Topical Minoxidil Does Not Act as an Antiandrogen in the Flank Organ of the Golden Syrian Hamster

Barbara A. Nuck; Stephen L. Fogelson; Anne W. Lucky, MD

• **Little is known about the mechanism of action of minoxidil-induced hair growth in male pattern baldness. We studied the potential antiandrogenic effect of topical minoxidil administered at the same dose and in the same vehicle that has been used successfully clinically in human subjects on the androgen-dependent cutaneous structures of the flank organ of the golden Syrian hamster. Minoxidil applied topically to one flank organ had no androgenic effect. Neither 1% nor 5% minoxidil topically applied for three weeks prevented the androgen-dependent growth of the pigmented spot, the sebaceous gland, or the hair follicle diameter induced by subcutaneous Silastic capsules filled with crystalline testosterone. As a positive control in the same experiments, 5% progesterone did significantly inhibit pigment and sebaceous gland enlargement. We conclude that there is no antiandrogenic component to the mechanism of action of topical minoxidil in the hamster flank organ, and thus there is probably no antiandrogenic role in man either.**

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Minoxidil is a potent antihypertensive agent that causes hypertrichosis as a side effect. There is little known about the mechanism of action of minoxidil-induced hair growth.[1] Male pattern baldness is known to be an androgen-dependent phenomenon, and it has been reported that topically applied minoxidil solutions stimulated hair regrowth in balding patients.[2-4] Thus far, there have not been any laboratory studies assessing whether minoxidil-induced hair growth is mediated by an antiandrogenic mechanism. In this study, we tested the antiandrogenic potential of minoxidil in three androgen-dependent structures of the golden Syrian hamster skin: pigment, sebaceous glands, and hair follicles.

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From the Department of Dermatology, University of Cincinnati College of Medicine.

Reprint requests to Department of Dermatology, University of Cincinnati College of Medicine, 231 Bethesda Ave, Mail Location 592, Cincinnati, OH 45267 (Dr Lucky).

MATERIALS AND METHODS

Female golden Syrian hamsters (Charles River Laboratories, Wilmington, Mass) were used for this study. To provide continuous androgen stimulation, testosterone-filled capsules, 0.5 cm in length, were implanted subcutaneously in the ventral neck folds using a method described previously.[5]

The animals were randomly assigned to one of five groups listed in the Table. The vehicle solution consisted of ethanol, distilled water, and propylene glycol (7:2:1). Minoxidil powder (Upjohn Co, Kalamazoo, Mich) was dissolved in the vehicle in 1% (data not shown) and 5% concentrations, and 50 μL was topically applied to the right flank organ of each hamster daily for three weeks. The left flank organ served as a control. The antiandrogenic effect of minoxidil was assessed by comparing right with left flank organs re three androgen-dependent cutaneous structures: pigment, sebaceous glands, and hair follicles. As a positive control, 5% progesterone, a known antiandrogen, was applied topically in the same vehicle to the right flank organ of animals stimulated with testosterone.

At three weeks, the pigmented spot area was shaved, and the length and width were measured to the nearest 0.5 mm. The hamsters were then killed, the flank organs excised, and the tissue fixed in 10% formaldehyde solution for routine histologic sectioning and staining. After the flank organs were excised, they were flattened onto small squares of stiff paper to assure that they did not curl. In this position, they were then bisected at the largest diameter and embedded with the two halves in parallel.

Experimental Groups*		
Group (No. of Animals)	Testosterone Capsule, 0.5 cm	Topical Treatment of Right Flank Organ
1 (10)	. . .	. . .
2 (9)	+	Vehicle
3 (10)	. . .	5% minoxidil
4 (10)	+	5% minoxidil
5 (4)	+	5% progesterone

*Plus sign indicates treatment.

Thus, each paraffin section contained two halves from the center of the gland. Five such sections were measured, and the largest of each of these was considered to be the thickest and major portion of the gland. Sebaceous gland surface area and hair follicle diameter were determined on the histologic sections. Using a computerized digitizing tablet (Zidas, Carl Zeiss Inc, Thornwood, NY) with a microscope extension tube (Zeiss), the image seen in the microscope was traced by an electronic diode cursor connected to the tablet, and the measurements were recorded.

To assess pigmentation, surface area index (SAI) was calculated as the product of length times width for each oval pigmented spot. The maximal SAI of the area of the sebaceous gland was determined by outlining the individual lobules of the five centermost histologic sections. The total sebaceous gland area for each section was calculated by the computerized digitizing tablet. To assess hair follicle diameter, the hairs under the sebaceous gland were counted and measured. The perimeter of the hair shaft was traced, and the minimum diameter was calculated and recorded. Statistical analyses were performed on a computer (Clinfo) in the General Clinical Research Center at the University of Cincinnati College of Medicine. Student's *t* test or the Wilcoxon rank-sum test were used to assess the differences between group means.

