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COMPARISON OF TRICHOSCAN FINDINGS OF DIFFUSE HAIR LOSS IN FEMALES VERSUS CONTROLS

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Abstract

Introduction – Diffuse hair loss is a worrisome complain specially in females, commonly encountered in clinical dermatological practice. Trichoscan is a non-invasive method of hair analysis that is a combination of epiluminescencetrichoscopy and automatic image analysis system. In this study we analyzed 115 number of volunteer females.

Aim -To find association between clinical presentation and objective parameters found by trichoscan.

Method – All Participants went through detailed history taking, for grouping them under acute telogen effluvium, chronic telogen effluvium, pattern hair loss (PHL) and control group. On day 0, scalp hair of approx 1 cm2 area were shaved to leave an approx 0.5 mm hair stump on the area 1 cm lateral to midline on a line joining highest point of pinna. On day 3, patient would undergo hair pull test, trichoscopy and trichoscan study. Trichoscan parameters like anagen:telogen ratio, density/cm2,vellus and terminal hairs were considered for analysis.

Statistical analysis – Raw data was analysed using appropriate one way ANOVA or Kruskal-Wallis Test followed by appropriate post-hoc test.

Results – The mean hair density/cm2 is lowest in PHL group with having significant difference with control ($p<0.01$), chronic and acute TE group ($p<0.05$). Anagen ratio is lowest in acute TE followed by chronic TE, PHL and control group. The terminal hair ratio is significantly low in PHL group as compared to control, acute and chronic TE groups. ($p<0.001$)

Conclusion – Trichoscan is easy useful tool for female hair analysis and satisfactorily used in differentiating PHL, acute telogen effluvium and normal.

Key Words - Trichoscan, Acute telogen effluvium, Chronic telogen effluvium, Pattern hair loss (PHL)

Introduction

Diffuse hair loss is a very common complaint encountered in clinical dermatological practice which can be acute or chronic. Diffuse hair loss is a very common complaint encountered in clinical dermatological practice. It includes various conditions like acute and chronic telogen Though females are less prone to get bald naturally, it is found to be more troublesome among themit found more troublesome among them.. It is noticed that female seek medical advice more than men. Any non-patchy hair loss appreciable to patient can be considered as diffuse hair loss. Various pattern of diffuse hair loss seen in female are: telogen effluvium, anagen effluvium, patterned hair loss (androgenetic alopecia), diffuse alopecia areata and other shaft disorders.[1] Trichoscan is a unit containing epiluminescence surface microscopy and digital image analysis system.[2,3] It is recently developed, computer assisted non-invasive method of assessment of hair biological peculiarities that is explained as user-friendly, time-saving and reproducible method.[3,4]

The aim of our study was to find out if there is any association between clinical presentation and objective measurements of hair biological parameters determined by trichoscan method.

Subjects and Methods:

After approval from the Institutional Review Board,total of 115 females visiting the outpatient department of Dermatologyduring the period of one and half years were enrolled after written informed consent.We included the females between second and fifth age groups. Females with other dermatological scalp conditions like psoriasis, lichen planus, discoid lupus erythematosus, hair shaft abnormality and cicatricial alopecia as well as those who deny for consent were excluded from the study.All participants went through detailed history including associated dermatological and other systemic diseases and thorough scalp examination. This helped us furtherfor grouping them as-anagen effluvium, acute telogen effluvium, chronic telogen effluvium, patterned hair loss, diffuse alopecia areata and control group. But, we did not comeameacross cases of anagen effluvium and diffuse alopecia areata. Routine laboratory investigations like complete blood count,renal function test, liver function test, urine analysis, HbsAg, erythrocyte sedimentation rate and serum ferritin level were carried out. Thyroid function test and other respective investigations were done if required.

Scalp examination for trichoscan was carried out as belows:

On day one, scalp hairs of approximately1 cm^2 area were trimmed to leave an approximately 0.5 mm hair stump. The area was fixed as -one cm lateral to midline on a line joining highest points of both pinna in all subjects to maintain the uniformity. Gross photographs were captured. All participants were called back after 48 hours with an instruction not to wash their hair during this period. The site was re-examined by putting contact mode dermoscope (Heine Delta 20 Plus P set, Germany) with polarized light that is attached with digital camera (Canon EOS 550 D Camera, Tokyo, Japan). After proper alignment the image was captured, saved and analysed with trichoscan software. This programme analyses 1.195 cm^2 area and gives different hair parameters like density/cm^2,anagen hairs (%), telogen hairs (%), density vellus hairs (per cm^2), density terminal hairs (per cm^2), ratio vellus hairs (%) and ratio terminal hairs (%)(Figure 1 and 2).

