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Original Article

Anatomy Section

Analysis of Ratio of Length of 2nd Digit to 4th Digit (2D:4D) among Transgender Women (MtF) and Cisgender People in South Indian Population: A Cross-sectional Study

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ABSTRACT

Introduction: Transgender refers to an individual whose gender identity differs from the sex that was assigned at birth. Transgender women (MtF) were assigned male at birth but later recognised themselves as female. The length of the index finger (2D) in males is shorter than the length of the ring finger (4D), resulting in a low finger ratio due to higher androgen levels, whereas in females, the ratio is high due to low androgen levels. The index to ring finger length ratio in males and females is influenced by testosterone hormone levels during early foetal life, exhibiting sexual dimorphism. The 2D:4D ratio serves as one of the determinants for future generations to identify the transgender women population.

Aim: To determine the 2D:4D ratio among transgender women and the cisgender population.

Materials and Methods: A cross-sectional study was conducted in the Department of Anatomy, Annapoorana Medical College and Hospitals, Vinayaka Mission's Medical College Hospitals, Salem, Tamil Nadu, India, from September 2018 to October 2019. A total of 392 adult participants between 19 years and 60 years of age were selected. The samples of transwomen were collected with the help of the President of Salem Thirunangaigal Nala Sangam (STNS) and Koovagam Koothandavar Temple in Tamil Nadu. Cisgender samples were collected from volunteers in Salem, Tamil Nadu. The 2D and 4D digit lengths of both hands were measured using Digimizer software with the help of images obtained by a Canon 220 photo scanner. Another type of classification was done by naked eye analysis without any calculations, based on which the 2D:4D ratio among males, females and transgender women was categorised into types 1, 2 and 3. The paired t-test was used to compare the male, female and transwomen populations.

Results: Out of 392 participants, 122 were cis females, 130 were cis males and 140 were transgender females. The p-value of the mean 2D:4D ratio was found to be significant in the right and left hands when comparing the 2D:4D ratio of males with females and transgender women with females. The p-value of the 2D:4D ratio for males with transgender women in the right and left hands was 0.451 and 0.943, respectively, which was statistically insignificant due to the high levels of androgen in these two groups.

Conclusion: The present study helps scientists in the gender study field to advance their knowledge about transgender individuals, which can fill the gaps in available information and overcome existing lacunae in this field.

Keywords: Androgen, Digit ratio, Index finger, Sexual dimorphism

INTRODUCTION

Transgender studies have become increasingly prominent over the past several decades. The word "transgender" was coined by psychiatrist John F. Oliven of Columbia University in 1965. Transgender refers to an individual's sense of personal identity that does not conform to their anatomical sex. For gender non conformity, the term transgender can be used. The various terms commonly used under the transgender umbrella are (FtM) for men and (MtF) for women, where individuals change themselves according to what they feel inside [1,2]. Cisgender identity and gender, however, match with the sex at birth. Prenatal androgen exposure in foetuses plays a vital role in determining the digit ratio. Male foetuses show lower (masculine) digit ratios due to more androgen exposure, whereas in female foetuses, the possibility of androgen exposure is less reflected as a higher (feminine) digit ratio [3]. Males with a feminine digit ratio are more vulnerable to have low sperm counts, infertility and metabolic syndrome [4]. The length of the index and ring finger (2D:4D) ratio varies in males and females as prenatal androgen exposure determines the digit length before birth [5]. The 2D:4D ratio was found to be greater in females than males due to exposure to low androgen levels in utero, and the gender difference tends to be more for the right hand than the left hand as the majority of the population were right-handed individuals [6].

The present study not only helps scientists in transgender studies but also assists forensic experts in dealing with medicolegal cases to identify the gender of an unknown individual with residual body parts having only a detached upper limb by measuring the digit (2D:4D) ratio as it shows sexual dimorphism. Therefore the present study was aimed to determine gender differences in digit ratio (2D:4D) between transgender women and cisgender individuals in the South Indian population and to calculate the 2D:4D ratios, determine the percentage distribution among the three genders, and categorise them into types 1, 2 and 3.

MATERIALS AND METHODS

A cross-sectional study was conducted in the Department of Anatomy, Annapoorana Medical College and Hospitals, Vinayaka Mission's Medical College and Hospitals, from September 2018 to October 2019. The Institutional Ethics Committee (IEC) granted ethical clearance for the present study (VMKVMC/IEC/18/62).

