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Which characteristics of the episiotomy and perineum are associated with a lower risk of obstetric anal sphincter injury in instrumental deliveries

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ABSTRACT

Objetive: Operative vaginal delivery (OVD) is the most important risk fact for obstetric anal sphincter injury (OASI). Knowledge of possible risk factors for their occurrence may therefore reduce the likelihood of faecal incontinence. The aim is to analyse the effect of mediolateral episitomy and perineum characteristics on the occurrence of OASI in OVD.

Study Design: Case-control study, which included 958 OVD that were reviewed in *Pelvic Floor and Puerperium Clinic*. The episiotomy and perineum characteristics of those women who experienced OASIs (n = 150) were compared with those who had no evidence of anal sphincter injury (n = 788).

Results: In multivariate logistic regression analysis the factors which were independently associated were nulliparity, persistent occipitoposterior position, birthweight >3500 g, an angle of episiotomy <30°, a distance episiotomy-fourchette <5 mm and a distance of perineal body <30 mm. The analysis of subgroups show that only the multiparous women does not benefit from any feature of the episiotomy, and an angle greater than 30° and a distance episiotomy-fourchette >5 mm are associated with a risk reduction of OASI in nulliparous, perineal bodies ≤30 mm and occipitoanterior position.

Conclusions: Two modifiable risk factors at the time of performing the episiotomy, the angle and distance episiotomy-fourchette, have been identified as the risk modification of OASI. It is necessary to achieve an adequate angle to reduce the probability of OASIs in OVD, and in nulliparous women with an anterior position and a distance of perineal body ≤30 mm could benefit from increasing the episiotomy-fourchette distance.

Introduction

Obstetric anal sphincter injuries (OASIs) are the leading cause of anal incontinence (AI) in women [1]. Thirty percent of women were symptomatic one year after OASI [2], and at 25-year revealed persistent AI in 40% [3].

Operative vaginal delivery (OVD) is used to facilitate childbirth and to avoid cesarean section delivery (CS) and its associated morbidities. Nevertheless, operative techniques are associated with a greater tendency for birth injury than spontaneous delivery [4]. OVD has been shown to be a significant contributor to the number of OASIs [5–7]. Episiotomy has traditionally been a routine component of OVD, the aim being to avoid injury to the anal sphincter and to minimise the risk of pelvic floor dysfunction in later life. In the only randomized clinical trial (RCT) comparing routine versus restrictive use of episiotomy for instrumental delivery, routine use of episiotomy was not associated with a statistically significant difference in the incidence of OASIs (8.1% vs. 10.9%) [8]. However, subsequently and with regard to the same population, Macleod et al. [9] found that restrictive use of episiotomy for instrumental delivery may increase immediate postpartum morbidity, in particular the incidence of perineal pain and stress urinary incontinence. The latest Cochrane review recommends that further research in instrumental delivery may help to clarify routine episiotomy is useful in this particular group [10].

Abbreviations: OVD, operative vaginal delivery; OASI, obstetric anal sphincter injury; AI, anal incontinence; CS, cesarean section delivery; RCT, randomized clinical trial.

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There is growing evidence that the exact placement of episiotomy plays an important role in the degree of perineal trauma [11–16]. This has also been evaluated in instrumental deliveries [17]. Mediolateral episiotomy is a compromise between midline and lateral episiotomy. The use of instrumental delivery in combination with midline episiotomy was associated with a significant increase OASIs risk in both primiparous and multiparous women [18,19]. However two large retrospective population-based register studies have suggested that mediolateral episiotomy reduces the risk of OASIS in instrumental delivery [20,21], and the lateral episiotomies are associated as a protective factor [22].

A wide variety in the clinical performance of mediolateral episiotomy has been observed between countries and institutions [23] as well as between the accoucheurs in the same institution [24–26]. Some authors have found an association between an angle too acute, or too wide [27] and the risk of OASIs, the instrumental deliveries with an acute angle was a also risk factor [17]. Stedenfeldt27 found that an episiotomy shorter or with introitus onset closer to fourchette is associated with an increased risk of anal sphincter injury. However in instrumental delivery there is no association with other episiotomy characteristics [17].

