Group 4 (coal and jaggery). Sialic acids are among the major constituents of mucin secreted in the respiratory tract. In the bronchoalveolar lavage of jaggery-treated rats, the sialic acid content was increased two-fold in comparison to normal rats (unpublished data). The sialic acid content of mucin molecules is responsible for the high viscosity of mucin, and imparts a negative charge of varying density (*10,11*). This attracts the positively charged respirable dust particles to the mucus, where they are trapped prior to clearance from the lung (*12*). Enhanced translocation of dust particles from lungs and peritoneal cavity to TLN was observed. However, the TLN are the site of immune-cell proliferation, and the enhanced translocation of particles following jaggery treatment may be due to the induction of some immune response. A recent report on the effect of dietary intake of fruits and vegetables on the risk of lung cancer among Yunnan tin miners demonstrated a significant protective effect of dietary habits (*13*). This supports the views presented in this study, of the protective effects of jaggery against the risks entailed in occupational exposure to dusts and fibers.

Protein is essential for fibrogenesis (*14,15*) but malnutrition and protein deficiency do not modulate silicotic fibrogenesis (*16*). The effect of carbohydrates on fibrogenesis is not as well studied as the effects related to nucleic acids or proteins, but it does appear that jaggery and its constituents are capable of enhancing the defense mechanisms of the lungs and protecting them against lesions induced by dust particles.

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