ORIGINAL RESEARCH

Is Primary Opening of Fistula-in-Ano Always at Dentate Line: Correlation Between MRI and Operative Findings in 379 Patients

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Background: The primary opening of the cryptoglandular fistula-in-ano is generally assumed to be present at the dentate line as the cryptoglandular glands open there. However, no study has ever systematically studied the location of the primary opening.

Methods: All fistula-in-ano patients operated-on over two years were screened and those who were never earlier operated on were included. Magnetic Resonance Imaging (MRI) was done on all patients. The primary fistula opening was localized on the MRI and corroborated with the operative findings. The primary opening was categorized at three levels - at the dentate line, above the dentate line, and below the dentate line.

Results: 744 anal fistula patients were operated on over two years and 379 patients, who had never been operated on before, were included in the study. 35 patients were excluded (the primary opening could not be localized). In 344 patients (finally analyzed), the primary opening was at the dentate line in 223 patients (64.8%), above the dentate line in 79 (22.9%), and below the dentate line in 42 (12.2%) patients. The primary opening was located above the dentate line in significantly higher numbers in complex fistulas than in simple fistulas (73/102 in complex vs 6/242 in simple fistulas, p<0.00001).

Conclusion: Unlike commonly presumed, the primary opening is located at the dentate line in only two-thirds (64.8%) anal fistulas. In 22.9% it was located above the dentate line and in 12.2%, below the dentate line. This is the first study in which the level of primary opening has been systematically analyzed.

Keywords: rectal fistula, anal fistula, MRI, internal opening, primary opening, dentate line

Introduction

An anal fistula is a difficult disease to treat.¹ Unlike other gastrointestinal fistulas, an anal fistula passes through the anal sphincters- external anal sphincter (EAS) and internal anal sphincter (IAS). Due to this, there is an inherent risk of damage to the anal sphincters leading to disturbance in continence.^{2–4} One of the pivotal steps in fistula management is accurate localization of the primary (internal) opening.^{5,6} Of all the risk factors, the inability to locate the fistula's internal opening is associated with the highest risk of fistula recurrence.^{3,5–7}

The anatomic anal canal is defined as the part between the anal verge and the dentate line. But clinically, the definition used more commonly is 'surgical anal canal' which is defined as the part of the gut between the anal verge and the anorectal sling.^{8–10}

© 2025 Garg et al. This work is published and licensed by Dove Medical Press Limited. The full terms of this license are available at https://www.dovepress.com/terms.php you hereby accept the Terms. Non-commercial uses of the work are permitted without any further permission from Dove Medical Press Limited, provided the work is properly attributed. For permission for commercial use of this work, please see paragraphs 4.2 and 5 of our Terms (http://www.dovepress.com/terms.php). The anal glands are presumed to be the origin of perianal abscesses and fistulas.^{11–13} These anal glands are located at the base of the column of Morgagni and open into the crypts through a duct in the anal canal at the dentate (pectinate) line.¹¹ The anal gland ducts, from their opening at the dentate line, enter the submucosa, where two-thirds enter the IAS and approximately half terminate in the intersphincteric plane.¹⁴ It is postulated that the obstruction of these ducts leads to perianal abscesses and anal fistulae and due to this reason, the primary internal opening of the cryptoglandular anal fistula is presumed to be located at the dentate line.⁸

However, in our clinical experience, it was observed that the primary opening was not located at the dentate line in all cases. We could not come across any study in the literature that had analyzed the exact location of the primary internal opening in a systematic manner. So, the main objective of the study was to ascertain whether the primary opening always opens at the dentate line or not.

Methods

All consecutive anal fistula patients operated at a specialized referral center over the last two years, between July 2022 to July 2024 were screened. Magnetic Resonance Imaging (MRI) was done on all the patients. Only the patients who were never operated for anal fistula were included in the study. The primary fistula opening was localized on the MRI and corroborated by the operative findings. The primary opening was categorized to be at three levels - at the dentate line, above the dentate line, and below the dentate line.

