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Study of first-trimester serum levels of β-hCG and PAPP-A as a screening test for fetal development of intrauterine growth restriction

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Abstract

Objective To evaluate the association between first-trimester serum levels of pregnancy-associated plasma protein-A (PAPP-A) and free beta-human chorionic gonadotropin (β-hCG) and the development of intrauterine growth restriction (IUGR), in order to assess their utility in early screening for improved perinatal outcomes.

Methods A retrospective case–cohort study was conducted at Marqués de Valdecilla University Hospital in 2021, including 119 pregnancies with IUGR and a randomly selected subcohort of 383 pregnancies from the same population. Serum levels of PAPP-A and β-hCG were analyzed both as continuous variables and as categorical variables based on population-specific percentiles (< 10th and < 5th). Logistic regression models were used to estimate odds ratios (ORs) and 95% confidence intervals (CIs), adjusting for relevant maternal and obstetric covariates.

Results Lower PAPP-A levels were significantly associated with an increased risk of IUGR, both as a continuous variable (OR = 0.50; 95% CI: 0.34–0.76, $p = 0.001$) and when categorized below the 10th percentile (OR = 4.01; 95% CI: 1.78–9.01, $p < 0.001$) and 5th percentile (OR = 4.45; 95% CI: 1.57–12.63, $p = 0.005$). β-hCG showed no association when analyzed continuously ($p = 0.164$), but values below the 10th percentile were significantly associated with IUGR (OR = 4.45; 95% CI: 1.97–10.04, $p < 0.001$). No reliable estimate could be obtained at the 5th percentile due to the small number of cases.

Conclusion Incorporating PAPP-A and β-hCG into first-trimester screening protocols could enable earlier identification of pregnancies at risk of IUGR, facilitating timely interventions and potentially improving maternal and neonatal outcomes. These findings support the clinical utility of these biomarkers in routine obstetric care.

Keywords Intrauterine growth restriction, First-trimester screening, PAPP-A, β-hCG, Maternal-fetal health, Perinatal outcomes

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Introducción

The first-trimester combined screening test (11 + 0 to 13 + 6 weeks) for chromosomal abnormalities includes maternal serum markers—PAPP-A and β-hCG—along with nuchal translucency (NT) and maternal age. While the association of these markers with aneuploidies is well established, growing evidence suggests that altered levels, particularly of PAPP-A, may also be linked to adverse obstetric outcomes such as intrauterine growth restriction (IUGR) [1–3].

PAPP-A is a zinc-binding enzyme that belongs to the metalloproteinase family (metzincins). This glycoprotein is primarily secreted by the trophoblast of the placenta. In uncomplicated pregnancies, PAPP-A is detectable in maternal blood from implantation, with its concentration increasing as gestational age progresses, reaching peak levels at the end of pregnancy. PAPP-A interacts with insulin-like growth factors (IGF). When PAPP-A levels are insufficient to activate IGF, the latter remains in its inactive form, leading to impaired fetal and placental development. This can result in a significant reduction in placental volume and alterations in fetal growth [1]. Additionally, decreased PAPP-A levels have been associated with impaired fetal growth and adverse pregnancy outcomes [4–6].

β-hCG is a glycoprotein composed of two amino acid chains (alpha and beta) linked by a disulfide bond. It is secreted by the syncytiotrophoblast and can be detected in maternal blood as early as the ninth or tenth day after ovulation. Its concentration rises rapidly, peaking at the end of the first trimester before gradually declining. Elevated β-hCG levels are associated with placental proliferation and invasion, whereas decreased levels may indicate trophoblastic insufficiency and an increased risk of pregnancy complications, including IUGR [1]. Similarly to PAPP-A, low levels of β-hCG in fetuses without abnormalities have also been associated with fetal development issues and pregnancy complications [1]. Although better known for its role in early pregnancy maintenance, low β-hCG levels have also been associated with placental dysfunction and increased risk of adverse outcomes, including IUGR [1].

Fetal intrauterine growth restriction (IUGR) is a condition that develops in 20–30% of small-for-gestational-age fetuses during pregnancy. In Europe, it is estimated to affect up to 10% of pregnancies [7]. IUGR is one of the major contributors to adverse perinatal outcomes and a significant risk factor for neonatal morbidity and mortality. Its pathophysiology primarily involves chronic vasoconstriction of the tertiary stem villi in the placenta, caused by inadequate trophoblastic invasion of the maternal spiral arteries. Currently, the most commonly used screening method begins with a second-trimester ultrasound (18–20 weeks) using Doppler studies of the maternal uterine arteries. Although the possibility of advancing screening to the first trimester has been explored, there is still insufficient evidence to support this change [8]. Earlier screening could enable timely detection, closer monitoring, and the application of interventions earlier in pregnancy.

In clinical practice, risk calculators are available that incorporate the previously mentioned proteins along with other data [9]. However, no population-based screening protocol for IUGR in the first trimester currently exists, highlighting the importance of this project.

The purpose of this study is to evaluate first-trimester serum levels of PAPP-A and β-hCG and their relationship with the development of intrauterine growth restriction over the course of pregnancy. Our findings may provide a foundation for establishing population-based IUGR screening in the first trimester, enabling earlier monitoring, prevention, and management of this obstetric condition.

