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RESEARCH ARTICLE

Incidence of Risk Factors for and Evolution of Depression in Prenatal and Postpartum Pregnant Women: A Prospective Cohort Study

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Abstract

Background: Postpartum depression (PPD) is associated with severe adverse maternal outcomes, significantly contributing to avoidable maternal mortality. In Brazil, most studies on the incidence of and risk factors for PPD have been cross-sectional and conducted in public maternity hospitals. Given the scarce evidence of studies on populations of pregnant women in private hospitals, this study was conducted with pregnant women at private maternity hospitals.

Objectives: The objectives of this study were to verify the incidence of depression and evaluate the risk factors for depression in pregnant women in the prepartum period, in the immediate postpartum period, and at six and twelve weeks postpartum.

Method: This prospective cohort study included pregnant women (gestational age of ≥ 38 weeks) older than 18 yo, a physical status class II and III, and undergoing delivery (cesarean section, vaginal or instrumental). The incidence of PPD, as determined using the EPDS depression scale, was assessed in the prepartum period, 24 to 48h postpartum (PPi), and 6 and 12 weeks postpartum (PP 6 weeks and PP 12 weeks). The patients were divided into two groups: EPDS score ≥ 11 and EPDS score < 11. The null hypothesis was rejected if the two-tailed P value was < 0.05.

Results: 200 patients were selected, and 53 were excluded from the study for distinct reasons. Therefore, 147 patients started the study. The final 12-week sample included 117 patients. No patient had a family history of depression, and none reported using illicit drugs. The medians and 25th and 75th percentiles of EPDS scores at the different time points of the study were: pre delivery, 5 [3-8]; PPi, 4 [2-7]; PP 6 weeks and PP 12 weeks 3 [2-5] (p = 0.0286). There was a statistically significant reduction in the incidence of PPD (p < 0.0001) between the four study moments, and only the incidence of PPD at the Pre-del and PPi time points was significantly different (p = 0.067). Preoperatively, only the number of previous pregnancies and history/presence of psychiatric disease were significantly different between the EPDS score ≥ 11 group and EPDS score < 11 group, with a higher incidence suggestive of PPD in patients with more than one previous pregnancy and in those with a history of psychiatric disease. Of the patients with a history of psychiatric illness, two were treated with antidepressants and had an EPDS score < 11, and the other, without treatment, had an EPDS score > 11. Patients with an EPDS score ≥ 11 at PPi, PP6 weeks, and PP12 weeks already had an EPDS score ≥ 11 pre-delivery.

Conclusions: This study, conducted in a private maternity hospital, showed a 12.9% incidence of PPD in the prepartum period, which decreased in the immediate postpartum period until the end of follow-up, i.e., at 12 weeks postpartum. History of untreated psychiatric illness and multiparity were the only risk factors associated with a higher incidence of PPD.

Keywords

Depression, Postpartum, Pregnant women, Risk factors

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Abbreviations

PPD: Postpartum Depression; EPDS: Edinburgh Postnatal Depression Scale; STROBE: Strengthening of Reporting of Observational Studies in Epidemiology; ASA: American Society of Anesthesiologists; Nbs: Newborns; ICU: Intensive Care Unit; Pre-Del: Pre-Delivery; Ppi: 24 to 48H Postpartum; PP 6 Weeks: 6 Weeks Postpartum; PP 12 Weeks: 12 Weeks Postpartum; BMI: Body Mass Index; ACOG: American College of Obstetricians and Gynecologists

Introduction

Postpartum depression (PPD) is a serious health problem, with an increasing incidence in the last 15 years, affecting 1 in 7 postpartum women in the United States. A recent meta-analysis that included studies conducted in forty-six countries estimated the overall rate of PPD at 17.7%. In Brazil, studies have reported an incidence of PPD between 7 and 39% [1-3].

PPD is associated with severe adverse maternal outcomes, significantly contributing to avoidable maternal mortality. Evidence suggests that routine screening for depression in pregnant and postpartum women may improve the detection and management of perinatal depression, reducing depressive symptoms in these women and the incidence of depression in a given population. Evidence for pregnant women is more scarce but is consistent with that for women in the postpartum period regarding the benefits of screening and treatment and the accuracy of triage instruments [4-13].

