

ORIGINAL RESEARCH

A Prospective Study of Estimation of Amniotic Fluid Optical Density for the Prediction of Fetal Maturity and Outcome

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ABSTRACT

Background: Objectives: 1. To establish correlation between Amniotic fluid Optical density (AFOD), gestational age, birth weight and functional maturity of the newborn. 2. To obtain mean AFOD at spontaneous onset of labor. 3. To study the functional maturity of the newborns, especially in terms of lung maturity by means of presence or absence of RDS in babies born out of spontaneous labor and in those born by elective termination.

Materials and Methods: From November 2019 to November 2021, 80 pregnant women attending Katuri Medical College in Guntur were studied descriptively. We chose 80 singleton pregnant women in spontaneous labour who had a first trimester ultrasound and estimated crown rump length. AF samples were taken aseptically during amniotomy and caesarean section after meticulous hysterotomy from the bulging membranes. Amniotic fluid optical density measured at 650 nm. All the babies' birth weights were noted. NICU admission, respiratory distress, etc.

Results: we see few babies born even at 35+ wks are fully mature and do not develop RDS. But on the other hand few born even after 40 wks are functionally premature and develop RDS. Some AFODs match the newborn's functional maturity whereas others don't. AFOD can assess a fetus's functional maturity. At AFOD 1, newborns are completely functioning. Babies with AFOD 0.4 are immature and have RDS regardless of gestational age, while those with AFOD 1+ are fully mature. Those with AFOD 1.75+ had postnatal alterations.

Conclusion: Each foetus has its own maturity potential, which occurs at various gestational ages and birth weights. AFOD 1.05820.36 babies had functional maturity, brown skin with little vernix, and no respiratory distress. Amniotic fluid OD.

Keywords: AFOD, Birth Weight, RDS, Gestational ages, Premature.

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INTRODUCTION

Despite major scientific progress, the mechanism that determines the gestational age at which spontaneous commencement of labour occurs with each pregnancy, as well as the gestational age at which functional maturity is achieved with each foetus, has remained a mystery to obstetricians. Different foetuses reach completion of functional maturity at varied gestational ages and with different birth weights, and this can occur at any time between 36 weeks and 42+ weeks of pregnancy. An essential preventive step in obstetrics is the study of each

patient's unique situation to determine the most acceptable timing for termination of a pregnancy.^[1]

The reason for inducing labour prematurely nowadays is that the calendar time of pregnancy duration has exceeded 287 or 294 days from the date of the last menstrual period, as determined by the mother, at the time of conception.^[2,3] Scientist have no reliable means of estimating the exact date (of confinement) but are forced to content ourselves with the method proposed by Naegele, which is based on the belief that labour occurs two hundred and eighty days from the beginning of the last menstrual period," J. Whitworth Williams wrote in the first edition of what would become the classic text Williams Obstetrics.^[4] It was not until 1948 that the World Health Assembly suggested an official definition of a preterm infant, which defined one as having a birth weight of less than 2,500g, a gestational age of less than 38 full weeks, or both as being early.^[5] One year later, in 1949, the Standard Certificate of Live Birth developed by the United States National Center for Health Statistics of the Centers for Disease Control and Prevention was revised to include reporting of the length of pregnancy in weeks, and it was revised again in 1956 to specify reporting of "completed weeks" of gestation, which was the first time this was done.^[6]

NICU admissions, transient tachypnea of the newborn (TTN), respiratory distress syndrome (RDS), need for ventilator support, increased sepsis rates (suspected or proven), and higher incidence of newborn feeding problems and other transition issues are all associated with non- medically indicated (elective) deliveries between 37 and 39 weeks, according to the ACOG committee. Having a Caesarean section performed before 39 weeks of pregnancy increases the risk of infant morbidity, which includes respiratory distress, hypoglycemia, sepsis, NICU admissions, and hospitalisation for more than 5 days, among other things.^[7]

In the case of "late to care" (after 20 weeks), where dates can only be calculated by ultrasound after 20 weeks, ACOG recommends FLM before a scheduled elective operation is scheduled. An open conversation about the risks and advantages of repeat caesarean section in an uncomplicated pregnancy would be necessary.^[7,8]

The following description of the condition of amniotic fluid may be found in an obstetrics textbook from 1862: "At first, it is clear, but later on it becomes murky and flocculent." The same textbook also includes a description of the changing amount and distribution of vernix on the newborn, which is very interesting.^[9]

Objectives

1. To establish correlation between Amniotic fluid Optical density (AFOD), gestational age, birth weight and functional maturity of the newborn.
2. To obtain mean AFOD at spontaneous onset of labor.
3. To study the functional maturity of the newborns, especially in terms of lung maturity by means of presence or absence of RDS in babies born out of spontaneous labor and in those born by elective termination.