This study is designed to analyse the effect of characteristics of mediolateral episitomy and distances of perineum on the occurrence of anal sphincter injury in OVD using the data from our database.

Materials and methods

Study population

This is a observational retrospective case-control study, which included all OVDs which were reviewed in *Pelvic Floor and Puerperium Clinic* in a single tertiary centre from January 2012 to June 2017. The episiotomy features of these women who experienced OASIs and were diagnosed in the delivery room (the OASIs group), were compared with those who had no evidence of anal sphincter injury (the control group). OASI was defined as any rupture involving the anal sphincter muscles with or without rupture of the anal mucosa clinicallly diagnosed in the delivery room. Women without epistiotomy or twin deliveries were excluded from analysis. The study protocol was approved by the local Research Ethics Board.

The indications for OVD at our center are prolonged second stage and non-reassuring fetal heart rate. We performed only low or outlet instrumental deliveries. The OVD were divided into three groups: a) Vacuum extractors (VE) (including: Ventouse/suction or Kiwi delivery); b) Forceps deliveries (including: Forceps or Thierry's spatulas), and c) Sequential deliveries (which include the different combinations of at least two types of instrumental deliveries). The choice of operative delivery for the initial attempt was left to the discretion of the attending physician. In the absence of epidural analgesia, local infiltration was usually added.

All instrumental deliveries were cited in *Pelvic floor and Puerperium Clinic* between the 8th and 12th weeks after delivery. On this visit, a history was taken and a complete examination were performed, including assessment of episiotomy and perineum. With the patient in lithotomy position and legs resting in knee holders, the perineum and episiotomy scar was assessed and the following characteristics were measured (Fig. 1): 1) Angle of episiotomy (angle between the episiotomy and the midline, which was measured using a digital goniometer in degrees), 2) Length of episiotomy, 3) Distance episiotomy-fourchette (the distance from the origin of the episiotomy scar in the introitus to the fourchette), 4) Distance of perineal body, and 5) Distance of genital hiatus. Distances were measured in millimetres. Two types of episiotomy in terms of their relationship to the anus can be observed (Fig. 1), the type I was defined when the end of episiotomy scar was below the anus, and the type II when above [17].

Data collection

Data for the study were drawn from the computerized puerperium records and their linked maternal hospital discharge records. All characteristics known from the literature as a possible risk factor, and available from the database, were analysed in this study as potential attributing factors for anal sphincter injury in assisted vaginal