The majority of women at risk for PPD can be identified before delivery based on already-identified risk factors, including psychiatric history, depressive symptoms during pregnancy and recent psychosocial stressors. Antenatal depression is the strongest predictor of postpartum depression. Research has confirmed that the levels of symptoms of depression, anxiety and stress vary throughout pregnancy and that prenatal depression is more prevalent than postnatal depression, peaking in the third trimester, suggesting that a proportion of women with postpartum depression experience a continuation of symptoms from the prenatal period [9,14-20].

Screening for depression involves the use of instruments, and the most frequently used PPD screening scale is the Edinburgh Postnatal Depression Scale (EPDS). Although it was created with the primary objective of evaluating PPD, subsequent studies have validated its use in women during pregnancy and in non-obstetric situations [21-28].

In Brazil, most studies on the incidence of and risk factors for PPD have been cross-sectional and conducted in public maternity hospitals, identifying the following risk factors: Marital status, age, low education, low socioeconomic level, multiparity, smoking, use of alcohol and drugs, domestic violence, and previous history of depression. Given the scarce evidence of studies on populations of pregnant women in private hospitals, this study was conducted with pregnant women at private maternity hospitals [7,20,23,28,29].

Objectives

Main objective: To verify the incidence of depression in pregnant women in the prepartum period, in the immediate postpartum period, and at six and twelve weeks postpartum.

Secondary objective: To evaluate the risk factors for depression in pregnant women in the prepartum period, in the immediate puerperium, and at six and twelve weeks postpartum.

Method

Setting and design

A prospective cohort study was conducted to evaluate pregnant women with a gestational age of at least 38 weeks hospitalized for delivery during the period from 11/15/2021 to 03/15/2022.

The results of this study are reported in accordance with the STROBE (Strengthening of Reporting of Observational Studies in Epidemiology) guidelines for cohort studies [4].

Participants

The study population included pregnant women (gestational age of at least 38 weeks) older than 18 years of age who were physical status class II and III according to the American Society of Anesthesiologists (ASA) and undergoing delivery (cesarean section, vaginal or instrumental). All patients whose newborns (NBs) died and/or required ICU admission after delivery and those who were unable to read or understand Portuguese were excluded.

Measures

The incidence of PPD, as determined using the EPDS depression scale, was assessed in the prepartum period, 24 to 48h postpartum (PPi), and 6 and 12 weeks postpartum (PP 6 weeks and PP 12 weeks).

The EPDS is a self-report Likert-type scale that contains ten items (with four response options for each item), with the total score ranging from 0 to 30. The higher the score, the greater is the risk of developing depression/the greater are the depressive symptoms, and the cutoff points for distinguishing clinical depression in prenatal care were statistically predetermined for different languages and cultures. In Brazil, studies have indicated cutoff points between 10 and 13, with values ≥ 11 being considered the best cutoff point for screening for PPD, with 82.6% sensitivity and 65.4% specificity. The cutoff point for PPD screening in this study was a score equal to or greater than eleven points [2,27,28,30,31].

The following potential risk factors for PPD were evaluated based on biological plausibility and risk factors previously studied in the literature: Age, physical status (ASA class), BMI (weight/height^2), education level, number of previous pregnancies, presence of comorbidities, presence or history of psychiatric illness, family history of depression, use of medication(s), use of illegal drugs, anesthesia or analgesia technique, type of delivery, Apgar score at 1 and 5 min and length of hospital stay.

Procedures

Patients were recruited, from 8:00 am to 5:00 pm, based on the availability of the researchers who collected the data. After the pre anesthesia evaluation, the pregnant women were evaluated by a member of the research team, and those who met the inclusion criteria received information about the study and were invited to participate. Those who decided to participate signed a free and informed consent form. The pregnant women were then asked to complete a printed copy of the EPDS [27].

Within 24 to 48h after delivery, the pregnant women were asked to complete another printed copy of the EPDS. For pragmatic reasons, the mothers were allowed to complete the EPDS at a time of the day convenient for them if they were resting or breastfeeding. At 6 and 12 weeks postpartum, one of the researchers conducted a telephone interview, during which they were asked to verbally answer the questions on the EPDS.

Sample size

A convenience sample (n = 200) was used, as the number of patients included was limited by the eligibility criterion (evaluation of PPD up to the 12th postpartum week).