The fistulas were classified according to Park,^{15,16} St James University Hospital (SJUH),¹⁷ and Garg classifications¹⁸ and were further classified as 'simple' and 'complex' fistulas per the Garg classification (Table 1). The latter was utilized as this was the only classification that categorizes fistulas into simple and complex categories.¹⁸ Moreover, Garg classification has been shown to correlate accurately with fistula complexity, guides fistula management and was validated in a large patient cohort.¹⁸ Garg grades I & II are categorized as 'simple fistulas' (fistulotomy can be done safely with low risk to continence) and Garg grades III–V are categorized as 'complex fistulas' (fistulotomy is contra-indicated due to high risk of incontinence and a sphincter-sparing procedure is recommended) (Table 1).¹⁸

Inclusion Criteria

Cryptoglandular anal fistula patients who had never undergone any surgical procedure for anal fistula before

	Parks Classification	St. James University Hospital Classification	Garg Classification	
Grade I	Intersphincteric	Intersphincteric linear	Low intersphincteric /trans-sphincteric: Single tract	Simple
Grade II	Transsphincteric	Intersphincteric with extension/s or associated abscess	Low intersphincteric /trans-sphincteric: Multiple tracts, horseshoe or associated abscess	
Grade III	Suprasphincteric	Trans-sphincteric linear	IIIA: High trans-sphincteric: Single tract; IIIB: (1) Anterior fistula in a female or any lower; (2) Grade I or II fistula with associated comorbidities#	Complex
Grade IV	Extrasphincteric	Trans-sphincteric with extension/s or associated abscess	High trans-sphincteric: Multiple tracts, horseshoe or associated abscess	
Grade V	Nil	Supralevator and translevator extension	Suprasphincteric or Supralevator or Extrasphincteric or RIFIL fistula	

Table I		Classifications	of	Anal	Fistulas	
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Notes: #Comorbidities: Crohn's disease, existing sphincter injury/weakness or post-radiation Low fistula involves $\leq 1/3$ and high fistula involves > 1/3 of the sphincters. **Abbreviation**: RIFIL, Fistula at 'Roof of ischiorectal fossa inside levator muscle'.

Exclusion Criteria

- 1. Patients with anal fistula who were operated on earlier for anal fistula, perianal abscess, or any other proctological disorder
- 2. Non-cryptoglandular fistulas Crohn's fistula, Tuberculosis fistula, extrasphincteric fistula
- 3. MRI not available
- 4. Primary internal opening could not be localized on MRI or intraoperatively.

Study Protocol

The data of all the patients operated on between 1 July 2022 and 1 July 2024 was retrieved, including operative notes and MRI scans. Apart from demographic details, the relevant history, clinical examination, fistula characteristics like previous fistula surgeries performed, associated abscess, number of tracts (single or multiple), horseshoe tracts, supralevator extension, suprasphincteric tract were noted. The location of the primary internal opening was noted and tabulated from the operative notes. After this, the MRI of all the patients was assessed independently by two experts (BK, PG) both of whom had independent experience of analyzing more than 8000 anal fistula MRIs over the last 20 years.¹⁹ A consensus was reached after discussion in cases in which their findings differed. The cases, where a consensus could not be reached, were excluded from the study.

Categorizing the Level of Primary Opening on MRI

On MRI, the primary opening was categorized to be at three levels - at the dentate line, above the dentate line, and below the dentate line. The length of the surgical anal canal [distance between the anal verge and the anorectal sling (lower border of the levator ani muscle)] was calculated on MRI by the method described below. The primary internal opening was categorized on MRI as

Above the Dentate Line

When the primary internal opening was located >33% of the total length of the surgical anal canal from the anal verge (upper 2/3 of the surgical anal canal)

Below the Dentate Line

When the primary internal opening was located <25% of the total length of the surgical anal canal from the anal verge (below the lower 1/4 of the surgical anal canal)

At the Dentate Line

When the primary internal opening was located between 25 to 33% of the total length of the surgical anal canal from the anal verge (between lower 1/4 and upper 2/3 of the surgical anal canal)

There is no paper or study in the literature which attempted to find the level of the internal opening and correlate it with the dentate line. So, based on our experience and a pilot study, we defined the level of the dentate line on the MRI. The dentate line is an anatomical landmark present on rectal mucosa. It can be seen intraoperatively on visual inspection but is not visible on MRI. So, before initiating the study, we analysed a cohort of 50 patients of anal fistula who were never operated before for the fistula and in whom the internal opening was present at the dentate line (these patients were not included in the study). Then, in these patients, we correlated the level of the internal opening with relation to the boundaries/ landmarks of the surgical anal canal on the MRI scan. This demonstrated that the level of the internal opening (hence the dentate line) was present between 25 to 33% of the total length of the surgical anal canal from the anal verge (between lower 1/4 and upper 2/3 of the surgical anal canal). This led to further deduction of the level of the internal opening above or below the dentate line as mentioned above.