Materials and methods

Study design and population

This was a retrospective case–cohort study, conducted at Marqués de Valdecilla University Hospital (HUMV), a tertiary referral center managing approximately 2,000 to 3,000 deliveries annually. The study aimed to evaluate the association between first-trimester biomarkers and intrauterine growth restriction (IUGR) within a well-defined obstetric population.

The study population was drawn from pregnancies resulting in live births between January 1 and December 31, 2021. After applying exclusion criteria—namely, multiple gestations, chromosomal abnormalities, and major congenital malformations—and performing data validation procedures, including consistency checks and removal of duplicate entries, a total of 2,660 eligible pregnancies were identified.

From this population, all pregnancies diagnosed with IUGR ($n = 119$) were included as cases. In parallel, a random subcohort of 383 pregnancies, approximately 15% of the eligible population, was selected as the reference group, regardless of IUGR status. Due to the random sampling procedure, five IUGR cases were also included in the reference group and were retained in both categories to preserve statistical efficiency and avoid bias. This hybrid design, which combines the comprehensive identification of cases with a population-based comparison group, enables efficient risk estimation while maintaining internal validity and representativeness [10, 11].

No additional exclusion criteria, such as chronic maternal diseases or infections during pregnancy, were applied, in order to maintain a representative, real-world sample and enhance the study's external validity. To address potential confounding, key epidemiological and clinical

variables were collected and included in all multivariable models.

Data collection

Epidemiological and clinical data were retrospectively collected from the hospital's electronic obstetric records. Variables included maternal age and body mass index (BMI) at the time of first-trimester screening, parity, race, toxic habits (smoking, alcohol, and drug use), relevant obstetric and medical history, and whether the pregnancy was achieved via assisted reproductive techniques. Additional information was gathered on maternal-fetal complications during pregnancy and postpartum, including miscarriage, fetal death, hypertensive disorders of pregnancy, placental abruption (DPPNI), gestational diabetes, preterm birth, and intrahepatic cholestasis.

First-trimester serum biomarker levels of pregnancy-associated plasma protein-A (PAPP-A) and free beta-human chorionic gonadotropin (β-hCG) were obtained from standardized laboratory reports. These biomarkers were measured from maternal serum samples collected during routine first-trimester screening, conducted between 11 + 0 and 13 + 6 weeks of gestation, in accordance with clinical guidelines. Multiples of the median (MoM) were calculated using population-specific reference medians for each gestational week. When available, first-trimester biochemical risk scores were also recorded.

Neonatal data included gestational age at delivery, type of delivery, Apgar scores, estimated fetal weight, umbilical cord pH, neonatal intensive care unit (NICU) admission, and early perinatal outcomes. All pregnancies were monitored from the first-trimester screening through delivery and up to 48 h postpartum to identify pregnancy- and birth-related complications.

All data were obtained without direct patient contact. The study complied with Spanish legislation on biomedical research and data protection, including the General Data Protection Regulation (GDPR, Regulation EU 2016/679) and Spanish Organic Law 3/2018 on Personal Data Protection and Guarantee of Digital Rights.

Outcomes

The primary outcome of the study was intrauterine growth restriction (IUGR). Cases were defined according to established clinical and ultrasound criteria as pregnancies in which the estimated fetal weight was below the 10th percentile for gestational age, accompanied by abnormalities in fetal blood flow distribution, as assessed by Doppler studies. This definition reflects both biometric and hemodynamic parameters, in line with current recommendations for the diagnosis of fetal growth restriction.

All pregnancies included in the study were monitored from the time of first-trimester screening through delivery and up to 48 h postpartum. This follow-up period allowed for the identification of relevant obstetric and neonatal complications associated with IUGR.

Statistical analysis

Descriptive statistics were used to characterize the study population. Continuous variables were reported as means and standard deviations, along with minimum and maximum values, while categorical variables were summarized using absolute frequencies and percentages. Comparisons between the IUGR and reference groups were conducted using the Student's t-test for continuous variables and the Chi-square test or Fisher's exact test, as appropriate, for categorical variables.

To assess the association between first-trimester biomarkers and intrauterine growth restriction, logistic regression models were fitted. Biomarkers (PAPP-A and free β-hCG) were analyzed both as continuous variables (MoM values) and as categorical variables based on population-specific percentiles (< 10th and < 5th). Crude and adjusted odds ratios (ORs) with 95% confidence intervals (CIs) were estimated.

All multivariable models were adjusted for key maternal and pregnancy-related confounders, including: maternal age, pre-pregnancy body mass index (BMI), smoking, alcohol use, drug use, parity, assisted reproductive technologies, uterine malformations, and gestational anemia. The selection of these variables was based on their established association with IUGR risk and their availability in the dataset.

A two-tailed p-value < 0.05 was considered statistically significant. All statistical analyses were performed using Stata 18/SE software (StataCorp, College Station, TX, USA).