Ethical considerations and consent to participate

Ethical clearance was obtained from the Ethics Committee for Research on Human Beings of the Santa Joana Hospital and Maternity Hospital (50167421.8.0000.5443). Informed consent was obtained from each participant. Confidentiality and anonymity were ensured by excluding the names of the participants on the questionnaire, emails of participants were not collected as well. The participants were assured that their participation is entirely voluntary.

Statistical analysis

For the analysis of risk factors, the patients were divided into two groups based on the EPDS results in the preoperative period: EPDS score ≥ 11 and EPDS score < 11.

Descriptive statistics for quantitative variables are reported as means and standard deviations for data with a normal distribution or medians and inter-quartile ranges for data with a non-normal distribution. Student's *t* test was used to compare the means of quantitative variables with a normal distribution, and the Mann-Whitney test was used to compare the means of variables with non-normal distribution. For qualitative variables, absolute frequencies and percentages are reported, and comparisons were performed using Fisher's test or the x^2 test. Statistical analysis was performed using Graph Pad Prism (9.5.1, USA) and R 4.0.5 software (R Core Team, 2021). The null hypothesis was rejected if the two-tailed P value was < 0.05.

Results

Two hundred patients were selected, and 53 were excluded from the study for distinct reasons (age < 18 years, gestational age < 38 weeks and 17 with newborns admitted to the ICU). Therefore, 147 patients started the study and remained in the study until PPi. The final 12-week sample included 117 patients (Figure 1).

Patient characteristics

The mean age of the 147 study participants was 31.8 ± 5.6 years, mean weight was 30.8 ± 5.4 kg.cm^{-2}, and the mean length of hospital stay was 3.0 ± 0.6 days. Most patients completed higher education (66%) and were married (64%) (Table 1). All patients were ASA II and received dual block analgesia (30.6%) or spinal anesthesia (69.4%). A total of 44.9% of the patients had comorbidities: hypertensive disease of pregnancy (n = 23), hypothyroidism (18), anemia (n = 10), myoma (n = 6), thrombophilia (n = 5) and gestational diabetes mellitus (n = 4).

Most patients were nonsmokers (95.2%) and had no history or presence of psychiatric illness (96.6%), and the newborns (NBs) had Apgar scores > 7 at 1 min (96.6%). No patient had a family history of depression, none reported using illicit drugs, and all fifth-minute Apgar scores were greater than 7.

Most patients did not use medication (68.7%). Among medications, drugs for the treatment of hypothyroidism were predominant (43.5%), followed by antihypertensive drugs (28.3%), antidepressants (11%), anticoagulants (6.5%), drugs to treat anemia (6.5%) and oral hypoglycemic agents (4.2%).

Incidence of PPD

The medians and 25th and 75th percentiles of EPDS scores at the different time points of the study were as follows: Pre-delivery, 5 [3-8]; PPi, 4 [2-7]; PP 6 weeks and PP 12 weeks 3 [2-5]. In a comparison of the 4 time points, p = 0.0286 (Mann-Whitney test).

Table 2 shows the incidence of EPDS score ≥ 11 and EPDS score < 11 at the different time points the study. There was a statistically significant reduction in the incidence of PPD (p < 0.0001) between the four study moments. When analyzing the time points separately, only the incidence of PPD at the Pre-del and PPi time