This categorization of the primary internal opening in relation to the dentate line was apart from the location of the primary opening, the amount of external anal sphincter (EAS) by the fistula was also noted and tabulated. This parameter, "height of penetration of external sphincter (HOPE)", is considered important to classify fistulas.²⁰ The fistula involving

 \leq one-third of EAS is considered low, and the fistula involving > one-third of EAS is considered high.^{2,18,21–28} So, the HOPE parameter is pivotal to categorizing fistula as low or high.

The approval was taken from the Ethics Committee of Indus International Hospital via reference No EC/IIH-IEC /SP6. The patients were informed about the purpose of the study, written informed consent was obtained, and the study was conducted in accordance with the Declaration of Helsinki.

Calculation of the Level of Primary Internal Opening on MRI Step-I: Understanding the Anatomy of the Anorectum on MRI

The two important landmarks to be identified on an MRI are the 'anorectal sling' and the 'anal verge' as the distance between these two is the length of the 'surgical anal canal'. The anorectal sling is identified on MRI as the lower border of the levator ani muscle (horizontal yellow arrow in Panel-A, Figure 1 and upper red arrow in Panel-B, Figure 1).²⁹ The anal verge is identified by the point (upper limit) of the medial part of the subcutaneous part of the external anal sphincter (EAS) (lower red arrow in Panel-B, Figure 1).²⁹ The subcutaneous part of the EAS descends lateral to the internal anal sphincter (IAS) and then curves around the inferior border of the IAS (vertical green arrow in Panel-A, Figure 1) and ends a few millimeters medial to the IAS. The point where this (subcutaneous) part of the EAS ends is the "anal verge" (lower red arrow in Panel-B, Figure 1).²⁹

Step-2: Measuring the Length of the Surgical Anal Canal

Once the two landmarks ('anorectal sling' and 'anal verge') are identified, the distance between the two is measured on MRI by drawing a scale (provided by "Radiant Dicom Viewer", an MRI viewing application) (Vertical green line in Panel-B, Figure 1).²⁹



Coronal Section

Figure I Normal anatomy- Coronal Section (MRI) and measurement of length of the surgical anal canal. Panel (A) (Left)- Green arrows showing the external anal sphincter. Vertical green arrow showing the lower border of the external anal sphincter (subcutaneous part). Horizontal green arrow showing the mid-part of the external anal sphincter (superficial part). Yellow arrows showing the levator ani muscle. The vertical yellow arrow shows the upper level of the levator ani muscle and its junction with the obturator internus muscle. The horizontal yellow arrow shows the lower border of the levator ani muscle. Panel (B) (Right)- Measuring the length of surgical anal canal. The part of the anal canal between the two red arrows [between the anorectal sling (lower border of levator ani muscle) and anal verge (where the subcutaneous part of the external anal sphincter)] is the surgical anal canal. This length is measured by a vertical scale drawn on the MRI section (vertical green line). The length of the surgical anal canal is 4.46 cm in this case. The dentate line is approximately between 25–33% of this line when measured from the anal verge.

Garg et al

Step-3: Utilizing Biplanar Mode on MRI

Most MRI viewing applications have a bi-planar and a multiplanar mode. It's a very useful feature. In bi-planar or multiplanar mode, two or multiple sections like axial, coronal, sagittal, diffusion, etc. can be opened simultaneously. While viewing a particular section, the level of that section can be viewed in another section. For example, suppose an axial (transverse) section is being assessed in a bi-planar mode and a coronal section is being opened in the second half (Figure 2). In that case, the corresponding level of each axial section image can be seen on the coronal section.