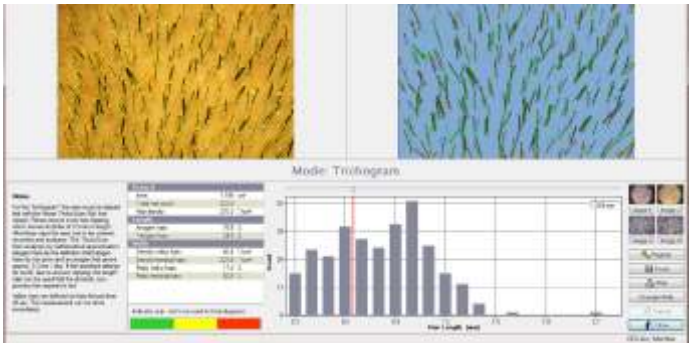

Figure 1: Example of the Trichoscan analysis of ATE

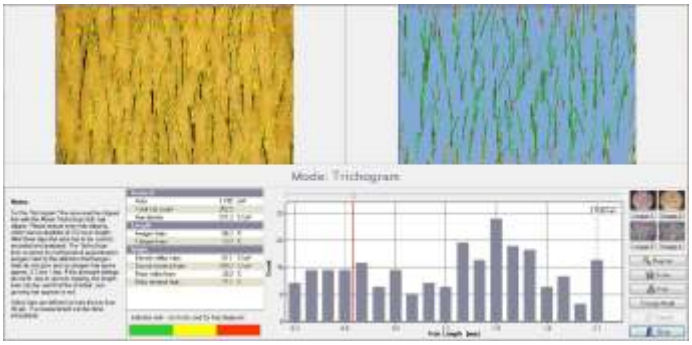

Figure 2: Example of the Trichoscan analysis of PHL

The principle of trichoscan is based on hair biologic characteristics like anagen hairs grows daily by approximately 0.3 mm while telogen hairs do not grow. Using this trichoscan counts, hairs with length of >0.7 mm wμm were consideredanagen and other non-growing hairs as telogen. This value of 0.7 mμm is manually adjustable. Black permanent hair colour (Streax hair colour) was used only in few persons with white hair to improve contrast between hair and scalp skin and only on the area to be examined with dermoscope. This was made possible by applyingmixture of cream and developer solution on examination area for 15 minutes in aproportion of 1:1 ratio with the help of a wooden spatula. Hair Pull Test(HPT)from area other than clipped hair was performed in all subjects. Pulled hairs were examined under microscope to confirm whether they areanagenor telogen hairs.Hair loss pattern werethen classified as: (1) Acute Telogen Effluvium (ATE) (2) Chronic Telogen Effluvium (CTE) (3) Patterned hair loss (PHL) and (4) Control.Patients coming for other dermatological conditions(excluding scalp), relatives accompanying the patients, hospital staff like nurse, medical students, technicians and other healthy volunteerswith normal hair cycle were included as controls.

Statistical analysis was done using Graph pad InStat software version 3.06. Raw data was put on normality test and data with Gaussian distribution like overall hair density and terminal hair density (per cm2) (Table 1) wereanalysed using oneway ANOVA while those with non-Gaussian distribution like anagen, telogen, vellus, terminal hair (%) and vellus hairs (per cm2) (Table 2) were analysed using Kruskal-Wallis test followed by appropriate post-hoc test. P value < 0.05 were accepted to be significant.

Groups → / Variables ↓	Group A Control (N=27)	Group B ATE (N=33)	Group C CTE (N=39)	Group D PHL (N=16)	P value (One Way ANOVA)
Hair Density (per cm^2)	277.9±51.8*	274.2±40.0†	269.9±44.6†	230.2±58.9	0.009
Terminal hairs (per cm^2)	222.1±38.8‡	220.2±33.1‡	217.8±35.2‡	167.0±39.9	<0.0001

* P< 0.01 as compared to group B† P < 0.05 and ‡ P < 0.001 as compared to group D using Tukey-Kramer multiple comparison test.