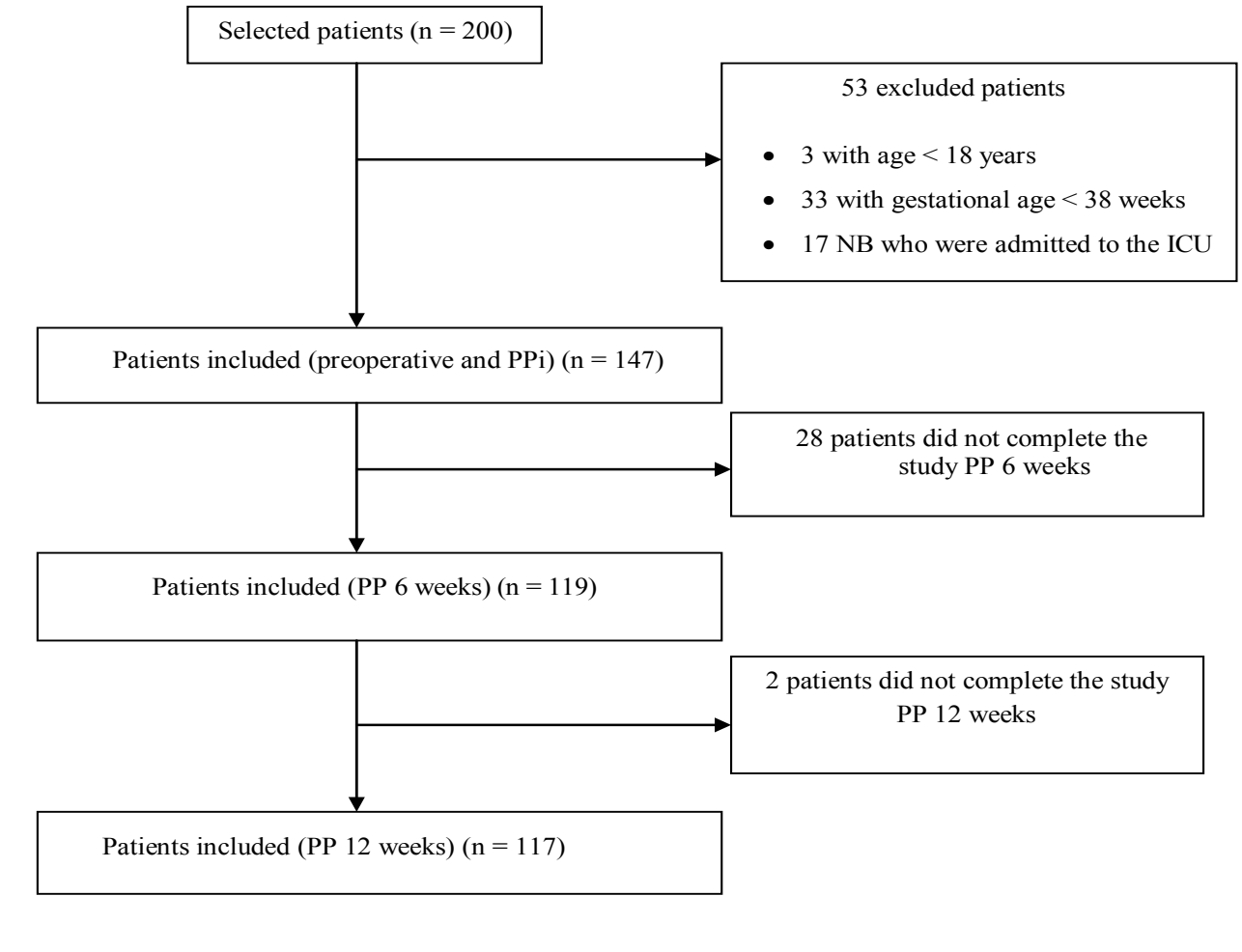

Figure 1: Study flow chart.

Table 1: Patient characteristics. Data are presented as the number of patients (%).

Characteristics	**n (%)**		**n (%)**
Age (years)		**No. of previous pregnancies**	
≤ 25 a	19 (12.9)	0	61 (41.5)
26-35 a	89 (60.5)	1	58 (39.5)
≥ 36 a	39 (26.5)	> 1	28 (19.1)
BMI (kg.cm^{-2})		**Marital status**	
18-24.9	25 (17.1)	Married	114 (77.6)
25-29.9	51 (34.7)	Single	32 (21.8)
≥ 30	71 (48.3)	Divorced	1 (0.6)
Types of delivery		**Education**	
Vaginal delivery	30 (20.4)	< Higher education	26 (12.9)
Cesarean section	114 (77.5)	≥ Higher education	121 (87.1)
Forced delivery	3 (2.1)		
Emergency cesarean section			
Yes	37 (32.4)		
No	77 (67.6)		
BMI: Body Mass Index			

points was significantly different (p = 0.067). Although there was a decrease in PPD incidence from PPi (4.08%) to PP 6 weeks and PP 12 weeks (1.6% and 1.7%, respectively), there was no significant difference.

Patients who had an EPDS score ≥ 11 at PPi, PP6 weeks and PP12 weeks already had an EPDS score ≥ 11 pre-delivery. No patient presented a score suggestive of PPD only in the postpartum period, i.e., No patient

Table 2: EPDS results at the different study times.