Sample size calculation: To calculate the sample size, reference values were available for males and females only; for transgender individuals, the reference values were not available. Therefore, the sample size was calculated based on the effect size. To calculate the sample size for three groups, the assumed medium effect, i.e., E=0.20, was used. The sample size was calculated using the

software G Power version 3.1.9.2 with 90% power, resulting in 107 per group and a total of 321 [7]. A non response rate of 15% was considered. Therefore, the final sample size of 126 per group and a total of 378 was used.

Inclusion criteria: Cisgender (both male and female) volunteers between 19 years and 60 years and government Certified adult Transgender women (MtF) between the age group 19 years and 60 years were included in the study.

Exclusion criteria: Other genderqueers like transmen (FtM), gay, lesbian, intersex and queer, and transwomen who did not give informed consent were excluded from the study.

Study Procedure

A total of 392 adult samples between 19 years and 60 years of age were selected. Samples from cisgender subjects were collected from Department of Anatomy, Annapoorana Medical College and Hospitals, Tamil nadu, India (cis females, n=50 and cis males, n=39), Vinayaka Mission's Medical College and Hospitals, Tamil nadu, India (cis females, n=23 and cis males, n=31), and at Salem cis females, n=49 and cis males, n=60). All the samples of transwomen (n=140) were collected with the help of President STNS and Koovagam Koothandavar Temple in Tamil Nadu, India.

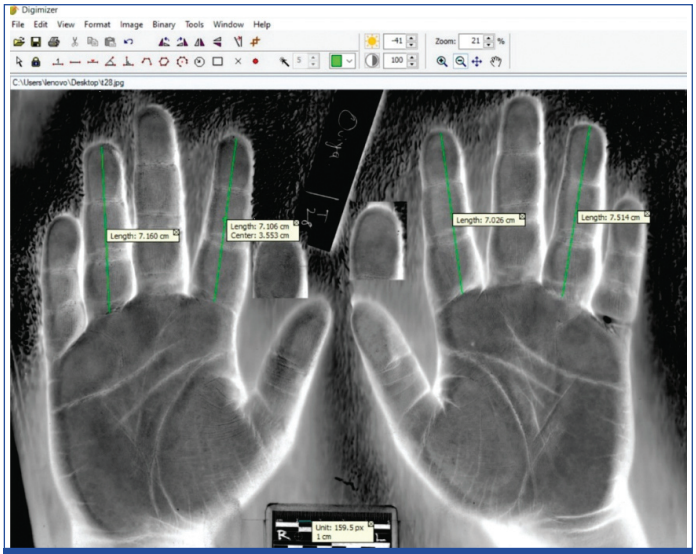
Subjects underwent multiple counselling sessions to help them understand the type of the research. Written informed consent was obtained from all interested participants. Out of 500 subjects approached, including cisgender and transgender women, only 392 subjects consented to participate.

Photo scanning image: All volunteers were asked to remove their ornamental jewellery from their fingers, and their hands were cleaned with soap and water to remove dirt. Once their hands were dry, a thin layer of non toxic white talcum powder was applied to the palms and fingers. The white powder enhances the ridges and creases of the hands for computer interpretation. A Canon CanoScan LiDe 220 photo scanner and a personal computer were used to record the image, ensuring that all fingers were straight and visible (A3). While scanning, a measurement scale, the subject's name and ID number were placed next to the subject's hand. Both the palmar sides, the subject's name with ID number and the measurement scale were scanned simultaneously (A1A2). The images were standardised by setting a 10 mm length to 157 pixels [Table/Fig-1].

[Table/Fig-1]: Scanning the palmar aspects of both right and left hands.

Software digimizer software: All the scanned images were labelled, converted to grayscale, and inverted to negative for better visualisation with the help of Photoshop 2018 software. Later, all the Joint Photographic Experts Group (JPEG) images were transferred to the Digimizer software for length measurements [Table/Fig-2] [8,9].

The following measurements were taken on both the right and left hands as specified below [10]:



[Table/Fig-2]: Shows 2D:4D measurements using Digimizer 4.3.2 software.

- D2 (Index finger) length was measured between the midpoint of the basal crease and the fingertip.
- D4 (Ring finger) length was measured between the midpoint of the proximal basal crease and the fingertip.