MATERIALS & METHODS

A descriptive study was conducted on one 80 pregnant women who attended Katuri Medical College, Guntur, during a period of 2 years from November 2019 to November 2021, with the following

Inclusion criteria:

- Women who underwent first trimester scan and crown rump length estimation, or
- Women with regular periods who underwent scan at less than 20 weeks gestation which is in agreement with the gestational age calculated from the last menstrual period

Exclusion criteria:

- Blood stained and meconium stained amniotic fluid samples
- Intrauterine growth restriction
- Premature rupture of membranes
- Preterm premature rupture of membranes
- Amniotic fluid index <5 and >25

Informed consent was taken from all women prior to artificial rupture of membranes and before LSCS

Under aseptic precautions amniotic fluid samples were collected while doing amniotomy after 3- 4cm dilatation by an intramuscular needle fitted with a 2ml disposable syringe. This procedure is done under vision gently by inserting one Sim's speculum, if necessary two, taking care to avoid injury to the presenting part.

Amniotic fluid samples are collected at cesarean section after careful incision on the uterus from the bulging membranes.

The color and turbidity of fresh uncentrifuged amniotic fluid samples thus obtained was measured subjectively by naked eye inspection and quantified subjectively as follows:

Table 1: Scoring for amniotic fluid turbidity

Color/ Turbidity of amniotic fluid	Score
Watery	1
Milky	2
Buttermilk like	3
Curd like	4

The color and turbidity of the fresh uncentrifuged sample was objectively quantified by colorimetry. The measurement of AFOD was done at 650 nm after the reading of control test tube with tap water. Babies are observed for the amount of Vernix on their skin immediately after birth before drying of the baby. Routine protocol for neonatal resuscitation was followed. Birth weights were recorded for all babies. APGAR scores at 1minute and 5 minutes were obtained. Babies were observed for classical signs of respiratory distress (tachypnea >60breaths /min, grunting, retraction of ribs, sternum. Suspected cases of distress were resuscitated with bag-mask ventilation with oxygen and referred to NICU for further management. Respiratory distress was graded using Downe's score. (see above)

RESULTS

Analysis was done using Microsoft excel 2007.

Table 2: percentage of deliveries at different gestational ages

Gestational age (days)	Percentage of deliveries	Number of deliveries
258 (36 weeks)	09	09
259-279 (37- 39 weeks)	48	48
280 (40 weeks)	1	1
GE 281	22	22
Total	80	80

In this observational study comprising 80 pregnant women, 65 were spontaneous deliveries. Out of these, 1 woman delivered on the day of EDD which corresponds to 1.25%, while 73.25% of the deliveries took place at gestational age between 37 and 40 weeks.16.25% delivered spontaneously before 37 weeks and 22.75% delivered after 40 weeks.

Out of a total 80 women, 42 were primigravidae, 43 were second gravidae, 13 were third and 2 were fourth gravidae.

Table 3: Distribution of cases according to parity

Parity	Frequency	Percent
G1	42	42
G2	43	43
G3	13	13
G4	2	2
Total	80	80

The mean AFOD was found to be 1,10 in the primigravidae and 1.02 in the multi gravidae in the non RDS group, while it was 0.29 in the primigravidae and 0.30 in the multi gravidae in the RDS group.

No statistically significant difference was found in the mean AFOD values between primi and multi gravidae either in the non RDS group or in the RDS group.

Table 4: Characteristics of AFOD and birth weight in RDS and non RDS compared to total population

	Without RDS	With RDS	Total
No. of cases	72	8	80
Mean period of gestation	271.52±9.38	255.5±13.65	272.08±10.71
AFODmean±SD	1.0582±0.36	0.2937±0.03	0.9969±0.37
Mean birth Weight± SD	3.1468±0.35	2.3875±0.35	3.086±0.45
AFOD<0.40	0	8	8
Birth weight/gestational Age± SD	11.5±1.27	9.31±0.95	11.32±1.37

Table 5: Distribution of cases with and without RDS in different groups of spontaneous onset and elective termination.