	Pre-del		PPi		PP 6 weeks		PP 12 weeks		c^2	P
	n	%	n	%	n	%	n	%		
EPDS score ≥ 11	19	12.9%	9	6.1%	2	1.6%	2	1.7%	21.2	< 0.0001
EPDS score ≥ 11	128	87.1%	138	93.9%	117	98.4%	115	98.3%		
Total	147		147		119		117			

Pre-del = predelivery; PPi = immediate postpartum; PP 6 and 12 weeks = 6 and 12 weeks postpartum; PSED = Postpartum Depression Scale

Table 3: Differences between the variables for patients with EPDS scores ≥ 11 in the predelivery and immediate postpartum periods.

Age	BMI (kg.cm^{-2})	Medication use	Education	Type of delivery	Psychiatric history
21	26.81	No	High school	Vaginal delivery	No
39	37.26	Antihypertensive	Complete university degree	Cesarean section	No
29	27.73	No	Complete university degree	Cesarean section	No
26	28.73	No	Complete university degree	Cesarean section	No
16	28.30	No	High school	Vaginal delivery	Yes
33	28.44	No	Complete university degree	Cesarean section	No
35	40.48	No	Complete university degree	Vaginal delivery	No
31	32.67	No	High school	Cesarean section	No
38	29.76	No	Complete university degree	Cesarean section	No
29.8 ± 7.7*	31.1 ± 4.7*				

BMI: Body Mass Index; *average ± standard deviation

Table 4: Differences between the variables for patients with EPDS scores ≥ 11 throughout the study period.

Age (years)	BMI (kg.cm^{-2})	No of previous pregnancies	Medication use	Education	Type of delivery
21	26.8	0	None	High school	Vaginal delivery
39	37.2	2	Antihypertensive	Complete higher education	Cesarean section

BMI: Body Mass Index

presented symptoms suggestive of PPD after delivery.

Of the 19 patients with EPDS scores suggestive of PPD pre-delivery and PPi, nine patients maintained EPDS scores ≥ 11 in PPi (Table 3). Only one had a history/presence of psychiatric illness but was not taking antidepressants and had no family history of depression. The number of previous pregnancies was similar among the nine patients (0 = 3 patients, 1 = 3 patients, > 1 = 3 patients). The nine patients had no other risk factors.

Only two patients maintained EPDS scores suggestive of PPD throughout the study period; both had no history/presence of psychiatric disease or a family history of depression, were not taking antidepressants, and had no other risk factors present. The variables that differed between the two are listed in the table below (Table 4).

Factors associated PPD

Preoperatively, among the risk factors for PPD studied, only number of previous pregnancies and history/presence of psychiatric disease were significantly different between the EPDS score ≥ 11 group and EPDS score < 11 group, with, respectively, a higher incidence suggestive of PPD in patients with more than one previous pregnancy and in those with a history of psychiatric disease (Table 5). Of the patients with a history of psychiatric illness, two were treated with antidepressants and had an EPDS score < 11, and the other three, without treatment, had an EPDS score > 11.

The separate analyses of age and BMI showed mean and standard deviation values of 31.5 ± 6.5 years (EPDS score ≥ 11 group) and 31.8 ± 5.5 years (EPDS score < 11 group) and 30.7 ± 4.3 kg.cm^{-2} (EPDS score ≥ 11 group) and 30.8 ± 5.6 kg.cm^{-2} (EPDS score < 11 group), respectively, with p values = 0.951 and 0.856 (unpaired Student's *t* test).

There was no significant difference between patients taking medication or not in relation to the incidence of PPD in the pre-delivery period (p - 0.997). Regarding the patients who used medication(s), there was a significantly higher incidence of EPDS score ≥ 11 (60%) among those who took antidepressants than among those who took other medication (7.8%) and those who did not use any medication (12.9%) (p =

Table 5: Incidence of EPDS score ≥ 11 and < 11 in the preoperative period for the preoperative risk factors.

