Teratogenic effects of nicotine on rat skin

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Summary

Beckground: Nicotine is a well-known toxic alkaloid substance with several teratogenic effects. In animal studies it has been observed that nicotine led to intrauterine growth retardation and intrauterine growth abnormalities including anancephaly, neonatal death and low birth weight. However, the teratogenic effects of nicotine have not previously been observed on skin.

Objective: We performed a study in order to observe histologically the teratogenic effects of nicotine on rat skin.

Materials & Methods: Ten female Wistar-albino rats were separated into two groups, a control and an experimental group (n=5). After the first week of pregnancy, the experimental group of rats were given nicotine intraperitoneally in a dosage of 2 mg/kg for two weeks.

Results: Striking teratogenic effects were observed in the experimental group of neonatal rats. Increased mitotic activity was noticed in the basal cells and hypertrophic epithelial cells were prominent in the epidermis. Chronic inflammatory cell infiltrate, fibrosis and extravasation of the erythrocytes were found in the dermis and hair follicles.

Conclusion: Considerable teratogenic effects of nicotine were observed histologically on newborn rat skin.

Key words: Newborn rat; Nicotine; Skin; Teratology.

Introduction

Nicotine is a liquid alkaloid obtained from the dried leaves of the tobacco plant, Nicotiana tobacum. It is a highly toxic substance with several well-known teratogenic effects. Maternal smoking in pregnancy is associated with low birth-weight infants and increased risk of abortion, stillbirth and neonatal death. In animal studies, it has been observed that nicotine administration led to intrauterine growth retardation and intrauterine growth abnormalities including anancephaly, neonatal death and low birth weight [1-3]. However, the teratogenic effect of nicotine has not been observed on skin previously. The purpose of the present study was to assess histologically the teratogenic effects of nicotine on rat skin.

Materials and Methods

Ten female Wistar-albino rats (200-220 g.) were caged and fed standard pellet food during the study. The rats were obtained from the Department of Medical Science Application and Research Centre of Dicle University. The female rats were confined in a special cage over 48 hours for copulation with adult males. After confirming pregnancy with the vaginal smear method, the primipara rats were separated into two groups (n=5), a control group and an experimental group.

After the first week of pregnancy, the experimental gravids were given nicotine intraperitoneally (i.p.) in a dosage of 2 mg/kg (in saline) for two weeks. During nicotine administration, all experimental gravids were examined daily by a radiologist using ultrasonography (US). A real time US (Toshiba SSA-270A) and 7.5 MHz linear transducer were used to detect the cardiac activation and to count the number of fetuses.

The gravid rats gave birth during the 21st and 22nd days of gestation. Immediately after, all litters were weighed and their lengths were measured. The litters were then anesthetised and several biopsies were taken and fixed in a solution of 10% formaldehyde. The tissues were then embedded in paraffin wax, sectioned and stained with hematoxylin-eosin (H&E). Histological assessments were performed using a light microscope.

Results

The experimental group of gravid rats showed some signs of distress and irritability including hyperactivity and anger immediately after nicotine administration. There were no resorbed fetuses or stillborns during nicotine administration. The average number of litters was the same in the control and exposed dams (Table 1). The mean weight of the experimental litters was found to be lower than the control group. However, the mean length of the experimental litters was identical to the control group of litters (Table 2).

There were no macroscopical abnormalities in either the experimental group or the control group of litters. However, abnormal microscopical findings were striking on experimental newborn rat skin. Under microscopic examination, normal epidermis and dermis were observed in the control group of neonatal rats (Figure 1). Increased mitotic activity was noted in the basal cells and hypertrophic epithelial cells were prominent in the epidermis of the experimental neonatal rats. In the dermal papillae, increased fibrosis and chronic mixed inflammatory cell infiltrate were noted in the same group (Figure 2). In the mid dermis, extravasation of erythrocytes was striking and the same type of inflammatory cell infiltrate was also observed around the hair follicles (Figure 3).

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Table 1. — *Number of births in the control and experimental groups.*

Indices (Dam rats)	Number of litters Control group	Number of litters (Experimental group)
1	9	8
2	7	9
3	10	9
4	10	8
5	9	10
Average	9	9

Table 2. — *Mean weight and length of the litters in the control and experimental groups.*

Indices (Dam rats)	Weight/Length Mean (Control group)	Weight/Length Mean (Experimental group)
1	6.09 g/4.7 cm	5.88 g/4.5 cm
2	6.36 g/4.4 cm	5.88 g/4.6 cm
3	6.11 g/4.6 cm	5.76 g/4.2 cm
4	6.03 g/4.4 cm	5.92 g/4.4 cm
5	6.05 g/4.2 cm	5.82 g/4.3 cm
Total Average	6.15 g/4.4 cm	5.85 g/4.4 cm

