# THE CYTOLOGICAL STUDY OF AMNIOTIC FLUID IN FOETAL MATURITY

by

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The exact foetal age is important in selecting the optimal time for induction of labour or in planning an elective caesarean section in cases of P.E.T., post-maturity, diabetes and RH incompatibility. It is of no less importance in placental insufficiency and in cases of stillbirths in previous pregnancies. This precaution of confirming the accurate foetal age will not only prevent stillbirths and intra-uterine deaths, but also avoid an iatrogenic death due to prematurity resulting from premature induction.

In our country, it is common for our women not to remember their last menstrual period, and some conceive during the lactational period.

The cytological study of amniotic fluid with Nile blue sulfate was carried out and its value in assessing foetal age was determined.

The cellular content of amniotic fluid consists of amniotic eithelial cells, squamous cells from the foetal skin and the fat ladden cells of foetal sebaceous glands. The amniotic epithelial cells are present in the early weeks, but disappear in the later half of pregnancy. Under 34 weeks, the average fat cell count is less than 1%. Between 34-38 weeks, it is 1-10% and between 38-40 weeks, the count is anything between 10-50% (Brosens and Gordon 1966).

The nile blue sulfate which stains the fat cells orange is a very rapid, simple and inexpensive technique that can be employed in clinical practice without much relying on the radiological investigation.

*Material*

One hundred and thirty cases were initially selected for the present study. The samples were obtained from patients attending the antenatal clinic, in labour, and during caesarean section.

Amniocentesis failed in 7 cases. In 5 cases, blood was aspirated and 10 samples were meconium stained, and these cases were therefore discarded from the study. Four patients had repeat amniocentesis at different gestational period. The technique of Brosens and Gordon (1966) was used.

The cases were divided into 3 groups:—

GROUP I:— Thirty-three samples were obtained from 32 normal pregnant women with known gestational period.

Six samples were obtained at 20-24 weeks pregnancy. None of the samples showed any particle of vernix and no orange cell could be seen when stained

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with .1% acqueous solution of Nile blue sulfate.

One case studied at 34 weeks showed 4% orange cells, and the repeat study at 39 weeks revealed 25% of these fat ladden cells. Twenty-five cases were between 37-40 weeks. Taking 20% count as indicating at least 38 weeks maturity, the correct result was obtained in 88% cases (22 samples). One patient had 15% count and another 5% at 40 weeks and third specimen showed only 1% count at 39 weeks. The naked eye appearance of the amniotic fluid however showed vernix caseosa and free fat globules were seen microscopically. There was no positive correlation between the cell count and the foetal weight. Four babies weighed less than 2500 gms but the cell count was over 20% in each specimen. This cytological test was found useful in identifying premature from "small for date" babies.

GROUP II:— Forty-seven cases had various obstetric complications as shown below :—

| | | |
|---|---|---|
| Pre-eclamptic toxaemia | — | 10 cases |
| Postmaturity | — | 21 cases |
| Diabetes | — | 6 cases |
| Bad Obst. history | — | 5 cases |
| Hydramnois | — | 5 cases |
| TOTAL | — | 47 cases |

*P.E.T. Cases*: There was I false negative finding amongst 10 cases. This patient had BP of 160/100 at 38 weeks gestation. The fat cell count was less than 1%. The naked eye appearance also failed to show any vernix. The X-ray however confirmed the foetal maturity of over 36 weeks. The patient had a caesarean section for foetal distress at 39 weeks and the baby weighed 3.0 kg.

*Postmaturity*: Two patients claimed to be postmature by 10 days. The cell count was, on the other hand, only 10% and 4% respectively. The X-ray too revealed maturity of 36 weeks in each case. The rest of the cases had cell count over 20% and were successfully induced.

The correct prediction of foetal age was done in all diabetic and bad obstetric cases.

Of the 3 cases of hydramnios, only I case at 40 weeks showed 50% cell count. The other 2 had a count of 4% and 2% respectively, but vernix was seen in the amniotic fluid and both these cases delivered mature babies within a week of amniocentesis.

The overall false negative reading was 2.1% (one out of 47). There was no positive correlation between the cell count and the foetal weight.

GROUP III: Twenty-nine cases of unknown gestational period were selected for the study. The pregnancies were otherwise normal.

There were 2 false negative findings A 3rd gravida with breech presentation showed a count of 2% with absence of vernix at about 36 weeks gestation.