Ethics

The study was approved by the Cantabria Clinical Research Ethics Committee (reference number: 2022.237). As this was a retrospective observational study based on data extracted from medical records, with no direct patient involvement or additional procedures, the requirement for informed consent was waived by the Ethics Committee, in accordance with Royal Decree 957/2020, which regulates observational studies in Spain. The study complies with the ethical principles established in the Declaration of Helsinki (World Medical Association, 2024) [12], as well as with national regulations on biomedical research, including Law 14/2007 of July 3 on Biomedical Research [13].

Results

Population description

The study population included 119 pregnancies diagnosed with intrauterine growth restriction (IUGR) and a randomly selected reference group of 383 pregnancies without IUGR, all drawn from the same cohort of eligible births in 2021.

Table 1 summarizes the clinical characteristics and maternal biomarkers in both groups.

No significant differences were observed in maternal age between IUGR and non-IUGR pregnancies (33.94 ± 5.66 vs. 33.64 ± 5.43 years, $p = 0.604$). However, pre-pregnancy body mass index (BMI) was significantly lower in the IUGR group (23.68 ± 4.24 vs. 25.19 ± 5.29 kg/m², $p = 0.006$), and the prevalence of smoking was notably higher (27.35% vs. 12.34%, $p < 0.001$). Conversely, previous cesarean section was less frequent among IUGR cases (4.20% vs. 11.23%, $p = 0.023$).

With respect to maternal complications, preeclampsia was significantly more common in pregnancies affected by IUGR (18.49% vs. 9.66%, $p = 0.009$), while gestational diabetes was less frequent (2.52% vs. 9.92%, $p = 0.010$). Analysis of first-trimester biochemical markers showed significantly lower PAPP-A levels in the IUGR group (1.06 ± 0.63 MoM vs. 1.38 ± 0.75 MoM, $p < 0.001$), whereas β-hCG levels did not differ significantly between groups (1.23 ± 0.89 MoM vs. 1.42 ± 1.00 MoM, $p = 0.072$).

Perinatal complications were also more frequent among IUGR cases, including higher rates of placental abnormalities (9.24% vs. 3.18%, $p = 0.006$), gestational anemia (46.61% vs. 34.92%, $p = 0.022$), and maternal intensive care unit (ICU) admission (7.56% vs. 1.57%, $p = 0.001$).

Neonatal outcomes

As detailed in Table 2, Apgar scores at 5 min were significantly lower in the IUGR group (9.36 ± 1.28 vs. 9.60 ± 0.90, $p = 0.022$). No significant differences were found in Apgar scores at 1 min ($p = 0.126$) or in umbilical cord pH ($p = 0.424$).

Overall, neonatal complications were more prevalent in IUGR pregnancies, with only 54.62% of these cases presenting without any complication, compared to 88.51% in the reference group ($p < 0.001$). Fetal well-being loss was more frequent among IUGR cases (12.61% vs. 3.39%, $p < 0.001$), as were perinatal death (4.20% vs. 0.78%, $p = 0.009$) and neonatal hospitalization (25.21% vs. 6.53%, $p < 0.001$). Although preterm birth (2.52% vs. 0.52%, $p = 0.055$) and placental abruption (0.84% vs. 0.26%, $p = 0.381$) were more common in the IUGR group, these differences did not reach statistical significance.

Biomarkers and fetal growth restriction severity

Table 3 presents the distribution of first-trimester biomarker levels and estimated fetal weight according to the severity of intrauterine growth restriction (IUGR). Cases were classified into two groups: mild IUGR (6th–10th percentile) and moderate-to-severe IUGR (≤ 5th percentile). Due to the limited number of severe cases ($n = 3$), these were merged with moderate cases to ensure statistical robustness and consistency in the analysis.

In the mild IUGR group, the mean PAPP-A concentration was 1.10 ± 0.68 MoM and β-hCG was 1.32 ± 0.82 MoM, while the estimated fetal weight averaged 2684.24 ± 270.38 g. In contrast, the moderate-to-severe group showed slightly lower levels of both biomarkers (PAPP-A: 1.05 ± 0.62 MoM; β-hCG: 1.20 ± 0.92 MoM)

Table 1 Clinical characteristics and maternal biomarkers in pregnant women with intrauterine growth restriction (IUGR) (Cases) and reference group (No IUGR)

Variable	Reference group (No IUGR) ($n = 383$)	Cases (IUGR) ($n = 119$)	*p*
Age (years), mean (SD)	33.64 (5.43)	33.94 (5.66)	0.604
Body Mass Index (kg/m²), mean (SD)	25.19 (5.29)	23.68 (4.24)	0.006 **
Previous cesarean section, n (%)	43 (11.23)	5 (4.20)	0.023 **
Smoking, n (%)	47 (12.34)	32 (27.35)	< 0.001 **
Preeclampsia, n (%)	37 (9.66)	22 (18.49)	0.009 **
Gestational diabetes, n (%)	38 (9.92)	3 (2.52)	0.010 **
Drug addiction, n (%)	5 (1.32)	3 (2.59)	0.342
PAPP-A (MoM), mean (SD)	1.38 (0.75)	1.06 (0.63)	< 0.001 **
β-hCG (MoM), mean (SD)	1.42 (1.00)	1.23 (0.89)	0.072
Fetal sex (male), n (%)	185 (48.30)	60 (50.42)	0.687
Assisted reproduction, n (%)	35 (9.14)	13 (10.92)	0.607
Placental alteration, n (%)	12 (3.18)	11 (9.24)	0.006 **
Gestational anemia, n (%)	132 (34.92)	55 (46.61)	0.022 **
Maternal ICU admission, n (%)	6 (1.57)	9 (7.56)	0.001 **