Table 1: Comparison of hair density and terminal hairs among different groups

Groups → / Variables ↓	Group A Control (N=27)	Group B ATE (N=33)	Group C CTE (N=39)	Group D PHL (N=16)	P value (Kruskal-Wallis Test)
Anagen (%)	82.6 (79.8-86.1)	75.0 (68.2-83.3)*	77.4 (73.7-82.9)	79.3 (76.8-82.2)	0.02
Telogen (%)	17.4 (13.9-20.2)	25.0 (16.7-31.8)*	22.6 (17.1-26.3)	20.8 (17.8-23.2)	0.02
Vellus hair (%)	19.1 (18.2-21.7)†	18.1 (16.8-21.9)†	18.9 (17.1-22.2)†	27.4 (24.0-29.7)	<0.0001
Terminal hairs (%)	80.9 (78.4-81.9)†	81.9 (78.1-83.2)†	81.1 (77.8-82.9)†	72.7 (70.3-76.0)	<0.0001
Vellus hairs (per cm^2)	53.5 (45.4-63.6)	50.2 (43.9-63.6)	52.3 (42.9-57.3)	58.8 (49.2-74.9)	0.3

*P < 0.05 as compared to group A; † P< 0.001 as compared to group D using Dunn's multiple comparison test.

Table 2: Comparison of Anagen, Telogen, Vellus and Terminal hairs among different groups

Results:

We enrolled total 115 females with age ranging between 13 to 50 years. The mean age of all with diffuse hair loss and control group were 28.9 ± 9.6 and 25.1 ± 7.6 years, respectively. The age-wise distribution was as follows: 35 (30.4%) in second decade, 39 (33.9%) in third decade, 27 (23.5%) in forth decade and 14 (12.2%) in fifth decade. We did not observe muchchanges in number of hairs per cm2, telogen hairs, vellus hairs or terminal hairs content with advancing age in any group.

In acute TE group, 45.5% cases showed positive hair pull test in whom duration of hair fall was less than or equal to two months. Specific triggering factors like febrile illness, major operative procedure and recovery from debilitating illness, etc. was identified from history only in 30.3% cases of acute TE and 17.9% cases of chronic TE.

Three out of 16 (18.8%) patients of PHL had positive family history of androgenetic alopecia. Only one participant of 16 from PHL group had clinical sign of hyperandrogenismcharacterised byexcessive hairs on upper lips and irregular menstruation.

The hair density was lowest in PHL group followed by CTE, ATE group and highest in control group with statistically significant difference between PHL and other three groups (Table 1). The control group showed highest anagen hair

content(%) while acute TE group showed lowest. The percentage of telogen hairs was highest in acute TE groupfollowed by the chronic TE and PHL group while lowest in control group.Terminal hair count was significantly lower infemales with PHL compared to other groups ($p < 0.000$). Vellus hair content was higher in PHL compared to other three groups though not significant ($p < 0.3$)(Table 2).

We reclassified all the females with diffuse hair loss in two groups based on Hair pull test results as Hair pull test positive and Hair pull test negative. There was no significant difference in mean values of different trichoscan parameters between two groups. Average Telogen hair count was slightly higher in females with positive HPT while average anagen hair count was slightly lower in HPT positive group (Table 3).

	Hair Density (per cm^2)	Anagen (%)	Telogen (%)	Vellus Hair (per cm^2)	Terminal Hair (per cm^2)
HPT Positive (N=27)	- -	75.9 ± 8.1	24.12 ± 7.9	55.21 ± 19.9	212.96 ± 35.69
HPT Negative (N=88)	267.32 ± 50.26	78.05 ± 7.9	21.95 ± 7.9	55.65 ± 18.2	212.23 ± 41.6

Table 3: Values of parameters between groups made as per Hair Pull Test results.

Serum ferritin level was done in 106 subjects. Low serum ferritin level (<11 ng/ml) was found in 40.6% (43) subjects. Eight out of 24 subjects in control group had low serum ferritin.