Based on the lengths of the index and ring fingers, they are categorised into three visual types [11]:

- Type 1: Length of the index finger longer than the ring finger.
- Type 2: Length of the index finger and ring finger equal.
- Type 3: Length of the ring finger longer than the index finger.

All the above measurements were taken thrice by two investigators, and the mean of the three values was considered as the final value. The readings were taken with 100% agreement by both observers.

STATISTICAL ANALYSIS

A detailed descriptive analysis was done by using Statistical Package for the Social Sciences (SPSS) software version 25.0. The mean, Standard Deviation (SD) and p-value were calculated and compared among cisgender males, cisgender females and transgender women. The paired t-test was used to compare the male, female and transgender women groups. The percentage distribution of each type based on the length of the index and ring fingers among the three genders was tabulated. A significance level of p-value <0.05 was considered significant.

RESULTS

Out of 392 participants, 122 were cis females, 130 were cis males and 140 were transgender females [Table/Fig-3]. The mean 2D:4D ratio in the right hand was comparatively greater than in the left hand in all three genders. The mean 2D:4D ratio of the right and left hands in females was higher than in the male and transgender women populations [Table/Fig-4].

Gender	Sample (n)	Age (years) (Mean±SD)
Cis female	122	36.7±11.5
Cis male	130	25.1±11.0
Transwomen	140	25.2±10.6
Total	392	28.8±12.2

[Table/Fig-3]: Mean and standard deviation age of the total population studied.

The mean 2D:4D ratio between females and males shows a p-value of 0.010 on the right-side and a p-value of 0.001 on the left-hand side. A similar significant result was noted by comparing transgender women and female groups, showing a p-value of 0.001 in both the right and left hands. On the contrary, the mean 2D:4D ratio between transgender women and males showed a p-value of 0.451 on the right hand and a p-value of 0.943 on the left hand,

Gender	2D:4D ratio	n	Min	Max	Mean	SD
Female	Right	122	0.89	1.31	0.974	0.04462
	Left	122	0.87	1.21	0.9618	0.04218
Male	Right	130	0.88	1.15	0.9595	0.04351
	Left	130	0.81	1.03	0.943	0.03625
Transgender women (MtF)	Right	140	0.85	1.08	0.9558	0.03654
	Left	140	0.84	1.15	0.9434	0.03989

[Table/Fig-4]: Descriptive analysis of mean values of 2D:4D ratios in males, females and transgender women.
Min: Minimum; Max: Maximum; SD: Standard deviation

confirming that there is no significant difference between these two groups, clarifying that transgender women's somatic features are analogous to males [Table/Fig-5].

Comparison group	2D:4D ratio	Mean diff.	Standard error	t-value	p-value
Female/male	Right	0.01447	0.00556	2.601	0.010*
	Left	0.01876	0.00496	3.785	0.001*
Transgender women (MtF)/female	Right	0.01815	0.00502	3.619	0.001*
	Left	0.01842	0.00507	3.631	0.001*
Transgender women (MtF)/male	Right	0.00368	0.00489	0.754	0.451
	Left	-0.00033	0.00466	-0.071	0.943

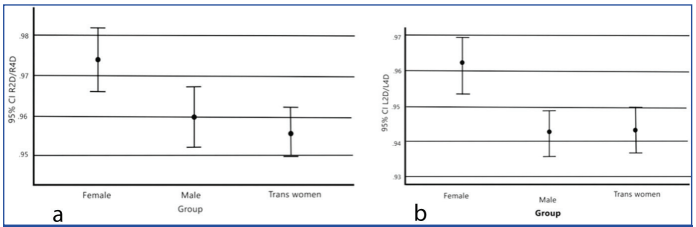
[Table/Fig-5]: D2/D4 ratios of both right and left hands among the three gender groups.

The percentage distribution of type 3 in males, females and transgender women on the right hand was 44.61%, 32.78%, and 46.43%, respectively, whereas on the left hand, it was 39.23%, 24.60%, and 42.86%, respectively. This implies that in males, the length of the ring finger was greater than the index finger, whereas in females, the length of the ring finger and index finger was the same (type 2) with 14.75% on the right hand and 18.03% on the left hand. Therefore, in males and transgender women, the type 3 finger pattern (Length of ring finger > Length of index finger) was more observed, whereas in females, the type 2 finger pattern was more prevalent than in males [Table/Fig-6].