Note: Values are presented as mean (standard deviation) for continuous variables and as number (percentage) for categorical variables. Statistically significant differences are indicated with ** ($p < 0.05$)

Table 2 Neonatal outcomes in pregnancies with intrauterine growth restriction (IUGR) and reference group

Variable	Category		Reference group *N* (%)	IUGR (Cases) *N* (%)	*p*-value
APGAR 1 min*, *mean (SD)			8.62 (1.23)	8.41 (1.40)	0.126
APGAR 5 min*, *mean (SD)			9.60 (0.90)	9.36 (1.28)	0.022 **
Ph cord*, *mean (SD)			7.25 (0.08)	7.26 (0.08)	0.424
Birth complications	**No**		44(11.49)	54(45.38)	< 0.001**
	Yes		339(88.51)	65(54.62)	
	Fetal well-being loss	**No**	370(96.61)	104(87.39)	< 0.001**
		Yes	13(3.39)	15(12.61)	
	Premature birth	**No**	381(99.48)	116(97.48)	0.055
		Yes	2(0.52)	3(2.52)	
	Placental abruption	**No**	382(99.74)	118(99.16)	0.381
		Yes	1(0.26)	1(0.84)	
	Perinatal death	**No**	380(99.22)	114(95.80)	0.009**
		Yes	3(0.78)	5(4.20)	
	Neonatal hospitalization	**No**	358(93.47)	89(74.79)	< 0.001**
		Yes	25(6.53)	30(25.21)	

Note: Statistically significant values are indicated with ** ($p < 0.05$). Data are presented as mean (standard deviation) for continuous variables or number (percentage) for categorical variables

Birth complications include the following adverse perinatal events:

– Fetal well-being loss, defined as abnormal fetal heart rate patterns or signs of distress requiring intervention

– Premature birth, defined as delivery before 37 weeks of gestation

– Placental abruption, defined as premature separation of the placenta from the uterine wall, potentially leading to fetal hypoxia or maternal hemorrhage

– Perinatal death, referring to fetal or neonatal death occurring around the time of birth

– Neonatal hospitalization, defined as admission to a neonatal intensive care unit (NICU) due to complications requiring specialized care

Table 3 Characteristics of biomarkers and estimated fetal weight in fetuses with intrauterine growth restriction (IUGR) by severity

Variable		*N*	Min	Max	Mean	SD
Mild growth restriction	*PAPP-A (MoM)*	27	0.34	3.26	1.10	0.68
	β-hCG (MoM)	27	0.33	3.55	1.32	0.82
	Estimated fetal weight	29	2100.00	3200.00	2684.24	270.38
Moderate-to-severe growth restriction (IUGR I–III)	*PAPP-A (MoM)*	80	0.14	3.75	1.05	0.62
	β-hCG (MoM)	80	0.18	4.57	1.20	0.92
	Estimated fetal weight	90	552.00	2900.00	2054.97	478.75
Total	*PAPP-A (MoM)*	107	0.14	3.75	1.06	0.63
	β-hCG (MoM)	107	0.18	4.57	1.23	0.89
	Estimated fetal weight	119	552.00	3200.00	2208.32	513.64

Note: Due to the limited number of severe IUGR cases ($n = 3$), these were grouped under "moderate-to-severe growth restriction" for analytical purposes

Data are presented as mean (standard deviation), minimum, and maximum values for each group

PAPP-A: pregnancy-associated plasma protein A; β-hCG: beta-human chorionic gonadotropin

and a substantially reduced mean fetal weight of 2054.97 ± 478.75 g.

Overall, across the entire IUGR cohort, the mean PAPP-A and β-hCG levels were 1.06 ± 0.63 MoM and 1.23 ± 0.89 MoM, respectively, with an average estimated fetal weight of 2208.32 ± 513.64 g.

Association between PAPP-A, β-hCG, and IUGR

As shown in Table 4, lower PAPP-A levels were significantly associated with an increased risk of IUGR. When analyzed as a continuous variable, PAPP-A was inversely associated with IUGR (OR = 0.50; 95% CI: 0.34–0.76, $p = 0.001$). This association remained significant when categorized: at the 10th percentile, the odds of IUGR were four times higher (OR = 4.01; 95% CI: 1.78–9.01, $p < 0.001$), and at the 5th percentile the risk further increased (OR = 4.45; 95% CI: 1.57–12.63, $p = 0.005$).

For β-hCG, no significant association was observed when assessed as a continuous variable (OR = 0.83; 95% CI: 0.63–1.08, $p = 0.164$). However, β-hCG values below the 10th percentile were associated with a significantly higher likelihood of IUGR (OR = 4.45; 95% CI: 1.97–10.04, $p < 0.001$). Due to the limited number of events, no valid estimate could be obtained at the 5th percentile.