Discussion:

Diffuse hair loss is defined as acute or chronic generalised thinning of hairs from the scalp. It includes various conditions like acute and chronic telogen effluvium, anagen effluvium, pattern hair loss and diffuse alopecia areata. Of them, telogen effluvium is the most common encountered form of diffuse hair loss which is similar to our observation.[5]

It is difficult to monitor the therapy of hair loss. Previously, dermatologist used to measure the hair length and diameter manually, but it is very tedious and is subjected to subjective variation. Though, invasive procedure like scalp biopsy is standard for measuring the anagen: telogen ratio and for diagnosing hair disorders but in every follow up, patient would not get convinced for biopsy to monitor the therapy. Also patient would feel discomfort for hair pulling in semi invasive methods like trichogram and unit area trichogram. Other non invasive hair evaluation methods like global hair count, hair weight test and daily hair count are prone to manual errors and often results are not reproducible.[2,3,5]

So, a reliable,non invasive, safe, easy and OPD tool is required to assess the biological hair parameters.Trichoscan, introduced in 2001 is a novel, handy, non- invasive, reproducible, easy, validated and investigator independent automated image analyser which is a computer based video dermatoscopy which identifies anagen, telogen, terminal, vellus hairs and hair density.[3,5] One validation trial conducted proved excellent correlation between hair parameter results analysed by trichoscan and manually.[6]

There is no defined or fixed normal value for hair density as there are differences according to race and genetic constitution.In the review of various studies, Ru[sh]ton et al. demonstrated hair density as average 181 hairs/cm^2 by phototrichogram and 237 hairs/cm^2 by unit area trichogram in female subjects.[7] Saraogiet al., found hair density 291.3/cm^2 in normal control females and in study by Aktanet al., mean hair density of healthy volunteers was found to be 141.7 hairs/cm^2 by Trichoscan while it was 212.8 hairs/cm2 by photomacrographs.8,9 Birch et al., detected hair density of 293 hairs/cm^2 on using photomacrographs.10 In our study we found hair density of 277.9 ± 51.8 hairs/cm^2 in control group (Table 1, Figure 3).[3]

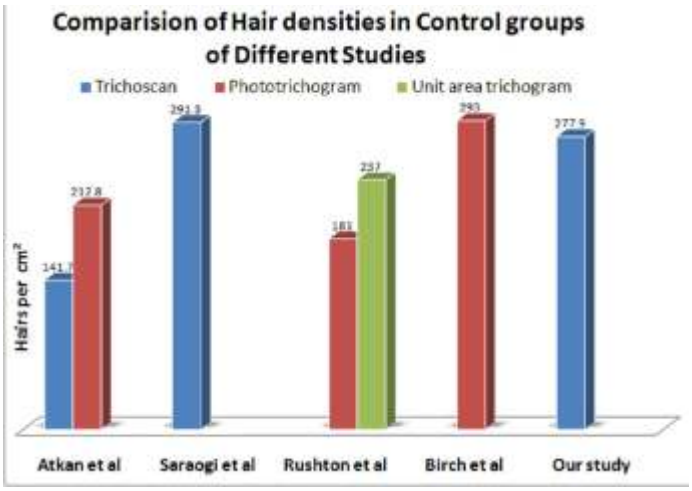

Figure 3: Bar chart showing comparison of hair densities in control groups of different studies

Acute telogen effluvium is defined as diffuse hair loss occurring from all over scalp around 6 weeks to 3 months of inciting event and have self-limiting course. If precipitating event is repeated or continued, hair loss may persist for more than 6 months and considered under chronic telogen effluvium. CTE was first described by Whiting DA in 1996 as idiopathic telogen hair loss primarily affecting middle-aged women, having long fluctuating course and diffuse hair shedding that may show bitemporal thinning.11Other secondary causes of chronic telogen shedding like thyroid dysfunction, iron deficiency anemia, acrodermatitisenteropathica and others were known.12,13In our study, ATE and CTE group did not show any statistically significant difference in trichoscan parameters. Hair density and terminal hair density (per cm^2) were higher in ATE group than CTE while terminal and vellus hair ratio wasfound to be almost similar (Table 1, 2).

A study by Haticeet al. showed statistically significant difference in the hair density and terminal hairs between the TE group and PHL.5 In our study also, we found similar results for hair density and terminal hairs ($p<0.05$ and <0.001 respectively) in the acute TE and PHL group. We observed slight difference of hair density between chronic TE and PHL group (Table 1). Thus, it can be said that clinical dilemma of differentiation of chronic telogen effluvium and patterned hair loss does not completely resolve with trichoscan.

Sinclair et al., in their using punch biopsy for differentiation between chronic telogen effluvium and PHL the ratio of terminal hairs to vellus hairs (T:V) >8:1 in CTE and 4:1 in PHL.14In contrast to these findings, in our study carried out by trichoscan, we calculated the Terminal : vellus hair ratio to be 4:1 in CTE group and 3:1 in PHL group (Table 2).