Step-4: Calculating the Level of Primary Internal Opening

In a biplanar mode, an axial section is opened on one side (Left side in Figure 2) and the coronal section of the patient is opened (right side in Figure 2). The axial section is opened at the level of the primary internal opening. This will



Figure 2 Calculation of the height of primary (internal) opening on a biplanar mode (simultaneously analyzing both axial and coronal sections of MRI). The biplanar mode shows the level of one section in the other and vice versa. The axial section (left panels) is at the primary (internal opening) level. The vertical yellow arrow shows the primary opening. The coronal sections (right panels) show the height at which the axial section is taken (the level of the primary opening in this case). Panel (**A**) (Upper)-The coronal section (right) shows the level of the primary opening (horizontal yellow line). Panel (**B**) (Lower)- The length of the surgical anal canal (as depicted in Figure-1, panel-(**B**)) is measured. This shows both the level of the primary opening (horizontal yellow line) and the length of the surgical anal canal by drawing a vertical scale (vertical green line). The intersection of both lines (Point-P, Panel-(**B**), right side) shows the precise level of the primary internal opening. If this point is between 25–33% of the total length of the surgical anal canal, then the primary opening is AT the level of the dentate line. If this point is < 25% of the total length of the surgical anal canal, then the primary opening is ABOVE the level of the dantate line.

highlight the level of the primary internal opening on the coronal section (Figure 2, Panel-A, right side, yellow horizontal line). Once this is done, then a scale is drawn in the coronal section (Figure 2, Panel-B, right side, green vertical line).

The intersection of these two lines (Point P- Figure 2, Panel-B, right side) shows the exact level of the primary internal opening in the 'surgical anal canal.²⁹ The distance between the intersection of the lines (Point-P) and the anal verge (lower point of the green vertical line) is the height of the primary opening from the anal verge. This is divided by the total length of the surgical anal canal (green vertical line). This gives the proportionate (percentage) level of primary opening in the anal canal.

If this point is between 25-33% of the total length of the surgical anal canal, then the primary opening is AT the level of the dentate line (Figure 3).²⁹

If this point is < 25% of the total length of the surgical anal canal, then the primary opening is BELOW the level of the dentate line (Figure 4).²⁹

If this point is >33% of the total length of the surgical anal canal, then the primary opening is ABOVE the level of the dentate line (Figures 5–7).

The sphincter is shorter anteriorly, especially in females. Therefore, in addition to the above calculation, the sagittal view is also helpful in ascertaining the height of the internal opening and the point where the fistula tract penetrates the external sphincter (HOPE parameter). Similarly, in patients with previous surgery, the resultant fibrosis would need to be considered while calculating the height of the internal opening. In the present study, all the patients who had undergone any previous proctological procedure were excluded.

MRI Imaging protocol³⁰

All the MRI scans were performed on the same 1.5 Tesla MRI scanner (Achieva, Phillips Medical Systems, Best, Netherlands). The machine utilized a phased-array four-channel sense body coil. As the first step, the long axis of the anal canal was identified utilizing a midline sagittal localizing sequence. After this, the subsequent transfers and coronal short inversion recovery sequences (STIR -Short Tau Inversion Recovery) were determined for the anal canal axis. Additional axial T-1 W scans and coronal T-2 W for the anal canal axis were done in all the patients. Sagittal T-1-weighted images were used in approximately 15% of patients when a presacral extension was suspected from the short inversion time recovery (STIR) images. No intravenous contrast material or endorectal/endoanal receiver coil was used in any of the patients and no three-dimensional reconstruction was done. Established criteria for MR diagnosis of fistula in-ano were used. The radiologist noted her opinion of the internal opening (location and height), number of fistula tracts, associated features, and classification of the fistulas as per Parks, SJUH, and Garg classifications.

Results

744 anal fistula patients were operated between July 2022 to July 2024. 365 patients were excluded as they had earlier undergone a single or multiple anal fistula surgery. 379 patients who had never earlier undergone surgery for anal fistula were included in the study. 35 patients were excluded (Crohn's or tubercular disease, the primary opening could not be localized during surgery or MRI or both), and 344 patients were finally included and underwent analysis (Figure 8). The patient and fistula characteristics are tabulated in Table 2. The fistulas were classified as Parks, St James University Hospital (SJUH), and Garg Classifications (Table 3).

In the patients studied (n=344), the primary opening was at the dentate line in 223 patients (64.8%) (Figure 3), below the dentate line in 42 (12.2%) patients (Figure 4), and above the dentate line in 79 (22.9%) (Figure 5–7) (Table 4).