Table 4 Association of PAPP-A and β-hCG, assessed as continuous variables and categorized in the 5th and 10th percentiles, with intrauterine growth restriction (IUGR)

Variable	Category	a/*n*	OR (95% CI)	*p*
PAPP-A (MoM)	Total		0.50 (0.34–0.76)	0.001 **
	p10			
	No	65/366	1(ref.)	
	Yes	14/34	4.01 (1.78–9.01)	< 0.001 **
	p5			
	No	89/421	1(ref.)	
	Yes	9/19	4.45 (1.57–12.63)	0.005**
β-hCG (MoM)	Total		0.83 (0.63–1.08)	0.164
	p10			
	No	65/366	1(ref.)	
	Yes	17/34	4.45 (1.97–10.04)	< 0.001**
	p5			
	No	89/421	1(ref.)	
	Yes	0/1	1.00 (1.00–1.00)	.

Note: The analysis was adjusted for maternal age, pre-pregnancy body mass index (BMI), smoking, alcohol use, drug use, parity, assisted reproductive technologies, uterine malformations, and gestational anemia

Statistically significant values are indicated with ** ($p < 0.05$). Data are presented as odds ratios (OR) and 95% confidence intervals (CI)

1 (ref.) indicates the reference category against which the odds ratio is compared

Cut-off points:

– p5 (5th percentile): PAPP-A or β-hCG values below the 5th percentile of the population distribution

– p10 (10th percentile): PAPP-A or β-hCG values below the 10th percentile of the population distribution

Discussion

Our findings demonstrate a strong association between low PAPP-A levels and the development of intrauterine growth restriction (IUGR). Pregnant women with PAPP-A levels below the 5th percentile exhibited a significantly increased risk of IUGR (OR = 4.45; 95% CI: 1.57–12.63, $p = 0.005$), while levels below the 10th percentile also showed a strong association (OR = 4.01; 95% CI: 1.78–9.01, $p < 0.001$). This reinforces the role of PAPP-A as an early predictive biomarker, in line with previous literature such as Papamichail et al. (2020), which reported an association between low PAPP-A and placental insufficiency and IUGR risk [14]. Similarly, Gupta et al. (2015) described a 21.5% prevalence of IUGR among women with reduced PAPP-A levels, highlighting the need for closer monitoring [4]. Shah et al. (2020) further reported a twofold increased risk of low birth weight in pregnancies with reduced PAPP-A levels [6].

The biological role of PAPP-A helps explain this association. As a zinc-binding metalloproteinase, PAPP-A modulates IGF bioavailability, promoting trophoblastic invasion and adequate spiral artery remodeling—key processes for optimal placental vascularization and fetal growth. Insufficient PAPP-A levels impair IGF activation, leading to poor trophoblastic invasion, chronic vasoconstriction of tertiary stem villi, and reduced placental volume, ultimately contributing to fetal growth restriction [1].

Regarding β-hCG, our data suggest a more modest but still relevant role. While β-hCG levels did not show a significant association with IUGR when analyzed as a continuous variable (OR = 0.83; 95% CI: 0.63–1.08, $p = 0.164$), levels below the 10th percentile were significantly associated with an increased risk (OR = 4.45; 95% CI: 1.97–10.04, $p < 0.001$). This suggests a potential complementary utility of β-hCG in early screening. However, at the 5th percentile, no valid estimate could be derived due to the small number of cases. These findings are partially aligned with prior studies such as Dugoff et al. (2004) and Rodríguez-Zurita et al. (2021). Dugoff et al. (2004) found that β-hCG levels below the 1st percentile were associated with fetal growth restriction before 24 weeks [15]. Likewise, Rodríguez-Zurita et al. (2021) and Yiming Chen et al. (2024) reported a correlation between low β-hCG levels and IUGR development [1, 16]. These associations may be explained by β-hCG's role in trophoblastic invasion and placental development—key processes that, when impaired, contribute to placental insufficiency [16, 17].

A trend toward a progressive decline in biomarker levels and estimated fetal weight was observed in relation to IUGR severity. PAPP-A and β-hCG levels were lower in moderate-to-severe IUGR cases compared to mild cases (PAPP-A: 1.05 vs. 1.10 MoM; β-hCG: 1.20 vs. 1.32 MoM), supporting their biological plausibility in the pathogenesis of fetal growth restriction. Furthermore, estimated fetal weight was notably lower in more severe forms of IUGR (2055 g vs. 2684 g), reinforcing the association between biochemical markers and clinical severity.

In terms of maternal and perinatal characteristics, our study confirmed known risk factors for IUGR, such as lower maternal BMI ($p = 0.006$), higher smoking prevalence ($p < 0.001$), and increased incidence of preeclampsia ($p = 0.009$). The association between IUGR and increased perinatal morbidity in our study—evidenced by significantly lower Apgar scores at 5 min ($p = 0.022$), higher neonatal hospitalization ($p < 0.001$), and greater incidence of perinatal death ($p = 0.009$)—further underscores the clinical relevance of early detection. These findings align with prior research [18, 19].