	Total	EPDS score ≥ 11	EPDS score < 11	
	n (%)	n (%)	n (%)	Q
Age (years)				
≤ 25 a	19 (12.9)	3 (15.8)	16 (88.2)	0.751*
26-35 a	89 (60.5)	10 (11.2)	79 (88.8)	
≥ 36 a	39 (26.5)	6 (15.4)	33 (84.6)	
BMI (kg.cm^{-2})				
18-24.9	25 (17.1)	1 (4.0)	24 (96.0)	0.871*
25-29.9	51 (34.7)	9 (17.6)	42 (82.4)	
≥ 30	71 (48.3)	9 (12.7)	82 (87.3)	
No. of previous pregnancies				
0	61 (41.5)	5 (8.2)	56 (91.8)	**0.022***
1	58 (39.5)	6 (10.3)	52 (89.7)	
> 1	28 (19.1)	8 (28.6)	20 (71.4)	
Marital status				
Married	114 (75.5)	15 (13.4)	96 (88.6)	0.749*
Single	35 (23.8)	4 (11.4)	31 (88.6)	
Divorced	1 (0.7)	0	1 (100)	
Education				
< Higher education	26 (12.9)	3 (12.0)	22 (88.0)	> 0.999**
≥ Higher education	121 (87.1)	16 (13.1)	106 (86.9)	
Types of delivery				
Vaginal delivery	30 (20.4)	2 (6.7)	28 (93.3)	0.376*
Cesarean section	114 (77.5)	17 (14.9)	97 (85.1)	
Forced delivery	3 (2.1)	0	3 (100.0)	
Emergency cesarean section				
Yes	37 (32.4)	6 (16.2)	31 (83.8)	> 0.999**
No	77 (67.6)	13 (16.9)	64 (83.1)	
Type of anesthesia				
Double blocking	45 (30.6)	5 (11.1)	40 (88.9)	0.793**
Spinal anesthesia	102 (69.4)	14 (13.7)	88 (86.3)	
Psychiatric illness (Hist/Pres)				
Yes	5 (3.4)	3 (60.0)	2 (40.0)	**0.016****
No	142 (96.6)	16 (11.3)	126 (88.7)	
Comorbidities				
Yes	66 (44.9)	9 (13.6)	57 (86.4)	0.881**
No	81 (55.1)	10 (12.3)	71 (87.7)	
Smoking				
Yes	7 (4.8)	0	7 (100.0)	> 0.999**
No	140 (95.2)	19 (13.6)	121 (86.4)	
Apgar 1st minute				
< 7	5 (3.4)	0)	5 (100.0)	> 0.999**
≥ 7	142 (96.6)	18 (12.2)	129 (90.8)	

BMI: Body Mass Index; chi-square (c^2)* or Fisher's** test; Hist/Pres = history/presence

0.024 and p = 0.12, respectively). Of the 13 patients taking antihypertensive drugs, eight were using alpha-methyldopa, and only one had an EPDS score ≥ 11 pre-delivery, which returned to < 11 in the postoperative follow-up period.

The incidence of PPD (EPDS score ≥ 11), of the variables emergency cesarean section and 1-minute Apgar were assessed at PPi, and there was no statistically significant differences between patients who underwent emergency or elective cesarean section

and those whose NBs had a 1-minute Apgar score < 7 or ≥ 7.

Discussion

In this prospective cohort study, the incidence of PPD was assessed using the EPDS and patients at 38 weeks of gestation or more before delivery (pre-delivery period) and in the immediate puerperium period and six and twelve weeks postpartum. The incidence of PPD was 12.9% in the pre-delivery period and in the immediate PPD, being more frequent in pregnant women with a greater number of children and in those with a psychiatric history without treatment. Patients with a history of psychiatric illness who were treated did not have symptoms suggestive of PPD, as determined using the EPDS.

The comparison of our findings with results reported in the literature is difficult because systematic reviews and meta-analyses have shown great heterogeneity in the studies analyzed, including in number of patients, EPDS cutoff point (9 to 13), and PPD evaluation time points (pre-delivery period to after pregnancy), with different results, leading to a higher incidence of PPD [32-37].

In this study, there was a reduction in the incidence of PPD in the late postpartum period, i.e., 6 and 12 weeks postpartum. There was a 20% loss to follow-up of patients who did not complete the study within 12 weeks of PP. Of these, 4 patients who had EPDS scores suggestive of PPD at the initial time of the study did not remain in the study for 6 weeks postpartum, and 6 more did not remain in the study for 12 weeks postpartum. Studies show a reduction in the incidence of PPD over time, up to 5 years [15,22,36].