The height of the fistula as determined by the HOPE (Height of penetration of external anal sphincter by the fistula tract) parameter was assessed for each patient (Table 4). The level of the opening of the primary internal opening correlated well with the fistula height in most cases. When the primary opening was located at the dentate line (n=223), then the fistula was also low in most cases (98.2%-219/223) and high (>1/3 EAS involved) in only 1.8% (4/223) cases. (Table 4). On the other hand, when the primary opening was located above the dentate line (n=79), the fistula was high (>1/3 EAS involved) in the majority of cases (92.4%-73/79) (Table 4). When the primary opening was located below the dentate line (n=42), then the fistula was always low ($\leq 1/3$ EAS involved) (Table 4). The difference was statistically significant (p<0.00001, chi-square test).



MRI (T-2 Weighted)



MRI (STIR)



Figure 3 A 48-year-old male with a posterior low transsphincteric with a primary anal opening at 6 o'clock position (posterior midline) at the dentate line (at approx 30% height from the anal verge). Panel (A)- Schematic diagram- Axial section. Panel (B)- Schematic diagram- Coronal section. Panel (C)- MRI (T-2 weighted) and Panel (E)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (D)- MRI (T-2 weighted) - Coronal Section: The yellow line shows the level of the axial section in Panel-(C) (the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (5.30 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 30% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (F)- MRI (STIR) - Coronal Section: The yellow line shows the measurement and extent of the surgical anal canal (approx. 30% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (F)- MRI (STIR) - Coronal Section: The yellow line shows the level of primary opening at the 9 o'clock position (posterior midline) [Yellow arrow]. Panel (F)- MRI (STIR) - Coronal Section: The yellow line shows the level of the axial section in Panel-(E) (the level of primary internal opening). The Red line shows the level of primary internal opening. The Red line shows the level of primary internal canal (approx. 30% from the axial section in Panel-(E)]. Panel (E)- MRI (STIR) - Coronal Section: The yellow line shows the level of the axial section in Panel-(E) (the level of primary internal opening). The Red line shows the level of primary opening in the anal canal (approx. 30% from the axial section in Panel-(E)].





Figure 4 A 35-year-old male with a Left low transphincteric with a primary anal opening at 6 o'clock position (posterior midline) below the dentate line (at approx 15% height from the anal verge). Panel (**A**)- Schematic diagram- Axial section. Panel (**B**)- Schematic diagram- Coronal section. Panel (**C**)- MRI (T-2 weighted)- Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**D**)- MRI (T-2 weighted) - Coronal Section: The yellow line shows the level of the axial section in Panel-(**C**) (the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (4.0 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 15% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**F**)- MRI (STIR) - Coronal Section: The yellow line shows the level of the axial Section: The yellow line shows the level of the axial section in Panel-(**C**). The transaction of both lines shows the level of primary opening in the anal canal (approx. 15% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**F**)- MRI (STIR) - Coronal Section: The yellow line shows the level of the axial section in Panel-(**E**) (the level of primary internal opening). The Red line shows the level of primary opening in the anal canal (4.0 cm). The transaction of both lines shows the level of primary internal opening). The Red line shows the level of primary internal opening. The yellow line shows the level of primary opening in the anal canal (4.0 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 15% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow in Panel-(**E**)].





MRI (STIR)



Figure 5 A 47-year-old male with a high transsphincteric fistula with an intersphincteric horseshoe tract with a primary anal opening at 6 o'clock position (posterior midline) above the dentate line (at 75% height from the anal verge). Panel (**A**)- Schematic diagram- Axial section. Panel (**B**)- Schematic diagram- Coronal section. Panel (**C**)- MRI (T-2 weighted): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**D**)- MRI (T-2 weighted) - Coronal Section: The yellow line shows the level of the axial section in Panel-(**C**) (the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (5.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 75% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**F**)- MRI (STIR)- Coronal Section: The yellow line shows the level of the surgical anal canal (s.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 75% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**F**)- MRI (STIR)- Coronal Section: The yellow line shows the level of primary internal opening. The Red line shows the level of primary internal opening. The Red line shows the measurement and extent of the surgical anal canal (s.4 cm). The transaction of both lines shows the level of primary internal opening. The Red line shows the neasurement and extent of the surgical anal canal (s.4 cm). The transaction of both lines shows the measurement and extent of the surgical anal canal (s.4 cm). The transaction of both lines shows the level of primary internal opening. The Red line shows the measurement and extent of the surgical anal canal (s.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 75% from the anal verge in



MRI (T-2 Weighted)



MRI (STIR)