A biological explanation for these findings may lie in the mechanisms of placental dysfunction. Insufficient remodeling of spiral arteries leads to high vascular resistance, reduced villous surface area, and placental infarctions, impairing oxygen and nutrient delivery to the fetus. This hypoxic environment triggers cellular stress within the placenta, suppressing protein synthesis and limiting proliferation, increasing the risk of IUGR [20].

These findings are further supported by recent evidence suggesting that abnormal first-trimester levels of PAPP-A and β-hCG—often measured as part of routine aneuploidy screening—may reflect early placental dysfunction beyond chromosomal anomalies. In particular, Caveretto et al. (2024) demonstrated that these markers are associated with increased risk of spontaneous birth and hypertensive disorders in pregnancies without preeclampsia but classified as high-risk based on first-trimester screening results [21]. This reinforces the idea that these biomarkers capture broader aspects of placental maladaptation relevant to fetal growth.

Moreover, Spinillo et al. (2022) provided meta-analytic evidence linking low levels of PAPP-A and β-hCG with confined placental mosaicism (CPM), a placental abnormality characterized by the presence of genetically distinct cell lines restricted to the placenta [22]. CPM is a known contributor to placental insufficiency, IUGR, and small-for-gestational-age (SGA) outcomes. These findings suggest that abnormal biochemical markers may serve as non-invasive indicators of underlying placental genomic anomalies, thus supporting their role in early obstetric risk stratification.

Maternal malnutrition and exposure to harmful substances early in pregnancy contribute to placental abnormalities, reducing villous surface area and limiting fetal nutrient supply [18]. Rasmussen et al. demonstrated that smoking cessation during pregnancy could reduce IUGR incidence by 12%. Additionally, severe preeclampsia has been linked to lower birth weights compared to mild cases, further emphasizing the impact of maternal health on IUGR [19]. Histopathological studies of IUGR placentas have documented extensive vascular abnormalities, including fibrin deposition, infarctions, and impaired villous development, highlighting the severity of placental dysfunction and the importance of intensive monitoring in pregnancies with abnormal biochemical markers [20, 23, 24].

These results are consistent with previous studies that have documented a relationship between low PAPP-A and β-hCG levels and fetal growth impairments [11]. Kaijomaa et al. investigated the predictive value of extremely low first-trimester PAPP-A levels (< 0.3 MoM) for adverse pregnancy outcomes, concluding that PAPP-A levels < 0.1 MoM increased the risk of complications 3.8-fold compared to women with PAPP-A levels between 0.2 and 0.3 MoM [25]. Similarly, Górczewski et al. (2023) reviewed the role of first-trimester biochemical markers in predicting perinatal outcomes, showing that low PAPP-A levels were associated with hypertensive disorders, fetal growth restriction, and small-for-gestational-age (SGA) neonates [24].

Although our findings support the use of PAPP-A and β-hCG for early risk stratification of IUGR, it is important to highlight that current international clinical guidelines do not incorporate these biomarkers into formal screening protocols for intrauterine growth restriction. The 2019 FIGO recommendations suggest that PAPP-A may be included in preeclampsia risk models when already available from aneuploidy screening, but do not propose its use for IUGR prediction [26]. Similarly, the 2024 update of the Royal College of Obstetricians and Gynaecologists' Green-top Guideline No. 31 provides detailed recommendations for IUGR monitoring based on Doppler assessments and clinical factors, yet does not include first-trimester biochemical markers in its risk pathways [27]. Moreover, the ISUOG guidelines for first-trimester ultrasound performance do not address the use of PAPP-A or β-hCG in screening for fetal growth restriction [28, 29]. This absence underscores a significant translational gap that our study seeks to address through a cost-effective, real-world model based on existing screening infrastructure.

Strengths, limitations and implications

A key strength of this study lies in its real-world clinical applicability. Conducted in a tertiary referral center with a large, non-selected cohort, the study reflects standard prenatal care practices and population diversity in a real-life setting. The retrospective design enabled access to complete clinical and laboratory data, and the inclusion of routine first-trimester markers (PAPP-A and β-hCG) reflects existing clinical workflows. Adjustment for key confounding factors—maternal age, BMI, smoking status, and use of assisted reproductive technologies—enhances the robustness of the observed associations.

Our findings contribute to an evolving paradigm in prenatal care, highlighting the potential of first-trimester biochemical markers not only for aneuploidy screening

but also for early risk stratification of placental dysfunction and fetal growth restriction. Given that these markers are already part of standard screening protocols, their expanded use as predictors of IUGR is both feasible and cost-effective. Integrating them into broader risk models could enable more personalized surveillance and earlier interventions for high-risk pregnancies.

Moreover, the use of a case–cohort design represents a methodological strength of this study. This hybrid approach allowed for efficient risk estimation by including all IUGR cases and a randomly selected reference subcohort from the same population. It preserved internal validity while optimizing resource use and minimizing selection bias, as supported by methodological literature [10, 11].