	Spontaneous Onset of Labour						Elective Termination					
	Without RDS		With RDS		Total		Without RDS		With RDS		Total	
<246	0	0.0%	1	100.0%	1	100%	0	0.0%	0	0.0%	0	0%
246-252	4	66.7%	2	33.3%	6	100%	0	0.0%	1	100.0%	1	100%
253-259	3	60.0%	2	40.0%	5	100%	2	100.0%	0	0.0%	2	100%
260-266	8	100.0%	0	0.0%	9	100%	2	100.0%	0	0.0%	2	100%
267-273	20	100.0%	0	0.0%	20	100%	5	100.0%	0	0.0%	5	100%
274-280	8	88.9%	1	11.1%	9	100%	10	91.7%	1	8.3%	08	100%
>280	19	100.0%	0	0.0%	19	100%	5	100.0%	0	0.0%	5	100%

The mean period of gestation in days for the total number of cases was 272.08±10.8;

The mean AFOD for Non-RDS cases (n=62) 1.057±0.37 was found to be significantly higher when compared to the RDS cases (18) 0.293±0.03; p<0.001.

The total number of cases with AFOD <0.40 were 8 and all of them developed RDS.

The mean birth weight in total number of cases was 3087±430, while it was 3147±38 and 2387±351 respectively in the non RDS and the RDS groups.

The mean birth weight (grams) /gestational age(in days) ratio for total cases was 11.32 ± 1.34 . The birth weight/ gestational age for non RDS group was the mean being 11.5 ± 1.23 , and the median 10.467. For the RDS group, it ranged from, the mean was 9.31 ± 0.95 and the median was 9.479. There was no statistically significant difference in birth weights adjusted to gestational age between the non RDS and RDS groups ($p=0.0827$).

The 80 cases were divided into groups according to gestational age, for comparison; less than 35 weeks, 35weeks+1day to 36 weeks, 36weeks+1day to 37 weeks, 37weeks+1day to 38 weeks, 38weeks+1day to 39 weeks, 39 weeks +1 day to 40weeks, and greater than 40 weeks. The chart showing the number of cases in each group divided according to the gestational age is as follows:

Out of the total number of cases of spontaneous onset of labor group of 71 only 6 case developed RDS which was in the lowest gestational age group, while in the elective termination group, RDS was present in 3 out of 7 groups, including the group with period of gestation >280 days. But, the severity of RDS decreased with increasing gestational age (mild RDS with Downe's score <4 , severe RDS with >4 Downe's score).

Table 6: summary of characteristics of AFOD and birth weight in different gestational ages

	Gestational Age in Days							Total
	<246	246-252	253-259	260-266	267-273	274-280	>280	
NoofCase	1(1.25%)	5(6.25%)	5(6.25%)	9(11.25%)	22(27.5%)	18(22.5%)	20(25%)	80(100%)
Birth Weight Mean	2.1 ± 0	2.54 ± 0.22	2.671 ± 0.39	3.23 ± 3.12	3.14 ± 0.28	3.214 ± 0.30	3.332 ± 0.36	3.077 ± 0.40
Cases without RDS	0	4	5	9	22	18	24000000	78
Cases with RDS	1	3	2	0	0	2	0	8
Mean AFOD at without RDS	-	0.64 ± 0.03	0.84 ± 0.17	0.83 ± 0.11	1.04 ± 0.21	0.91 ± 0.20	1.34 ± 0.43	1.15 ± 0.36
Mean AFOD at with RDS	0.27 ± 0	0.31 ± 0.02	0.26 ± 0.02	-	-	0.34 ± 0.01	-	0.27 ± 0.03
BW/GA	8.812 ± 0.0	9.432 ± 0.86	10.13 ± 1.34	11.54 ± 0.98	11.53 ± 1.09	11.59 ± 1.13	11.74 ± 1.32	11.31 ± 1.32

Table 7: Correlation between AFOD and gestational age of the newborn

	Period of Gestation	AFOD(at 650nm)
Period of Gestation	1	
AFOD(at 650nm)	0.509	1

The correlation coefficient was 0.509 between period of gestation and AFOD showing that there is no correlation between them

Table 8: Correlation between functional maturity and gestational age of the new-born.

	Functional maturity	Gestational age
FunctionalMaturity	1	
Gestational age	-0.392	1

The correlation coefficient between functional maturity and gestational age was - 0.392 which shows that there is no significant correlation between them.