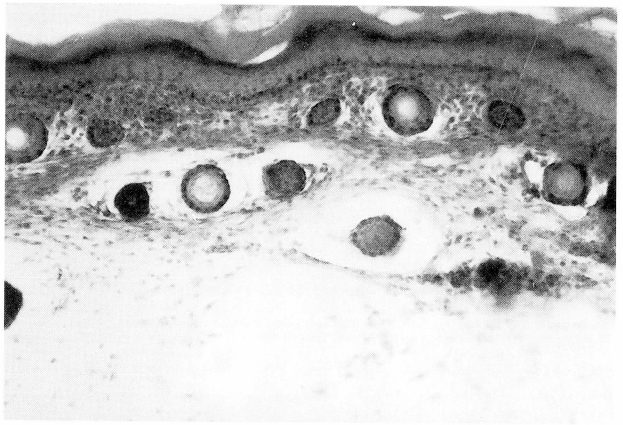
Discussion

In several human and animal studies, the teratogenic effects of nicotine administration during organogenesis have been investigated [2-4]. It was found that nicotine led to intrauterine growth retardation and intrauterine growth abnormalities including anacephaly, neonatal death and low birth weight. In our study, nicotine was also given during the organogenesis period (7 - 20 days) and low-birth weight and impaired skin maturation of rat skin were observed (Table 2 & Figures 2, 3).

Although teratogenic effects of nicotine have been observed in several studies, the pathogenesis has not been well documented. It has been suggested that the vasoconstriction of the placental vessels was the main pathology which led to a decrease in blood flow during pregnancy. Furthermore, increased blood pressure and palpitations were observed after intravenous nicotine administration in animals [5].

Among smokers, vasoconstriction and decreased capillary blood flow were found to play a major role in delayed wound healing, autonomic neuropathy in diabetics and ischemic necrosis of the skin flaps [6-9]. In these studies the vasoconstrictor effect of nicotine caused a reduction in nutritional blood flow to the skin resulting in tissue ischemia and impaired healing of injured tissue.

Nicotine was also found to increase platelet adhesiveness, raising the risk of thrombotic microvascular occlusion and tissue ischemia. In addition, proliferation of red blood cells, fibroblasts, and macrophages were reduced by nicotine. In a case report, leucocytoclastic vasculitis in association with the use of a nicotine patch was diagnosed after a skin biopsy [10]. Although we could not observe any obvious vasculitic damage in our study, extravasated erythrocytes were striking histologically in the experimental group of neonatal rats (Figure 3).



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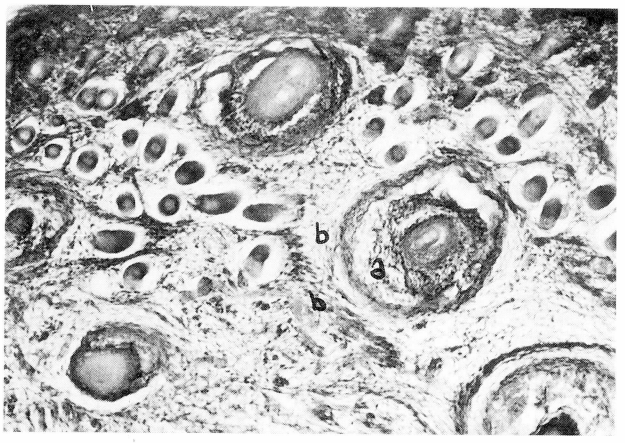

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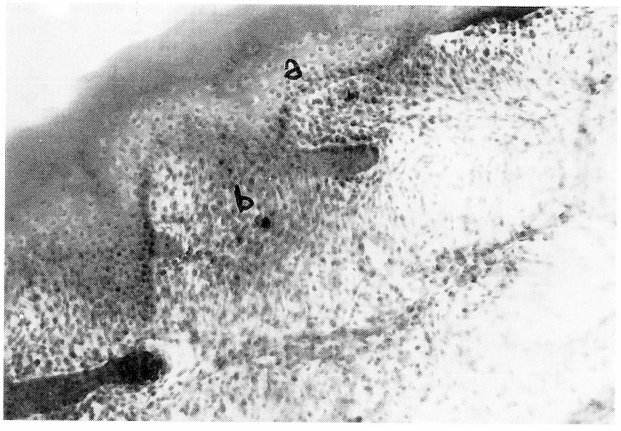

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Figure 1. — In the control group of neonatal rats, normal appearance of the epidermis and dermis (H&E, original magnification x 41).
Figure 2. — In the experimental group of neonatal rats, hypertrophic epithelial cells (a) and increased fibroblasts (b) in the dermal papillae (H&E, original magnification x 41).
Figure 3. — In the experimental group of neonatal rats, lymphocytic chronic inflammatory cell infiltrate around the hair follicles (a) and extravasated erythrocytes (b) in the mid dermis (H&E, original magnification x 41).

Nicotine has a low molecular weight and is easily absorbed after inhalation by buccal, nasal mucosa, alveoles and also by skin contact. In a case report of a patient who developed nicotine poisoning after cutaneous application of nicotine sulphate, a prolonged absorption of

nicotine was observed in the blood despite vigorous skin decontamination. It was suggested that the skin may be a reservoir for slow release of nicotine into the circulation [11]. Although nicotine treatment was found to be beneficial in pyoderma gangrenosum and dermatitis due to fluorouracil topical treatment [12, 13], we observed histologically that nicotine had several teratogenic effects on skin maturation in rats.

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