The trichoscopic features diagnostic for PHL are variation in hair shaft diameter due to miniaturization of hair follicle, peripilar sign, single hair bearing follicles, focal areas of atrichia and yellow dots while telogen effluvium is a diagnosis of exclusion (Figure 4).

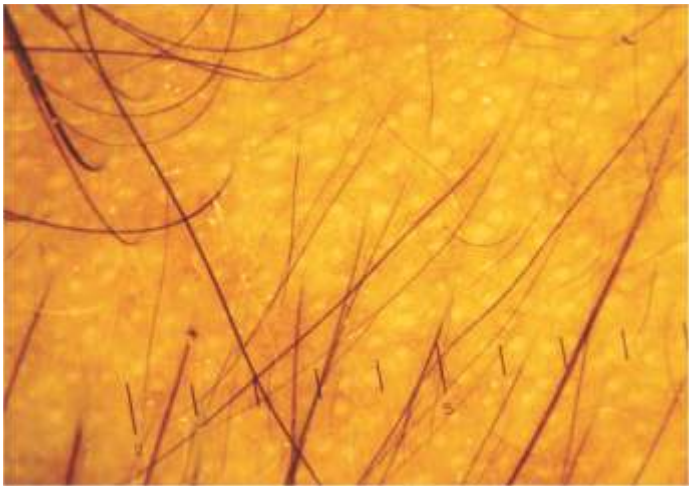

Figure 4: Dermoscopic features of Female androgenic alopecia: miniaturized hair follicles (arrow), variation in hair shaft diameter, single hair bearing follicles and yellow dots (arrow head).

Decreased hair density and follicular units with single hair in the absence of characteristic signs of other causes of alopecia is considered to support clinical diagnosis of telogen effluvium.[15,16] Scrutiny of trichoscopic images in our study lead to a perception that few images from telogen effluvium group showed thin hairs with equal diameter in contrast to PHL group where it showed thinned hairs with varyingdiameters.

Hair pull test which is used to determine disease activity is found to be positive during active shedding phase of both telogen effluvium and anagen effluvium.[17]Interesting finding noted in our study that the test is positive in cases of early androgenetic alopecia at the areas of thinningwhile negative at occipital area.There was no any statistically significant difference in telogen hairs (%) found between HPT positive PHL subjects and HPT negative PHL subjects in contradictory to that found by Haticeet al(Table 3).[5]

Earlier in a study by Van Nesteet al. with contrast-enhanced phototrichogram method, it was showed that increase in age and duration of complain were associated with decreased hair density in females with ludwig pattern hair loss.[18] Here, our observation showed no relation of hair density with patient's age and duration of hair loss unlike the inference made by Aktanet al.[9]

There are few studies which suggest relation between serum ferritin level and hair loss. Ideal ferritin level was considered to be at least40 ng/ml for healthy hair growth.[19] We found very low values of serum ferritin among cases and contols,all groups, where only 6.6% (7 out of 106) showed normal range. There was no any difference in the values of serum ferritin among study groups. No association between serum ferritin level and density of hairs per cm^2, anagen (%), telogen (%) and terminal hairs per cm^2 were noticed in our study.

Limitations

Limitations of the study design includes limited sample size for controls and PHL group.Our study is confined to a single centre. Few limitations experienced during procedure and the software are mentioned as below: 1) Crossing of two hair strands lead to false counting yielding higher telogen ratio 2) If the hair is cut very short, near to scalp surface it may not be detected by the software 3) Very thin hairs escape from the counting. These can be overcome by avoiding the processing of this kind of images. Similar errors are also mentioned by Saraogiet al.[8]

Conclusion

Trichoscan, being a non-invasive and simple office procedure, has a scope for use in routine OPD for evaluation of hair loss if the limitations of procedure are meticulously taken care off. It can be used satisfactorily in differentiating acute telogen effluvium, patterned hair loss and normal hairs. In cases of clinical conundrum, trichoscanalong with dermoscopic confirmation of condition will further allow clinician to manage patients with disease specific treatment and prognosis. Trichoscan as part of evaluation, gives psychological satisfaction to the patient which may lead to improved compliance with treatment prescribed by the physician.[3,5]

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