	Type 1				Type 2				Type 3			
Gender (n)	Rt (n)	Rt (%)	Lt (n)	Lt (%)	Rt (n)	Rt (%)	Lt (n)	Lt (%)	Rt (n)	Rt (%)	Lt (n)	Lt (%)
Female (122)	3	2.46	9	7.38	18.03	14.75	22	18.03	40	32.78	30	24.60
Male (130)	5	3.85	7	5.38	3	2.31	6	4.62	58	44.61	51	39.23
Transgender women (140)	2	1.43	5	3.57	3	2.14	5	3.57	65	46.43	60	42.86

[Table/Fig-6]: Percentage distribution of various types among males, females and transgender females.

The pair-wise comparison results show that the mean of females significantly differed from males and transgender women. However, the mean value of males did not differ significantly from transgender women [Table/Fig-7].



[Table/Fig-7]: a) The 2D:4D mean ratios of right-hand values are plotted with a confidence interval of the difference among males, females and transgender women; b) The 2D:4D mean ratios of left-hand values are plotted with a confidence interval of the difference among males, females and transgender women.

DISCUSSION

The 2D:4D ratio in humans is of considerable interest in research as it exhibits sexual dimorphism. We analysed the 2D:4D ratio in relation to gender identification in the transgender population. The relative 2D:4D ratio might serve as a suitable indirect biomarker for prenatal androgen activity, influencing the masculinising effects on behaviour and secondary sexual characteristics of an individual.

In the present study, the mean 2D:4D ratio of the right hand in males, females and transgender women was found to be 0.9595, 0.974 and 0.9558, respectively. The mean 2D:4D ratio of the left hand in males, females and transgender women was found to be 0.943, 0.9618, and 0.9434, respectively.

A comparison was made between previous and present studies. Results of mean 2D:4D ratios among male, female and transgender women populations are shown in [Table/Fig-8] [11-26].

Visual classification of index and ring finger: Robertson J et al., studied the 2D:4D ratio in males and females using hand radiographs. The mean 2D:4D ratio in males was less when compared to females and showed a significant difference with p-value <0.001. Based on finger length measurements, the hand was categorised into three types, and it was found that a majority of males fall under type 3 hand (ring finger greater than index finger). In females, type 3 was less in number since the index and ring finger lengths were almost similar, showing type 2. The male pattern 2D:4D finger type in males and transgender women was similar to the present study. However, in females, the percentage distribution of type 3 was less. The non gonadal somatic sex difference that masculinises males due to foetal androgen levels exhibits 2D:4D sexual dimorphism [11].

A 2D:4D ratio in the cisgender population: According to Van Hemmen J et al., Aboul-Hagag KE et al., and Gillam L et al., the mean 2D:4D ratio between females and males shows a significant p-value (<0.001) in both the right and left hand. It was found that the mean 2D:4D values were greater in females than in males, due to prenatal testosterone levels playing a vital role in digit length values, which correlates with the present study [12-14].

Canan F et al., conducted a study linking Problematic and Pathological Internet Use (PPIU) to 2D:4D ratios in a sample of both females and males, which shows a stronger association in males than in females due to prenatal testosterone concentration leading to addictive video gaming [15].

Dey S and Kapoor AK, and Jacob M et al., conducted a study in the Indian population, which revealed that the mean 2D:4D digit ratio in males (0.968) was significantly lower when compared to females (1.014). The sex differences were slightly different, higher for the right hand than the left hand, implying that the 2D:4D digit ratio in the right hand is more sensitive to foetal androgens than the left hand [16,17].

Asuku A et al., reported that the mean 2D:4D ratio in males was less than in females, and the authors also noticed that the difference was more pronounced in the right hand than the left hand when comparing the urban with rural male and female populations. Certain environmental factors influence the digit ratio during the embryonic period, which may manifest in later life, establishing such a difference in urban-rural populations [18].