Nonetheless, certain limitations should be acknowledged. The single-center design may limit external validity, and although the sample size was adequate for PAPP-A analysis, the statistical power to detect consistent effects for β-hCG—particularly at extreme percentiles—was more limited. In addition, potential residual confounding from unmeasured variables such as maternal nutrition, inflammatory status, or undiagnosed placental pathology cannot be entirely ruled out.

Future research should focus on validating these findings in multicenter, prospective cohorts, and assessing the clinical utility of incorporating PAPP-A and β-hCG into integrated screening algorithms that combine biochemical, biophysical, and maternal risk factors.

Conclusion

This study provides strong evidence that low first-trimester PAPP-A levels are independently associated with an increased risk of intrauterine growth restriction, reinforcing their value as early biomarkers of placental dysfunction. Although the association with β-hCG was less consistent, it remains clinically relevant and may complement PAPP-A in early risk stratification. Together, these markers could enhance current screening strategies by identifying at-risk pregnancies earlier in gestation, enabling tailored follow-up and timely interventions to improve perinatal outcomes.

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Author contributions
ACM, IGA and TDS have contributed to the conception and design of the study. ACM, IGA, CCGL, JAM, SVC, JMOF, MCM and TDS have acquired the data and have been involved in drafting the manuscript. The first draft of the manuscript was written by ACM, IGA and TDS. All authors reviewed and provided input on the final version of the manuscript.

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Data availability
Data cannot be made publicly available in order to protect infant privacy. The data are available upon request from the University of Cantabria Archive (http://repositorio.unican.es/) for researchers who meet the criteria for access to confidential data. Requests may be sent to Dr. Inés Gómez-Acebo (ines.gomez@unican.es).

Declarations

Ethics approval and consent to participate
This project was approved by the Ethics Committee for Clinical Research of Cantabria (Comité de Ética de la Investigación con Medicamentos de Cantabria, CEIm) on 01/12/2022, reference number 2022.237. The study was conducted as an observational study, collecting data from medical records without any direct intervention on participants, and therefore, no risks were associated with participation. In accordance with Royal Decree 957/2020, which regulates observational studies in Spain, a waiver of informed consent was requested and granted by the Ethics Committee. The processing, communication, and transfer of personal data complied with all applicable regulations, including the General Data Protection Regulation (GDPR, Regulation (EU) 2016/679 of the European Parliament and of the Council, April 27, 2016) and the Spanish Organic Law 3/2018, of December 5, on Personal Data Protection and Guarantee of Digital Rights. The study adhered to Spanish laws on biomedical research, European Union data protection regulations, and the Declaration of Helsinki on ethical principles for medical research involving human subjects.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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References

1. Rodríguez-Zurita A, Caamiña Álvarez S, González Gómez T, Gónzalez García M, Martín Santos L, García Bello MÁ, et al. Niveles séricos de PAPP-A y β-hCG En El primer trimestre Del Embarazo Como predictores de resultados obstétricos desfavorables En El hospital universitario Nuestra señora de Candelaria. Clínica E Investig En Ginecol Obstet; 2021. p. 49.
2. Sun X-W, Li X-H, Zhang C, Meng F-Q, Xing Y-G, Ding Y. Correlation analysis of serum placental growth factor, pregnancy-related plasma protein-A and disease severity in patients with hypertensive disorder in pregnancy. Eur Rev Med Pharmacol Sci. 2021;25:1788–95.
3. Pakniat H, Bahman A, Ansari I. The relationship of pregnancy-Associated plasma protein A and human chorionic gonadotropin with adverse pregnancy outcomes: A prospective study. J Obstet Gynaecol India. 2019;69:412–9.
4. Gupta S, Goyal M, Verma D, Sharma A, Bharadwaj N, Kabra M, et al. Adverse pregnancy outcome in patients with low pregnancy-associated plasma protein-A: the Indian experience. J Obstet Gynaecol Res. 2015;41:1003–8.
5. Turner JM, Kumar S. Low first trimester Pregnancy-Associated plasma Protein-A levels are not associated with an increased risk of intrapartum fetal