Although there was no significant difference, the incidence of PPD was 2.2 times higher among women who underwent cesarean section than among women who underwent a vaginal/forceps delivery (14.9% and 6.7%), a finding that agrees with the results of a meta-analysis, i.e., compared to vaginal delivery, cesarean delivery is associated with an increased risk of PPD. The possible mechanisms include delayed first interaction between mother and child and first breastfeeding attempt, postpartum pain and incompatibility between women's expectations for childbirth and the care provided [33,38]. The lack of a higher incidence of PPD with cesarean section may be the result of conducting the study in a private maternity hospital, where many cesarean sections are elective (67.4%), performed by maternal desire.

The sample in the present study consisted of pregnant women at a private maternity hospital, and probably because of this, the majority of pregnant women had higher education (87.1%), a finding that differs from the results of other studies; for this reason, it was not possible to evaluate the influence of schooling on the incidence of PPD [33,39-42].

Most of the pregnant women were married, and thus, it was not possible to evaluate the correlation between marital status and EPDS scores. One of the risk factors considered relevant in the literature is the history of child abuse and violence by an intimate partner, e.g., the husband. This factor was not evaluated in our study [2,33,39,40].

Another risk factor for PPD cited in the literature is fetal death, which leads to a higher incidence of PPD, sometimes leading to suicidal ideation. In the United Kingdom, maternal suicide is the leading cause of direct maternal deaths from 6 weeks to 1 year postpartum. There were no fetal deaths in this study [12,42].

In the analysis of the risk factor type of anesthesia, there was no difference between analgesia performed with double blockade or spinal anesthesia in relation to the incidence of PPD. A systematic review and meta-analysis concluded that pregnant women who underwent epidural analgesia for vaginal delivery had lower rates of PPD in the short and long term after delivery than did those who received other analgesia techniques. In another study, compared to patients under spinal anesthesia, pregnant women undergoing cesarean section under general anesthesia showed a significantly higher risk of PPD and greater need for antidepressant [41-43].

In a study with 100 pregnant women, Nayak, et al. found a statistically significant risk of postpartum depression associated with the use of alpha-methyldopa during pregnancy; 9 patients took methyldopa, and 7 had EPDS scores > 10, which was considered by the authors as the cutoff point for depression [43]. In the present study, 9 patients were using methyldopa, and 1 had an EPDS score > 11, only pre-delivery.

Although there are several PPD screening tests recommended by the American College of Obstetricians and Gynecologists (ACOG) (Postpartum Depression Screening Scale, Beck Depression Inventory, Center for Epidemiologic Studies Depression Scale and Zung Self Rating Depression Scale), in the present study, the EPDS was used as the PPD screening scale because it is the validated screening test most frequently used worldwide, has good accuracy, and is a quick and easy-to-understand test that can be self-applied [11,25].

Limitations

This present study was conducted with a limited number of patients; however, the study can be replicated in the future in a multicenter study with a larger sample size. Another possible limitation is that the patients were asked by telephone to answer questions on the EPDS, thus having to recall episodes of PPD. In addition, we did not verify economic status and partner support, which have already been shown to influence

PPD rates. Likewise, although patients were asked whether they had preexisting depression and were treated with medication or not and the medical records indicated preexisting depression, we may have missed undiagnosed or underreported baseline depression. Another limitation of the study was the non inclusion of pregnant women under 18 years of age, as some studies have indicated that this population is prone to PPD.

Conclusions

This study, conducted in a private maternity hospital, showed a 12.9% incidence of PPD in the prepartum period, which decreased in the immediate postpartum period until the end of follow-up, i.e., at 12 weeks postpartum. History of untreated psychiatric illness and multiparity were the only risk factors associated with a higher incidence of PPD.

The results of this study highlight the importance of always performing PPD screening during and after pregnancy in all pregnant women.

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Conflicts of Interest

The authors have no conflicts of interest relevant to this article to disclose.

Author Statement

EARA, CAPS, DSSC, MMS, and LASTM conceptualized and designed the study; LASTM performed the statistical analyses and drafted the manuscript; EARA, CAPS, DSSC, and MM Shelped draft the manuscript; All authors commented and contributed to successive drafts, and read and approved the final manuscript as submitted, and agree to be accountable for all aspects of the work.

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