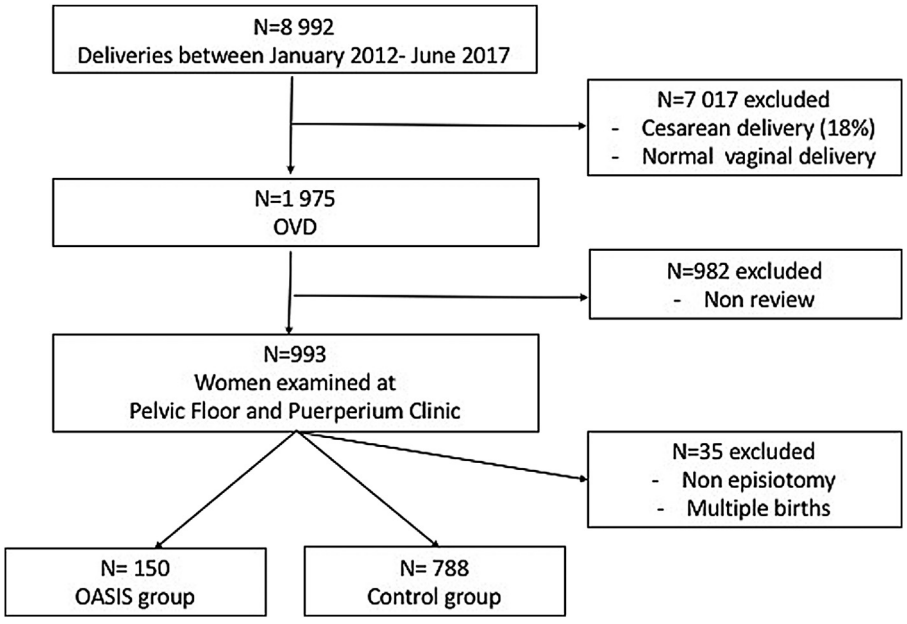

Fig. 1. The flow diagram of the included patients. OVD: Operative Vaginal Delivery. OASI: Obstetric Anal Sphincter Injuries.

deliveries. These factors were: matenal age, race, gestational diabetes, parity, gestational age, induction of labor, occipitoposterior position, mode of OVD, epidural anesthesia, duration of second stage, and fetal birthweight. The characteristics of the episiotomy (angle, length and distance to fourchette) and the distance of the perieneal body and the genital hiatus were also collected.

Data analysis

Categorical variables were evaluated using a chi-square test analysis, and the Mann-Whitney U test was used for continuous non-parametric variables and the Student-t test for continuous parametric. The Kolmogorov-Smirnov test was used to evaluate continuous data for normality prior to significance testing.

In order to account for the potentially confounding effect of differences in demographic, delivery and episiotomy characteristics between groups, a logistic regression model to predict OASIS was created using factors found to be significant in bivariate analyses. Prior to this the continuous variables were transformed into a dichotomous variable. Receiver operating characteristic (ROC) curve analysis was used to determine optimal cut-off values for episiotomy characteristics to predict any OASI risk. We defined the best cut-off value as the value with the highest accuracy which maximizes the Youden index (sensitivity + specificity-1).

Subgroup analyses were performed for the characteristics of the episiotomy that were associated with OASIS by dividing group according to predictive factors of OASI in the multivariate analysis.

Differences were considered significant when the probability value was 0.05. All data were managed and analyzed using the SPSS software (version 20.0 for Mac; SPSS Inc, Chicago, IL).

Results

Demographic, obstetrical and delivery characteristics

During the study period at our center, 1975 out of a total 8992 deliveries (21%) were OVD. The overall rate of episiotomy in all births was 45%. The rate of episiotomies and OASIs for OVD was 97% and 7.85% respectively (the rate of OASI for sequential, Espatulas, Forceps, Vaccum and Kiwi: 26.7%, 17.4%, 5.6%, 8.16% and 5.1% respectively). During this period 958 women with OVD were examined at the *Pelvic Floor and Puerperium Clinic,* and were included in this study, 150 of whom in the OASIs group and 788 in the control group. Thirty-five women were excluded due to twin delivery or no episiotomy. Fig. 1 illustrates the selection of study participants as a flow diagram. The demographic, obstetrical and delivery characteristics of the women in the OASIs and control group are presented in Table 1. Women in the OASIs group were more likely to be nulliparous (95.6% vs 89.1%, p = 0.023), and delivered at a more advanced gestational age (281 vs 280 days, p = 0.049). The OASIs group was characterized by higher rates of persistent occiput posterior position (32.5% vs 22%, p = 0.03) and a significantly higher neonatal birth weight (3388 g vs 3292 g, p = 0.037). There were no differences between the groups with regard to maternal age, nationality, gestational diabetes, weight gain, labor induction, meconium, mode of OVD, experience of the obstetrician, epidural analgesia, duration of labor, fetal sex, umbilical pH or Apgar (Fig. 2) (Table 2).

Episiotomy characteristics

On univariate analysis of episiotomy characteristics, the OASIs group was characterized by lower angle of episiotomy (30° Vs 22°, p < 0.0001), lower distance of perineal body (30 mm Vs 32.5 mm, p < 0.0001) and lower distance episiotomy-fourchette (4 Vs 3 mm, p = 0.002). Women with episiotomy type I were significantly more common in OASIs group (34.8% vs 16.7, p < 0.0001).