RESULTS

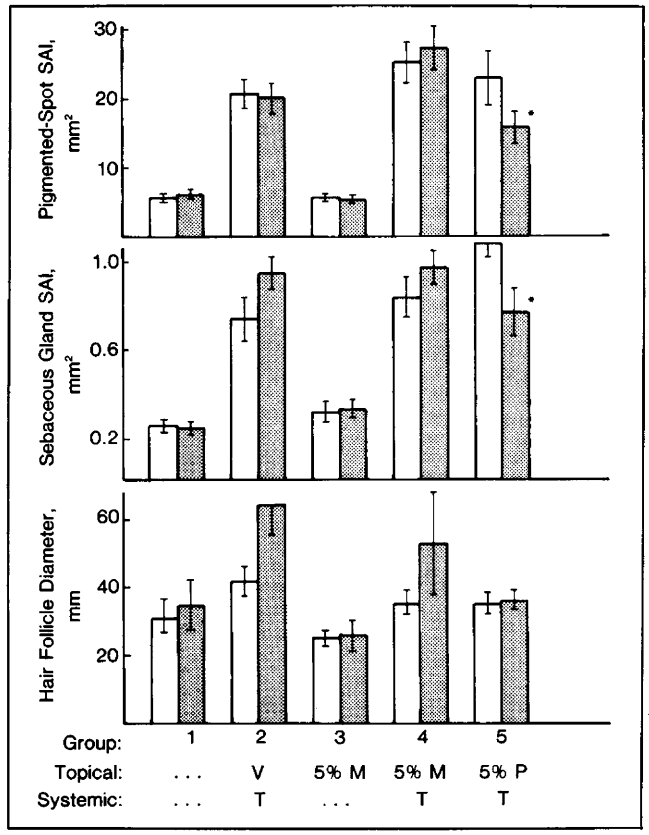
For all three skin parameters studied, testosterone caused significant growth compared with untreated flank organs (Figure). There was no difference between right and left flank organs in the control and vehicle-treated animals.

Pigmentation

The top portion of the Figure shows that the SAI of the pigmented spot of the right flank organs of the animals treated with 5% minoxidil alone (group 3) were not significantly different from those of the control group, which received a blank Silastic capsule (group 1), or from the left flank organ. One percent (data not shown) or 5% minoxidil applied topically (group 4) did not affect the growth of the pigmented spot in testosterone-stimulated animals.

Sebaceous Glands

Topically applied 5% minoxidil alone did not have an inhibitory or stimulatory effect on the sebaceous gland SAI (Figure, center). Although the mean values of the SAI appeared larger than in the untreated flank organ, 5% minoxidil-treated glands were not significantly different in size.



Response of pigmented spot (top), sebaceous gland (center), and hair follicle diameter (bottom) of flank organ of golden Syrian hamster to systemic stimulation with testosterone (T) and to topical inhibition by minoxidil (M) and progesterone (P). Systemic T significantly increased growth of all three structures compared with control (group one). Topical minoxidil applied to right flank organ (shaded) did not inhibit androgenic effect of T. Topical progesterone did inhibit pigment and sebaceous gland growth (asterisk indicates $P < .01$). Minoxidil alone had no androgenic effect. SAI indicates surface area index; V, vehicle.

Hair Follicle Diameter

Topically applied minoxidil alone had no effect on mean hair follicle diameter (Figure, bottom). Five percent minoxidil failed to inhibit testosterone stimulation of mean hair diameters in the flank organ.

Five percent progesterone (Figure, top and center) significantly inhibited the pigmented spot ($P < .01$) and the sebaceous gland SAI ($P < .01$) growth in the right flank organ (treated) when compared with the left flank organ. There was no significant effect on mean hair diameter (Figure, bottom).

COMMENT

The flank organ of the golden Syrian hamster has been used as a model to test topical antiandrogens.[5-8] We used this model to test the antiandrogenicity of 5% topical minoxidil and found no effect.

Patients taking therapeutic doses of minoxidil report a generalized increase in body hair. This observation encouraged dermatologists to test minoxidil for the treatment of hair loss. There is evidence that a 2% or 3% solution of minoxidil

applied topically stimulates hair regrowth in patients with male pattern baldness.[2-4] In other clinical trials, topical applications of minoxidil resulted in normal terminal hair regrowth at sites affected by severe alopecia areata.[1]

The precise mechanism of action by which minoxidil stimulates hair growth has not been determined.[1] Parker and Odell[9] performed in vitro and in vivo studies testing the possibility of an androgenic influence of minoxidil and other hypertrichosis-causing drugs. In vitro preparations of rat testis cells produced no stimulation of testosterone production when incubated with minoxidil. In vivo experiments showed no increase in steroid concentrations in the castrated dog adrenal gland after infusion of minoxidil intravenously. Receptor studies were also performed using tritiated testosterone and did not demonstrate any displacement of the radioactive label by minoxidil.[9]

We have examined the possibility of topical antiandrogenic activity of minoxidil. In our test system, minoxidil, in a 5% solution comparable with that that has been effective in male-pattern hair loss, had no inhibitory effect on the cutaneous androgen-dependent tissues of the hamster. Because of our small sample size, a small antiandrogenic effect might not have been detected in our study. However, we doubt that its hair-stimulating effect in man is mediated via an antiandrogenic mechanism. The mechanism of action of minoxidil, as yet undetermined, and the efficacy of this drug for cosmetic therapy deserve further study.

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