Figure 6 A 53-year-old male with a Left Suprasphincteric fistula with a primary anal opening at 6 o'clock position (posterior midline) above the dentate line (at 90% height from the anal verge). Panel (**A**)- Schematic diagram- Axial section. Panel (**B**)- Schematic diagram- Coronal section. Panel (**C**)- MRI (T-2 weighted) and Panel (**E**)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**D**)- MRI (T-2 weighted) - Coronal Section: The yellow line shows the level of the axial section in Panel-(**C**) (the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (5.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 90% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel F- MRI (STIR): Axial Section: The yellow line shows the measurement and extent of the surgical anal canal (5.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 90% from the axial section in Panel-C]. Panel E- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel F- MRI (STIR - Coronal Section: The yellow line shows the measurement and extent of the surgical anal canal (5.4 cm). The transaction of both lines shows the level of primary opening). The Red line shows the measurement and extent of the surgical anal canal (5.4 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 90% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel F- MRI (STIR) - Coronal Section: The yellow line shows the level of primary opening in the anal canal (approx. 90% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow in



MRI (T-2 Weighted)



MRI (STIR)



Figure 7 A 48-year-old male with a Left Suprasphincteric and a Translevator fistula with a primary anal opening at 6 o'clock position (posterior midline) above the dentate line (approx. 85% height from the anal verge) and translevator rectal opening at 3 o'clock. Panel (**A**)- Schematic diagram- Axial section. Panel (**B**)- Schematic diagram- Coronal section. Panel (**C**)- MRI (T-2 weighted) and Panel (**E**)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**D**)- MRI (T-2 weighted) and Panel (**F**)- MRI (STIR - Coronal Section: The yellow line shows the level of the axial section in Panel-(**C**) (the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (4.0 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 85% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow in Panel-(**C**)]. Panel (**E**)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow in Panel-(**C**)]. Panel (**E**)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow in Panel-(**C**)]. Panel (**E**)- MRI (STIR): Axial Section: showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**F**)- MRI (STIR - Coronal Section: The yellow line shows the level of primary opening in the anal canal (approx. 85% from the anal verge in this case) showing primary anal opening at the 6 o'clock position (posterior midline) [Yellow arrow]. Panel (**E**)- MRI (STIR - Coronal Section: The yellow line shows the level of primary internal opening). The Red line shows the measurement and extent of the surgical anal canal (4.0 cm). The transaction of both lines shows the level of primary opening in the anal canal (approx. 85% from the anal verge in this case) showing primary anal ope



Figure 8 Flowchart depicting the inclusion and exclusion criteria for the cases included in the study.

In simple fistulas (Garg grade I–II), the primary opening was at or below the dentate line in 97.5% (236/242) fistula whereas in complex fistulas, the primary opening was at or below the dentate line in only 28.4% (25/102) and above the dentate line in 71.6% (73/102) fistulas (p<0.00001) (Table 5). Also when the primary opening was above the dentate line, the fistula was complex in 92.4% (73/79) patients (Table 5).

Characteristics					
	N=344				
Age (years)	40.3± 11.3				
Sex (M/F)	303/41				
Fistula Characteristics					
Associated Abscess	81 (23.5%)				
Horseshoe tract	54 (15.7%)				

Table	2	Patient	and	Fistula
Charact	eristi	cs		

Table 2 (Continued).

	N=344
Single tract	91 (26.5%)
Multiple tracts	245 (71.2%)
Supralevator	12 (3.5%)
Suprasphincteric	8 (2.3%)
Translevator	I (0.03%)

Table 3 Fistula Categorization as per Different Classifications

Grade	Parks	St James University Hospital (SJUH)	Garg	
I	68	19	62	Simple
н	258	49	182	(Grades I & II)
ш	17	58	18	Complex
IV	I	200	66	(Grades II, IV, V)
v	NA	18	18	
Total	344	344	344	

Table 4 Level of Primary Opening of Anal Fistula and Correlation of the Level of the Primary Internal Opening to the Height of Fistula [(Determined by HOPE (Penetration of the External Sphincter by the Fistula Tract)]

Level of Primary Opening \rightarrow	At the Dentate Line	Above the Dentate Line	Below the Dentate Line	Test of Significance (Chi-Square Test)
Total (n=344)	223 (64.8%)	79 (22.9%)	42 (12.2%)	
LOW Fistula (≤ 1/3 EAS*(involved)	219 (98.2%)	6 (7.6%)	42 (100%)	P<0.00001
HIGH Fistula (> 1/3 EAS involved)	4 (1.8%)	73 (92.4%)	0	P<0.00001

Notes: Level of Primary Internal Opening. Below the Dentate line – located <25% of the total length of the surgical anal canal from the anal verge. At the Dentate line- located between 25–33% of the total length of the surgical anal canal from the anal verge. Above the Dentate line- located between >33% of the total length of the surgical anal canal from the anal verge. Above the Dentate line- located between >33% of the total length of the surgical anal canal from the anal verge. Above the Dentate line- located between >33% of the total length of the surgical anal canal from the anal verge. Above the Dentate line- located between >33% of the total length of the surgical anal canal from the anal verge.