Table 1
Characteristics of Operative Vaginal Delivery with and without OASI.

Characteristics	No OASI n = 788	OASI n = 150	ρ value
Patient characteristics			
Maternal age (years)*	33.22 (5.17)	32.01 (5.37)	0.03[a]
Spanish (%)	91.4	91.9	NS[c]
Gestational diabetes (%)	4.4	4.7	NS[c]
Nulliparity (%)	89.1	95.6	0.023[a]
Gestational age (days)**	280 (275-285)	281 (276-287)	0.049[b]
Weight gain (Kg)**	12 (10-16)	12 (10-15.25)	NS[b]
Delivery characteristics			
Prostaglandins Induction (%)	11.7	15.3	NS[c]
Meconium (%)	22.3	15.2	NS[c]
Occipitoposterior position (%)	22	32.5	0.03[c]
Mode of delivery (%)			NS[c]
Sequential	9.1	14.9	
Espatulas	2.9	1.2	
Forceps	45.2	50.3	
Vacuum	23.6	17.4	
kiwi	19.2	16.1	
Experience obstetrician >10 years (%)	52.9	57.8	NS[c]
Epidural anesthesia(%)	94.6	95.6	NS[c]
Duration of stage labor (min)			
1 st Stage labor**	240 (161-330)	240 (180-330)	NS[b]
2nd stage labor**	94 (30-150)	90 (45-135)	NS[b]
Neonatal characteristics			
Male (%)	55.7	58,8	NS[c]
Birthweight >3500 g (%)	27.3	37.8	0,007[c]
Apgar score <5 at 1 min (%)	5	7.6	NS[c]
Apgar score <7 at 5 min (%)	2	3.1	NS[c]
Umbilical artery pH < 7.20 (%)	37	34	NS[c]

[a] Student's test.
[b] Mann-Whitney U test.
[c] Chi-Square test.
* Mean(SD).
** Median(Interquartile range); NS: Not Significant.

Risk factors for oasis: multivariate analysis

In order to identify independent risk factors for OASIs, we used multivariate logistic regression analysis to control for potential confounders (Table 3). The factors that were independently associated with OASIs in OVD were the nulliparity, persistent occipitoposterior position, birthweight >3500 g, an angle of episiotomy <30°, a distance episiotomy-fourchette >5 mm and a distance of perineal body <30 mm.

The angle of episiotomy behaves as a factor associated to anal sphincter injury, so women with a mediolateral episiotomy and an angle of greater than 30° have an 81% less risk of having an OASIS (OR 0.19,95% CI 0.12–0.31). The odds ratio estimates show that there is a 48% (OR 0.52,95% CI 0.31–0.88) and 63% (OR 0.23,95% CI 0.23–0.58) reduced risk of sustaining an anal obstetric sphincter rupture when the distance episiotomy-fourchette was >5 mm and distance of perineal body was >30 mm, respectively.

Characteristics of episiotomy and perineum in subgroup analysis

In the analysis of subgroups (Table 4) to assess the association between the characteristics of episiotomy and perineum with the risk of OASIs, we found that an angle of the episiotomy greater than 30° behaves as a protective factor in all subgroups with a risk reduction of 80%, except in multiparous women, where there is no association with any characteristics. A length of episiotomy >30 mm and a distance of genital hiatus >25 mm, as in global analysis, are not associated with increased risk of OASIS in any subgroup. A distance episiotomy-fourchette >5 mm is associated with a risk reduction of OASIS of 50%, only in women with perineal body less than or equal to 30 mm (OR 0.46,95% CI 0.25–0.83), nulliparous (OR 0.46,95% CI 0.27–0.78) and occipitoanterior position(OR 0.54,95% CI 0.3–0.96). A lower distance of perineal