Table 9: Correlation between AFOD and Vernix score

	AFOD at 650 nm	Vernix Score
AFOD at 650 nm	1	
Vernix score	-0.514	1

The correlation coefficient between AFOD and vernix was -0.514 which shows a negative correlationthe correlation coefficient between AFOD and vernix was -0.512 which shows that there is no significant correlation between the two.

Table 10: Correlation between AFOD and functional maturity

	AFOD at 650 nm	Functional maturity
AFOD at 650 nm	1	
Functional maturity	0.837	1

The correlation coefficient between amniotic fluid optical density at 650nm and functional maturity was 0.837, showing a significant positive correlation.

Table 11: Correlation between Weight and functional maturity was 0.837 which shows that there is significant positive correlation between the two

	Functional maturity	Birth Weight
Functional Maturity	1	
Birth weight	-0.478	1

The correlation coefficient between functional maturity and birth weight of the newborn was found to be -0.478, showing no significant correlation between these two factors.

Table 12: Correlation between AFOD and birth weight of the newborn was -0.477 which shows that there is no significant correlation between the two

	AFODat 650nm	Birth weight
AFOD at 650nm	1	
Birth weight	0.477	1

The correlation coefficient between AFOD and birth weight was 0.477 which shows minimal correlation between the two.

DISCUSSION

Each baby has its unique we cannot define term in terms of gestational age A genetic predisposition to birth preterm is present in some women as a result of variances at the molecular level. Cervical ripening, which occurs in the absence of infection, is an inflammatory process similar to that of term labour. Polymorphisms in various genes that regulate cytokines, hereditary sensitivity to infections with low virulence, mutations in

collagen synthesis, oxytocin receptors, BMI, parity, and age are all factors that contribute to this condition. In addition, these parameters differ from one race or fetomaternal unit to another, resulting in physiological variance in the length of pregnancy.^[4,5]

When instances of RDS were eliminated from analysis in the current study, there was no significant difference in mean AFOD values between the two groups in the range of 3 to 7. The results remained the same after controlling for birth weights. All of the kids born with an AFOD value in the range of 1.0370.103 SD were fully functionally developed at the time of birth. Their skin was mature and pink, and there were very few vermicides attached to the surface of their bodies. When the AFOD level was more than 0.40, none of the newborns had RDS. In contrast, kids delivered with an AFOD score less than 0.40 (n=18) were functionally preterm, suffered varied degrees of RDS, and had birth weights ranging from 2300 to 3000 grammes, depending on the gender. Because of this, their skin was prematurely thin, glossy, and bright red in colour, with a thick layer of vermicides adhering to the surface. The lowest AFOD value at which newborns got RDS was 0.20, while the highest was 0.40. The post-mature skin alterations in babies born with significantly higher AFOD levels (>1.75) were more severe [Figure 4.3,4.4,4.5]. Prior to centrifugation, optical density measurements were made using centrifuged materials. It was established that the cutoff value was 0.15. The figures are subject to change depending on the centrifugation speed and length of time. The mean AFOD at spontaneous commencement of labour (1.08) was higher than the mean (0.80) for individuals who chose to terminate their pregnancy, indicating that the mother was fully mature at the onset of labour rather than at the end of the pregnancy. Centrifugation at 2000 rpm for 10 minutes produces an OD reading of 0.15 for the uncentrifuged samples with an OD of 0.40. The centrifugation results in an OD reading of 0.40 for the centrifuged samples. The researchers discovered that kids born with an AFOD value less than 0.40 at a younger gestational age suffered moderate to severe and protracted RDS, as opposed to babies delivered with an AFOD value less than 0.40 at a later gestational age, who developed milder RDS for a shorter period of time. The findings are consistent with those of the previous studies, which is optimistic.

The amount of surfactant is represented indirectly by the abbreviation AFOD. As a result, the severity of the disease is determined by how low the AFOD value is, or in other words, how low the surfactant phospholipids are, rather than how old the patient is chronologically.

Despite the fact that a 0.40 AFOD score is sufficient to prevent RDS, foetal maturity is considered complete only when all systems have reached full functional maturity. When the GI system is premature, necrotizing enterocolitis occurs, when the pulmonary system is premature, respiratory distress syndrome (RDS) occurs, and when the skin is premature, inability to regulate temperature occurs. When the AFOD value is around 1, it indicates that the infants have reached full maturity.