According to Jaiswal A et al., the mean 2D:4D ratio in males was lower compared to females due to prenatal testosterone levels,

Name of authors	Mean 2D:4D ratio (Right hand)			Mean 2D:4D ratio (Left hand)			Region/population where the study was conducted
	Male	Female	Trans women (MtF)	Male	Female	Trans women (MtF)	
Robertson J et al., 2008 [11]	0.906	0.921	-	0.909	0.922	-	Caucasian population
Van Hemmen J et al., 2017 [12]	0.956	0.976	-	0.963	0.980	-	Caucasian population
Aboul-Hagag KE et al., 2011 [13]	0.9672	0.9878	-	0.9680	0.9875	-	Egyptian population
Gillam L 2008 [14]	0.957	0.978	-	0.956	0.977	-	Nottingham
Canan F et al., 2017 [15]	0.991	1.009	-	0.990	0.997	-	Southern Turkey
Dey S and Kapoor AK, 2016 [16]	0.9678	1.0145	-	0.9689	1.0142	-	North Indian population (Uttarakhand)
Jacob M et al., 2015 [17]	1.01	0.98	-	0.97	0.97	-	South Indian population
Asuku A et al., 2018 [18]	0.96	0.99	-	0.96	0.99	-	Nigerian population
Jaiswal A et al., 2017 [19]	0.97	0.99	-	0.967	0.987	-	North India
Shokri H et al., 2021 [20]	0.98	0.97 (Schizophrenia-Male)	-	0.99	0.98 (Schizophrenia-Male)	-	Iran
Hisasue S et al., 2012 [21]	0.945	0.999	0.955 Transmen (FtM)	0.941	0.979	0.954 Transmen (FtM)	Japanese population
Leinung M and Wu C 2017 [22]	0.972	0.998	0.978 (MtF) 0.983 (FtM)	-	-	-	New York
Siegmann EM et al., 2020 [23]	0.959	0.975	0.963 (MtF) 0.972 (FtM)	0.961	0.974	0.969 (MtF) 0.971 (FtM)	Germany
Sadr M et al., 2020 [24]	0.959	0.983	0.972 (MtF) 0.981 (FtM)	0.974	0.991	0.981 (MtF) 0.991 (FtM)	Iran
Saglam T et al., 2020 [25]	0.97	1.00	0.97 (AMB-GD) 0.98 (AFB-GD)	0.97	0.99	0.98 (AMB-GD) 0.98 (AFB-GD)	Turkey
Vujovic S et al., 2014 [26]	0.928	0.921	0.920 (MtF)	0.935	0.945	0.926 (FtM)	Serbia
Present study 2021	0.9595	0.974	0.9558	0.943	0.9618	0.9434	South Indian population

[Table/Fig-8]: Comparison between previous and present study results of mean 2D:4D ratios among male, female and transgender women population [11-26].
MtF: Transgender women; FtM: Transmen; AMB: Assigned as male at birth; AFB: Assigned as female at birth

but no significant difference was observed in the 2D:4D ratio between the right and left hands. This comparison of the 2D:4D ratio among the cisgender group is in accordance with the present study [19].

Shokri H et al., compared the 2D:4D ratio between healthy males (n=72) and males with schizophrenia (n=62) and found that there was no significant difference (p-value >0.05) between the two groups except for the right index finger length. The index finger length serves as a biomarker to assess the prognosis of schizophrenia [20].

In a study on the 2D:4D ratio in the cisgender and transgender population, Hisasue S et al., described the mean 2D:4D ratio in gender identity disorder (FtM) as 0.955 and 0.954, in the right and left hand, which is lower when compared to female controls with 0.999 and 0.979, respectively in the right and left hand. This reflects early testosterone exposure in the foetus creating an impact on the 2D:4D ratio. Control males show a lower 2D:4D ratio than control females [21]. The present study also shows a lower 2D:4D ratio in control males than in control females. The comparison between male and female transgender women (MtF) and females shows a significant p-value <0.001. However, the comparison between transgender women (MtF) and males showed a p-value >0.001, which implies that the concentration of sex hormones in early foetal life decides the architecture of the human body.

According to Leinung M and Wu C, the mean 2D:4D ratio for the dominant hand in transmen (FtM) was 0.983, which was lower than in female controls (0.998) but showed a similar ratio to male controls (0.972). No difference in the 2D:4D ratio was noted between transgender women (MtF) and male controls with a p-value of 0.434, which correlates with the present study. It concluded that the gender identity of transmen (FtM) was influenced by prenatal androgen exposure, whereas for transgender women (MtF), gender identity was influenced by additional factors like prenatal stress [22].