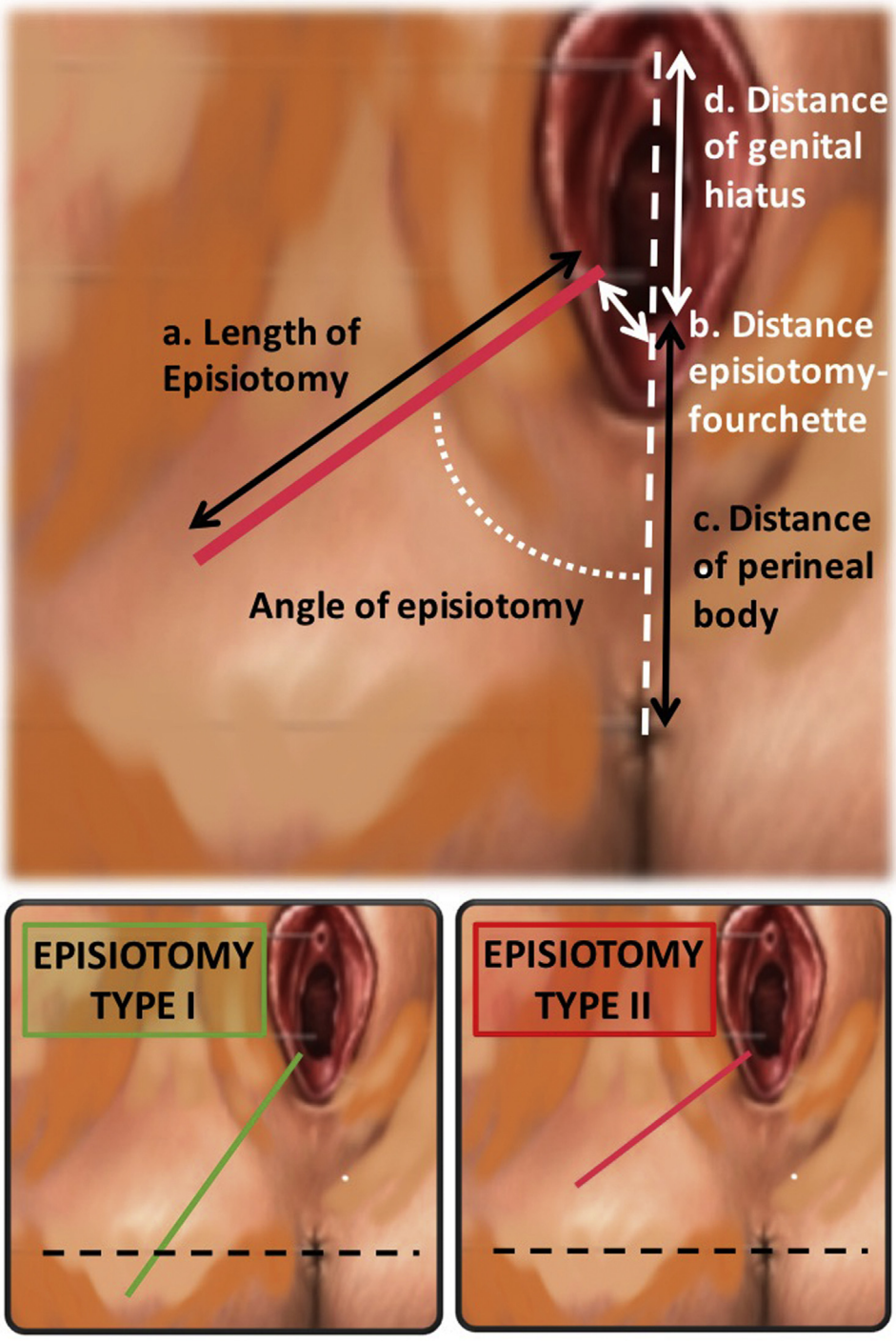

Fig. 2. Characteristics and types of episiotomies.

body is associated with an increased risk of OASIS, whether it is greater or less than 30 mm. However a distance of perineal body >30 mm is only associated with a reduction in the risk of sphincter injury in nulliparous women (OR 0.42,95% CI 0.27–0.65) and in occipitoanterior positions (OR 0.28,95% CI 0.17–0.46).