Table 5 Level of Primary Opening of Anal Fistula and Its Con	rrelation with Fistula Complexity
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Level of Primary Opening \rightarrow (n=344)	At Dentate line (n=223)	Above Dentate line (n=79)	Below Dentate line (n=42)
Complex Fistula (Garg grade III–V) (n=102)	24 (23.5%)	73 (71.6%)	5 (4.9%)
Simple (Garg grade I–II) (n=242)	199 (82.9%)	6 (2.5%)	37 (15.4%)
Test of Significance (Fisher's Exact test)	P<0.00001	P<0.00001	P=0.0064

Discussion

This is perhaps the first study that analyzed the level of primary internal opening in cryptoglandular anal fistulas based on a detailed study of MRI and its correlation with operative findings. The study brought out several interesting findings. Unlike commonly presumed, the primary opening was located at the dentate line in only two-thirds (64.8%) anal fistulas (Figure 3). In 22.7% of patients, it was located above the dentate line (Figures 5–7) and in 12.5% of patients, the primary opening was present below the dentate line (Figure 4). In complex fistulas, the primary opening was located above the dentate line in more than two-thirds (71.6%) of patients. Also, the fistula was mostly complex (92.4%) when the primary opening was above the dentate level (Table 5 and Figures 5–7). The height of penetration of the external sphincter (HOPE) parameter also correlated significantly with the level of the internal opening (Table 4, p<0.00001). The EAS involvement was $\geq 1/3$ in the majority of the patients when the internal opening was above the dentate line (92.4%) (Table 4). This highlights that the level of the internal opening was above the dentate line (92.4%) (Table 4). This highlights that the level of the internal opening was above the dentate line (92.4%) (Table 4).

The findings of the study raise a logical question and have significant clinical implications. The presumption that the primary opening would be at the level of the dentate line leads many surgeons to try to look for the primary opening at or around the dentate line. The most commonly employed method is probing the fistula through the external opening and getting the probe out from the primary internal opening. Also, the primary opening may not be locatable in about 20% of cases, especially in the complex fistulas.^{6,7,31} Probing the fistula in such cases, coupled with the findings of this study that in complex fistulas, the primary opening is located higher to the dentate line in more than 71% of cases, increases the risk of forming a false tract and a false internal opening. This would not only increase the recurrence rate in complex fistulas but would also further complicate the fistula, making its cure more difficult in the future.

The findings of the study raise a logical question. If most of the crypt gland opens at the dentate level, then how can the location of the primary opening away from the dentate line be explained? One possibility could be that the fistula initiates at the level of the dentate line, but a few of the more aggressive fistulas gradually spread superiorly in the submucous or intersphincteric plane. Subsequently, the fistula bursts open at the superior level, thereby forming a 'new primary' opening. Slowly, over time, the primary opening at the level of the dentate line closes down, leaving only the 'new' primary opening at a higher level. Another possible explanation that seems more plausible is that apart from opening at the dentate line, a few branches of the crypt gland extend superiorly and inferiorly as well, and these branches may be responsible for the primary internal opening at these locations away from the dentate line. Also, few cases would be the fistula developed due to the deepening of chronic anal fissures and most of these would be the subset in which the primary opening was below or at the level of the dentate line.

The strength of the study is that this is the first study in which the location of the primary opening is assessed on detailed analysis of MRI by experienced MRI experts (BK, PG). Another strong point is the large study cohort of 344 patients who were never operated before for anal fistula and whose MRI and operative notes were available for the analysis.