AF The level of lecithin in the blood increases from 43 micrograms per millilitre at 34-35 weeks gestation to 147 micrograms per millilitre at term before labour. Furthermore, it has been shown that its levels can rise to as high as 232 micrograms/ml during long-term labour. A significant increase in sebaceous gland hyperplasia, which is a main constituent of vernix caseosa and occurs in conjunction with the desquamation of foetal corneocytes during the last trimester, causes vernix caseosa. The separation of the vernix has made a significant contribution to the increase in AFOD during the third trimester. The lung develops in a parallel manner to the rest of the body throughout the same time span. Higher OD values were associated with a lower quantity of vernix on the body surface of newborns. In this study, the inverse relationship between AFOD values is demonstrated. the correlation coefficient is -0.512 (negative correlation coefficient).

Labor progresses as a result of the production of pro-labor cytokines such as interleukin-6, interleukin-8, interleukin-1 beta, and epidermal growth factor in amniotic fluid and maternal

serum. These cytokines are released by the foetal skin and accumulate in the amniotic fluid and maternal serum as labour progresses. It is more closely associated with foetal functional maturity than it is with either gestational age or birth weight when labour begins. Each fetomaternal unit has its own unique gestational age at the time of delivery. Klimek was the first to establish the concept of an individual word (Let man be born at his own due time). There are scoring systems that are independent of gestational age and birth weight, as well as computer-aided procedures, that can be used to determine the functional or biological gestational age of a foetus. Gestational age is often assessed postnatally using the Klimek and Ballard scoring methods, both of which are widely used. In India, the recommendations for the child survival and safe motherhood programmes recommend that newborns who do not show signs of disease can be handled at home with careful care, even if their birth weight is as low as 1800 grammes.





However, the significance of AFOD extends beyond simply determining lung maturity; it also has a significant impact on the identification of events that occur beyond lung maturity, such as skin maturity and the beginning of labour. The use of induced (non-spontaneous) labour has been demonstrated to result in higher Oxytocin requirements and an increased incidence of PPH. Even if the inductions were performed at an inopportune time, the iatrogenic consequences and suffering may have been prevented if the inductions had been performed according to AFOD guidelines. A single-age-based approach to inductions may result in the need for high doses of prostaglandins, dysfunctional labour, prolonged induction delivery intervals and an increased number of instrumental deliveries. It may also result in pain and higher doses of narcotic analgesia, neonatal respiratory depression and perineal tears as well as an increased rate of caesarean section. Mechanization of the natural process of labour has resulted in increased mother and newborn morbidity and mortality. AFOD readings or their corresponding indications, which define the preparation of labour and thus the biological gestational age, should be considered when inducing labour rather than the "EDC," which is a misnomer in and of itself.^[7,8]

The fetus's attainment of full foetal development occurs at the same time that the placenta's biological existence comes to an end and the woman is in the best possible internal state to give birth to her child. This modern understanding of the origin of the onset of labour is a subject of investigation for medical quantum thermodynamics, which serves as the foundation for the multifactoral physical, chemical, biological, and psycho-emotional conditionings of pregnancy that are present. As a result, it is critical to recognise both the concurrence and diversity of changes that characterise each individual birth in order to avoid rushing into delivery decisions without taking into consideration the unique foetal development of each individual.^[9]

In terms of AFOD, each foetus has a certain level of maturity potential. The result is that every fetomaternal unit is distinct and requires a customised strategy. In our study population, we infer that complete functional maturity and the commencement of labour occur at individual terms with mean AFOD about 1, independent of birth weight and gestational age, which is the point of complete maturation in our model.

Comparison of Studies:**The following studies show similar results to this study:**

According to a previous study by Matile et al., it has been found that the delivery of a mature newborn begins spontaneously in the vast majority of cases when milky amniotic fluid is detected by amnioscopy.^[8]

Many of Nishijima's investigations, including his most recent, demonstrate conclusively that each fetomaternal unit matures at its rate, and that this rate is independent of any single characteristic considered to date.

Using data from an article by Cetrulo et al,^[9] it is extrapolated that complete functional maturity and the commencement of labour occur at individual terms, with a mean AFOD of 0.98, regardless of birth weight or gestational age, which is the foundation for this research.

CONCLUSION

According to the findings of this study, each fetus has its own maturity potential. Furthermore, the optical density of amniotic fluid (AFOD) was found to be substantially associated with the functional maturity of newborns. It has been discovered that the risk of AFOD increases with pregnancy age. The prevalence of AFOD is higher among neonates who have a higher APGAR score.

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