Discussion

Main findings

The main risk factors of OASIs in the OVD are nulliparity, persistent occipitoposterior position, birthweight >3500 g and a distance of perineal body ≤30 mm. We also find two protective factors which can be modified by the obstetrician at the time of performing the episiotomy, such as an angle of episiotomy >30° and a distance episiotomy-fourchette >5 mm. Except in the case of multiparous women, where they do not benefit from any feature of episiotomy to reduce the risk of OASIs, an adequate angle is the most important modifiable protective factor. In addition, the nulliparous women with an anterior position and a distance of perineal body ≤30 mm could benefit from increasing the episiotomy-fourchette distance to reduce the risk of sphincter injury.

This outcome is clinically relevant because episiotomy technique is relatively easy to modify, and our findings suggest that

Table 2
Characteristics of episiotomy with and without OASI in OVD.

Characteristics of episiotomy (Quantitative variables)	No OASI n = 788	OASI n = 150	ρ value
Angle			
Angle of episiotomy (º)*	32 (14)	23 (9)	<0,0001[a]
Angle of episiotomy >30º (%)	53.0	18.4	<0.0001[b]
Distance			
Length of episiotomy (mm): distance a*	35 (5)	30 (10)	NS[a]
Length of episiotomy >30 mm (%)	49.7	44.7	NS[b]
Distance episiotomy-fourchette (mm): distance b*	4 (5)	3 (4)	0.002[a]
Distance episiotomy-fourchette >5 mm (%)	29.9	19.2	0.007[b]
Distance of perineal body (mm): distance c*	35 (10)	30 (10)	<0.0001[a]
Distance of perineal body >30 mm (%)	52.1	34.5	<0.0001[b]
Distance of genital hiatus (mm): distance d*	25 (5)	25 (10)	0.004[a]
Distance of genital hiatus >25 mm (%)	47.4	40.8	NS[b]
Classification of episiotomies (%)			<0.0001[b]
Epsiotomy type I (%)	16.7	34.8	
Episiotomy type II (%)	83.3	65.2	

NS: Not Significant.
* Median(Interquartile range).
[a] Mann-Whitney U test.
[b] Chi-Square test.

Table 3
Factors that predict OASIS in OVD.

Factors	Wald	ρ value	OR* crude	IC 95%
Maternal age <35 years	1.12	NS		
Nulliparity	4.45	0.035	0.31	0.1-0.9
Gestational age >40 weeks	0.87	NS		
Occipitoposterior position	16.77	0.001	0.31	0.18-0.54
Birthweight >3500 g	7.34	0.007	0.62	0.43-0.87
Angle of episiotomy > 30º	44.68	0	0.19	0.12-0.31
Distance episiotomy-fourchette > 5 mm	5.76	0.016	0.52	0.31-0.89
Distance of perineal body > 30 mm	18.32	0	0.37	0.23-0.58

Values reflect the results of multivariate logistic regression analysis that controlled for the variables that are detailed in Tables 1 and 2.

modified practice would potentially lead to reduction in anal sphincter injury rates, especially in a risk group such as instrumental deliveries.

Strenghs and limitations

The strength of this study is the large number of analyzed deliveries collected in a prospective way. The weaknesses are the retrospective study design, the characteristics of mediolateral episitomy are collected weeks after delivery, only 50% of the OVD of the study period attended at *the pelvic floor and puerperium Clinic* and the small sample size of the multiparous subgroup. The results should therefore be interpreted with caution.

As the measures studied were collected a few weeks after birth, we do not know the true measures of the episiotomy at birth. The perineal distension and oedema caused by the crowning of the head and subsequent retraction by the healing process cause changes in episiotomy between at the time of incision and puerperium control. Therefore, the measurements in this study are necessarily smaller than at the time of performing the episiotomy. So we should consider three different angles of episiotomy in three periods: a) Incision, b) repair and c) scar. It has been shown that an average difference between angle at incision and repair of 15–20° [28–30,32]. Another study has shown that the angle varies as a function of the distance to fourchette at which the introitus episiotomy begins [31]. On the other hand, the length of the perineal body is modified increasing up to 50% from the first stage to crowning [33,34].