Siegmann EM et al., observed that the mean 2D:4D ratio in both the right and left hands in MtF individuals was higher than in male controls. However, the mean 2D:4D ratios in FtM individuals were lower than in female controls in both the right and left hands. No significant results were found when comparing FtM individuals with male controls in both the right and left hands. In contrast, when comparing MtF individuals with male controls, significance was noted only in the left hand (p-value=0.049), while it was insignificant in the right hand. In the present study, the comparison between MtF individuals and female controls was significant in both the right and left hands, but with male controls, it was not significant in either hand. The results slightly differ from those of the above-mentioned authors regarding the left hand in MtF individuals' comparison with male controls since the 2D:4D ratio in the right hand is more sensitive to foetal androgens than the left hand, and prenatal androgen influences gender identity in individuals born as males [23].

Sadr M et al., compared the mean 2D:4D ratios of transgender women and transmen with a control group of the same natal sex. The results showed that transgender women had significantly less masculinised values compared to control males, while transmen had more masculinised values compared to control women. In the present study, the comparison between transgender women and control men showed similar results, with less masculinisation and more pronounced differences on the right hand. The results possibly predict weak prenatal testosterone effects in natal males, while strong prenatal testosterone effects in natal females may be a causative factor for gender dysphoria [24].

Saglam T et al., analysed the mean 2D:4D ratios of Assigned Female at Birth (AFB-GD) and Assigned Male at Birth (AMB-

GD) with male and female controls. The mean 2D:4D ratio of AMB-GD did not differ significantly from the male controls in both the right and left hand. The finger ratio in the female controls shows a significant difference on the right hand (p-value <0.001), but the ratio was found to be insignificant on the left hand. The mean 2D:4D ratio of AFB-GD shows a significant difference (p-value=0.028) from the female controls in the right hand. However, there was an insignificant difference in the left hand. In the left hand, the ratio was insignificant with female controls and showed a significant difference (p-value=0.045) with male controls. The present study on transgender women shows a significant difference (p-value <0.001) with cis females in both the right and left hands. However, on the contrary, with cis males, the p-value (0.451, 0.943) was not significant on both the right and left hands, which correlates with Saglam T et al.'s findings as the prenatal exposure of the foetal brain to testosterone level determines gender identity [25].

Vujovic S et al., observed that the mean 2D:4D ratio of MtF with control males doesn't show any significant difference, but values were similar to control females. However, the comparison between FtM with male and female controls shows the lowest ratio in the left hand. The present study correlates with Vujovic S et al.'s findings on transgender women, which show a significant difference with female controls in both the right and left hands. However, with male controls, it was not significant in both the right and left hands. The result describes that decreased androgen exposure in prenatal life may play a vital role in MtF gender identity [26].

A significant strength of the present study is the sample collection. Out of all genderqueers, selecting transwomen was a great challenge. The methodology of recording the 2D:4D ratio by a digital scanner is more accurate. It is a non invasive method that does not cause any harm to the participants. Further studies among transwomen and other genderqueer groups with a large sample size are needed to validate the findings of the present study. In the future, further research might explore a new standard methodology to robustly capture the dermatoglyphics pattern. In the present study, only the index and ring finger (2D:4D) ratio was studied. The study of other digits' ratios could provide additional information that may possibly fulfill the lacunae.

Limitation(s)

The larger the sample size, the more precise the results will be. The authors focused solely on transwomen, making it challenging to include a greater number of samples. When recording the finger scanning of both hands, the accuracy may be influenced by the extent of finger spreading, potentially leading to a small error.

CONCLUSION(S)

The 2D:4D ratio indirectly predicts the foetal testosterone to estradiol ratio, enlightening us to understand and explain different personality traits. The present study concluded that transgender women's somatic appearance falls in favour of males based on the 2D:4D ratio, which needs further extended research on the dermatoglyphics of the transgender women population to endorse the current result. India lacks considerable data regarding third genders. In view of that, the present article provides one of the crucial anthropological measurements, which will be additive to digit ratio research in India as well as be useful for forensic research.

Authors' contribution: KA: Concept, designing, data collection, interpretation of results and execution of research. DS: Designing and interpretation of results. SS: Interpretation of results and manuscript preparation.

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