The MRI analysis is much more accurate than clinical examination (preoperative) as well as examination under anesthesia (EUA) as it precisely outlines both the sphincters, the fistula tracts, the crossing of fistula tracts across the sphincter, and the primary internal opening.²⁰ This level of accuracy of the position of the primary opening and its relation to the sphincters is not possible on examination alone especially when there is associated scarring, fibrosis, and inflammation due to chronic fistulas. Therefore, the availability of MRI of every patient and its detailed analysis were pivotal for the execution of such a study.

Another important point about the anatomy of the anal canal needs consideration. The anal canal is a short tubular structure which is surrounded by a much wider ischiorectal fossa. A fistula can spread in much more superior and other directions in the ischiorectal fossa and then curve downwards to open into the anal canal. The 'surgical anal canal' is a small area (between the anal verge and the anorectal ring) and its length on visual inspection in gut mucosa is usually about 3 to 4 cm. It further gets shortened by the fibrosis caused by the fistula.⁸ So, in this short distance of 3–4 cm (or even shorter), the points where the fistulas would open inside the anal canal, even the different levels (of primary

openings) would be 'quite nearby'. This could give the impression that the primary opening is usually always at or around the level of the dentate line, which perhaps led to the basis of the presumption formed over the past several decades. It was only when a detailed analysis was done on MRI that the primary opening was unequivocally demonstrated to be significantly above the dentate line in so many cases. (Figures 5-7)

The dentate line is an anatomical landmark on the mucosal surface and is visible to the naked eyes intraoperatively but is not seen on MRI. Therefore, in this study, a relatively broad range of 25 to 33% of the distance of the anal canal from its lower end was taken as the region of the dentate line. Another surprising finding was that the primary opening was also located below the dentate line in 12% of cases.

The study also highlights the value of MRI in the management of anal fistulas especially the complex one. If the primary opening seems above the dentate line, the chances of a fistula being complex are very high, and it would be prudent to refer such a case to a referral institute for a sphincter-sparing surgery. However, we do not recommend a radiological investigation [MRI or EAUS (endoanal ultrasound)] in all anal fistula patients, and these investigations should be done in only recurrent cases, fistulas with no external opening, and fistulas with non-locatable internal opening.^{23,26,32-34} We also do not recommend that the method utilized in this study (MRI-based) to locate the height/ level of the primary opening be used in clinical practice routinely. In practice, the conventional methods used to locate the primary opening (clinical examination, injection of a colored dye through the external opening, and examination under anesthesia) should be utilized to assess the primary opening, and MRI/EAUS be done only in the indications mentioned above. However, the findings of the study (that not all primary internal openings are located at the dentate line, especially in complex fistulas) are pertinent and should be kept in mind while probing and trying to locate the primary opening so as to inadvertently create a false internal opening. Also, to manage fistulas, HOPE (height of penetration of EAS by the fistula tract) parameter remains equal or more relevant to the operating surgeon than the height/level of the primary opening. The EAS is the main sphincter that needs to be preserved during fistula for preserving continence and therefore, the knowledge of whether the fistula involves >1/3 of EAS or $\le 1/3$ of EAS is important to divide fistulas as low or high and to decide whether fistulotomy can be safely performed or not.²⁰ And the study highlighted that the HOPE parameter also correlated well with the height of the primary opening. When the primary opening was above the dentate line, the fistula was complex (involved >1/3 of the EAS) in 92.4% (73/79) of patients (Table-5).

Our previously published study highlighted that MRI is an excellent modality to assess fistula healing in the postoperative period.³⁰ However, the concept and point of study in both studies are entirely different. The earlier paper³⁰ was on the comparison of preoperative and postoperative MRI after anal fistula surgery. In contrast, the present manuscript is about finding the location of the primary opening of the fistula in the anal canal. Therefore, all patients in the present study were suffering from primary fistulas, none of whom had been operated on earlier for anal fistula. So, the present study is the first study in which the location of the primary opening was determined after the correlation of the operative findings with a detailed MRI analysis.

To conclude this is the first study to unequivocally highlight that all fistulas do not open at the dentate line. Less than two-thirds (64.8%) of anal fistulas open at the dentate line, while 22.9% open above the dentate line, and 12.2% have primary internal openings below the dentate line. In complex fistulas, the primary opening was located above the dentate line in about two-thirds (71.6%) of patients and when the primary opening is above the dentate line, the fistula is complex in 92.4% of patients. These findings have significant clinical implications. More prospective studies are needed to corroborate the findings of this study.

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Disclosure

The authors declare no conflicts of interest in this work.

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137