Despite the weaknesses of this study, the evident and considerable protective effect of the episiotomy angle and the distance episiotomy-fourchette clearly is of clinical importance.

Interpretation

Tincello et al. [24] were the first to question the technique of mediolateral episiotomy and to raise the issue of the degree of force relief upon the perineum related to the angle of episiotomy. Later on, Andrews et al. [25] with an observational study and Eogan et al. [13] and Stedenfeldt et al. [27] with case–control studies showed differences in the characteristics of episiotomies in women with anal sphincter injury. Eogan et al. [13] only studied the angle of episiotomy. Andrews et al. [25] also studied the length and depth but only finding differences in the angle. Stedenfeldt et al. [27] added distance episiotomy-fourchette and found an association with that distance, the depth and length of the episiotomy, but not in the angle or the perineal body. A recent random study in nulliparous found 2.4% OASI when the episiotomy angle was performed at 60° versus 5.5% if it were 40°, although the difference was not statistically significant due to the small sample size [30].

Finally a prospective case-control study also found that when a mediolateral episiotomy is performed in OVD, the technique has a strong effect on the occurrence of OASIs, and an angle greater than 20° have an 87% lower risk [17], but no association with other characteristics of the episiotomy was found. Our study confirms the important relationship between the angle of the episiotomy and the risk of OASI in OVD and also finds association with other parameters, such as distance to fourchette.

Conclusions

OVD is the most important risk fact for OASI. In daily obstetric practice, the use of OVD is necessary in the case of fetal distress or prolonged second stage of labor. Knowledge and modification of attributive risk factors may help reduce the number of anal sphincter injuries during OVD, may therefore reduce the likelihood of faecal incontinence. The obstetrician determines two factors when performing an episiotomy which can modify the risk of OASI: the angle and the episiotomy-fourchette distance. Only the multiparous subgroup does not benefit from any feature of the episiotomy. It is necessary to achieve an adequate angle >30° to reduce the probability of OASIs in OVD, as well as an adequate

Table 4
Characteristics of episiotomy in subgroup analysis.

Characteristics	Univariate analysis			Multivariate analysis		
	OR	95% CI	ρ-value	Adjusted[a] OR	95% CI	ρ-value
Perineal body						
				≤30 mm		
Angle of episiotomy > 30°	0.19	0.11–0.33	0.000	0.16	0.09–0.3	0.000
Length of episiotomy > 30 mm			NS			
Distance episiotomy-fourchette > 5 mm	0.5	0.29–0.87	0.014	0.46	0.25–0.83	0.01
Distance of perineal body (mm)	0.91	0.86–0.96	0.001	0.9	0.84–0.97	0.005
Distance of genital hiatus > 25 mm			NS			
				>30 mm		
Angle of episiotomy > 30°	0.22	0.1–0.45	0.000	0.17	0.07–0.4	0.000
Length of episiotomy > 30 mm			NS			
Distance episiotomy-fourchette > 5 mm			NS			
Distance of perineal body (mm)	0.088	0.8–0.97	0.011	0.85	0.76–0.95	0.000
Distance of genital hiatus > 25 mm			NS			
Parity						
				Nulipara		
Angle of episiotomy > 30°	0.20	0.13–0.31	0.000	0.21	0.13–0.34	0.000
Length of episiotomy > 25 mm			NS			
Distance episiotomy-fourchette > 5 mm	0.51	0.32–0.8	0.004	0.46	0.27–0.78	0.004
Distance of perineal body > 30 mm	0.47	0.32–0.68	0.000	0.42	0.27–0.65	0.000
Distance of genital hiatus > 25 mm			NS	NS		
				Multipara		
Angle of episiotomy > 30°			NS			
Length of episiotomy > 25 mm			NS			
Distance episiotomy-fourchette > 5 mm			NS			
Distance of perineal body > 30 mm			NS			
Distance of genital hiatus > 25 mm			NS			
Fetal head position						
				Occipitoposterior		
Angle of episiotomy > 30°	0.22	0.09–0.55	0.001	0.19	0.07–0.54	0.002
Length of episiotomy > 25 mm			NS			
Distance episiotomy-fourchette > 5 mm			NS			
Distance of perineal body > 30 mm			NS			
Distance of genital hiatus > 25 mm			NS			
				Occipitoanterior		
Angle of episiotomy > 30°	0.19	0.11–0.31	0.000	0.17	0.1–0.3	0.000
Length of episiotomy > 25 mm			NS			
Distance episiotomy-fourchette > 5 mm	0.63	0.37–0.95	0.071	0.54	0.3–0.96	0.035
Distance of perineal body > 30 mm	0.35	0.22–0.54	0.000	0.28	0.17–0.46	0.000
Distance of genital hiatus > 25 mm			NS			