compromise or adverse neonatal outcomes: A retrospective cohort study. J Clin Med. 2020;9:1108.
6. Shah KH, Anjum A, Nair P, Bhat P, Bhat RG, Bhat S. Pregnancy associated plasma protein A: an indicator of adverse obstetric outcomes in a South India population. Turk J Obstet Gynecol. 2020;17:40–5.
7. Arenas Ramírez JJ, Puerto Navarro B, Molina García FS. Martínez-Astorquiza Ortiz de Zarate T. Detección y Manejo de Las alteraciones Del Crecimiento fetal En españa. Encuesta Nacional. Prog Obstet Ginecol Rev Soc Esp Ginecol Obstet. 2017;60:306–13.
8. Roura LC i, Rodríguez EC, Arenas JMB, Alsina JC, Ramoneda VC, Armengol ED et al. Tratado de ginecología y obstetricia. 2014.
9. Calculadora de riesgo gestacional - Vall d'Hebron Barcelona - Medfetal. Medicina Fetal Vall d'Hebron. 2025. https://www.medfetal.org/riesgo-gestacional/
10. Rothman KJ, Lash TL, Greenland S. Modern epidemiology /. In: Kenneth J, editor. Rothman; Sander greenland; timothy L. Lash. 3rd. ed. Philadelphia: Lippincott Williams & Wilkins; 2008.
11. Onland-Moret NC, van der Schouw ADL, Buschers YT, Elias W, van Gils SG. Analysis of case-cohort data: a comparison of different methods. J Clin Epidemiol. 2007;60:350–5.
12. World Medical Association. World medical association declaration of helsinki: ethical principles for medical research involving human participants. JAMA. 2025;333:71.
13. Ley. 14/2007, de 3 de julio, de investigación biomédica. Bol Estado. 2007;159:28826–48.
14. Papamichail M, Fasoulakis Z, Daskalakis G, Theodora M, Rodolakis A, Antsaklis P. Importance of low pregnancy associated plasma Protein-A (PAPP-A) levels during the first trimester as a predicting factor for adverse pregnancy outcomes: A prospective cohort study of 2636 pregnant women. Cureus. 2022;14:e31256.
15. Dugoff L, Hobbins JC, Malone FD, Porter TF, Luthy D, Comstock CH, et al. First-trimester maternal serum PAPP-A and free-beta subunit human chorionic gonadotropin concentrations and nuchal translucency are associated with obstetric complications: A population-based screening study (The FASTER Trial). Am J Obstet Gynecol. 2004;191:1446–51.
16. Chen Y, Dai X, Hu T, Jiang C, Pan Y. Clinical value of prenatal screening markers in early pregnancy combined with perinatal characteristics to predict fetal growth restriction. Transl Pediatr. 2024;13:1071–85.
17. Honarjoo M, Zarean E, Tarrahi MJ, Kohan S. Role of pregnancy-associated plasma protein A (PAPP-A) and human-derived chorionic gonadotrophic hormone (free β-hCG) serum levels as a marker in predicting of small for gestational age (SGA): A cohort study. J Res Med Sci Off J Isfahan Univ Med Sci. 2021;26:104.
18. Carrascosa A. Crecimiento intrauterino: factores reguladores. Retraso De Crecimiento Intrauterino. 2003;:55–73.
19. Rasmussen S, Irgens LM. The effects of smoking and hypertensive disorders on fetal growth. BMC Pregnancy Childbirth. 2006;6:16.
20. Burton GJ, Jauniaux E. Pathophysiology of placental-derived fetal growth restriction. Am J Obstet Gynecol. 2018;218:S745–61.
21. Cavoretto PI, Farina A, Salmeri N, Syngelaki A, Tan MY, Nicolaides KH. First trimester risk of preeclampsia and rate of spontaneous birth in patients without preeclampsia. Am J Obstet Gynecol. 2024;231:e4521–7.
22. Spinillo SL, Farina A, Sotiriadis A, Pozzoni M, Giglio S, Papale M, et al. Pregnancy outcome of confined placental mosaicism: meta-analysis of cohort studies. Am J Obstet Gynecol. 2022;227:714–e7271.
23. Dankó I, Kelemen E, Tankó A, Cserni G. Placental pathology and its associations with clinical signs in different subtypes of fetal growth restriction. Pediatr Dev Pathol Off J Soc Pediatr Pathol Paediatr Pathol Soc. 2023;26:437–46.
24. Górczewski W, Górecka J, Massalska-Wolska M, Staśkiewicz M, Borowski D, Huras H, et al. Role of first trimester screening biochemical markers to predict hypertensive pregnancy disorders and SGA Neonates-A narrative review. Healthc Basel Switz. 2023;11:2454.
25. Kaijomaa M, Ulander V-M, Hämäläinen E, Alfthan H, Markkanen H, Heinonen S, et al. The risk of adverse pregnancy outcome among pregnancies with extremely low maternal PAPP-A. Prenat Diagn. 2016;36:1115–20.
26. FIGO Working Group on Good Clinical Practice in Maternal-Fetal Medicine. Good clinical practice advice: first trimester screening and prevention of pre-eclampsia in Singleton pregnancy. Int J Gynaecol Obstet Off Organ Int Fed Gynaecol Obstet. 2019;144:325–9.
27. Morris RK, Johnstone E, Lees C, Morton V, Smith G. The Royal college of obstetricians and gynaecologists. Investigation and care of a Small-for-Gestational-Age fetus and a growth restricted fetus (Green-top guideline 31). BJOG Int J Obstet Gynaecol. 2024;131:e31–80.
28. Salomon LJ, Alfirevic Z, Bilardo CM, Chalouhi GE, Ghi T, Kagan KO, et al. ISUOG practice guidelines: performance of first-trimester fetal ultrasound scan. Ultrasound Obstet Gynecol Off J Int Soc Ultrasound Obstet Gynecol. 2013;41:102–13.
29. Salomon LJ, Alfirevic Z, Da Silva Costa F, Deter RL, Figueras F, Ghi T, et al. ISUOG practice guidelines: ultrasound assessment of fetal biometry and growth. Ultrasound Obstet Gynecol. 2019;53:715–23.

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