Characteristics	Univariate analysis			Multivariate analysis			Univariate analysis			Multivariate analysis		
	OR	95% CI	ρ-value	OR	95% CI	ρ-value	OR	95% CI	ρ-value	OR	95% CI	ρ-value
Perineal body			≤30 mm						>30 mm			
Angle of episiotomy > 30°	0.19	0.11–0.33	0.000	0.16	0.09–0.3	0.000	0.22	0.1–0.45	0.000	0.17	0.07–0.4	0.000
Length of episiotomy > 30 mm			NS						NS			
Distance episiotomy-fourchette > 5 mm	0.5	0.29–0.87	0.014	0.46	0.25–0.83	0.01			NS			
Distance of perineal body (mm)	0.91	0.86–0.96	0.001	0.9	0.84–0.97	0.005	0.088	0.8–0.97	0.011	0.85	0.76–0.95	0.000
Distance of genital hiatus > 25 mm			NS						NS			
Parity			Nulipara						Multipara			
Angle of episiotomy > 30°	0.20	0.13–0.31	0.000	0.21	0.13–0.34	0.000			NS			
Length of episiotomy > 25 mm			NS						NS			
Distance episiotomy-fourchette > 5 mm	0.51	0.32–0.8	0.004	0.46	0.27–0.78	0.004			NS			
Distance of perineal body > 30 mm	0.47	0.32–0.68	0.000	0.42	0.27–0.65	0.000			NS			
Distance of genital hiatus > 25 mm			NS	NS					NS			
Position			Occipitoposterior						Occipitoanterior			
Angle of episiotomy > 30°	0.22	0.09–0.55	0.001	0.19	0.07–0.54	0.002	0.19	0.11–0.31	0.000	0.17	0.1–0.3	0.000
Length of episiotomy > 25 mm			NS						NS			
Distance episiotomy-fourchette > 5 mm			NS				0.63	0.37–0.95	0.071	0.54	0.3–0.96	0.035
Distance of perineal body > 30 mm			NS				0.35	0.22–0.54	0.000	0.28	0.17–0.46	0.000
Distance of genital hiatus > 25 mm			NS						NS			

CI, confidence interval; OR, odds ratio.
[a] Adjusted for: maternal age, parity, daytime obstetrics, birthweight, fetal head position.

distance to fourchette (>5 mm) in nulliparous, perineal bodies ≤30 mm and occipitoanterior position.

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Contribution to authorship

- E Gonzalez Díaz: Conception and Design, Data collection, Data analysis, Manuscript writing
- C Fernández Fernández: Manuscript writing
- JM Gonzalo Orden: Supervision
- A Fernández Corona: Supervision

All authors assisted in the critical revision of the manuscript and have read and approved the final version of the article.

Ethical approval

The *Comité Etico de Investigacion Clinica de Leon* approved the study protocol.

Disclosure of Interests

None declared.

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