The X-ray however confirmed the maturity over 36 weeks. Similarly, another patient near term showed the cell count of 2% and there was no vernix caseosa. The X-ray showed the foetal maturity of 40 weeks.

Three patients in this group had repeat amniocentesis. A low count at 34 weeks showed a rise to more than 20% at about 38 weeks. The babies too were mature and of normal weight.

The false negative finding was thus seen in 6.9% in this group (2 cases).

In the entire series, 3 cases showed low cell count (2.8% false negative). There was no false positive reading.

*Discussion*

Our observation tallies with that of Brosens and Gordon (1966) who could not find any orange cell under 28 weeks of gestation. Chan, *et al* (1969), and Barnett and Navin (1970) all reported the similar observation. Sharma and Trussell in 1970 detected orange fat ladden cells only after 32 weeks of gestation, whereas Balkrishna (1972) found orange cells only after 33 weeks.

All the authors found a gradual increase in number of fat ladden cells after 30 weeks upto term. Three cases studied in the present series showed a count of 4% at 34 weeks, but a repeat study at 38 weeks and over revealed more than 20% orange cells.

Of the 26 samples studied in normal pregnancies after 37 weeks, 23 samples (88.40%) showed more than 20% orange cells, which was taken as an index of 38 weeks maturity. All the 3 samples which showed the cell count of less than 20%, however, revealed the presence of vernix and clumps of free lipid could be seen under the microscope.

Andrews (1970) and Eite (1971) suggested that the count of 10% indicated foetal maturity of 36 weeks and 20% indicate 40 weeks maturity. Balkrishna *et al* (1972) also recommended 20% count as indicating at least 38 weeks maturity. Taking 20% count or presence of vernix is indicating at least 38 weeks maturity, we obtained 100% accurate results. Chandriok *et al* (1971) reported 12.1% false negative results, Dhar and Bazaz (1975) 12.3% and Sharma and Trussell 4.4.% false negative findings.

Wachtel *et al* (1969), Lind *et al* and Bishop and Carson (1970) suggested that in cases of equivocal findings, the study of foetal squames should form an additional guidance in assessing foetal maturity. In early week, these authors observed immature nucleated squames resembling parabasal cells of vaginal cytology. After 36 weeks, there was a rapid increase in anucleated mature squames and disappearance of nucleated squames.

Like most other authors, we did not find any correlation between the cell count and the foetal weight. Thus it is possible to differentiate premature from small for date babies.

Exceptional finding was reported by Bishop and Corsen (1968) and Gauther (1972) who could gauge the foetal weight in 85% of their cases by studying the orange cell count.

Group II—There was only one false negative result in pre-eclamptic toxaemia group. (accurate prediction rate of 90%). Except Grizic, (1976) all other authors as well as our findings confirm that cell count is only related to foetal maturity and not to birth weight. The "Small for date" babies due to placental insufficiency in pre-eclampsia did not influence the cell count.

Similarly, the cell count was related only to gestational age in diabetics, and bad obstetric history. All cases of postmaturity showed a cell count of over 20% but the count was no different than that found at term. Our findings are thus similar to those of Brosens and Gordon (1966) and Bishop Corson (1968) and we are inclined to believe that postmaturity cannot be diagnosed or confirmed by fat cell count. On the other hand, Bal-Krishna *et al* (1972) and Grizic (1976) found a rise in the number of cell count after 40 wks.

In 2 out of 3 cases of hydramnios, a low count of 2 and 4% was obtained at term. This perhaps could be due to dilution by a large quantity of liquor. The presence

of vernix however confirmed the foetal maturity. A low count was also reported by Sharp in 1968 and Murphy in 1969. This is a warning that a low cell count in hydramnios may be deceptive in some cases and other methods should be employed in case of doubtful report. X-ray is ideal, because radiological investigation is essential anyway to exclude skeletal deformity.

Group III—There were 2 false negative readings in this group and naked eye examination too failed to reveal any vernix.

In the entire series, the accurate prediction rate for assessing foetal maturity was 97.2% (2.8% false negative—3 cases).

## *Conclusion*

The cytological study of amniotic fluid using .1% acquous solution of Nile blue sulfate is very useful in predicting the foetal maturity in normal and abnormal pregnancies. This method also helps in differentiating prematurity from small for date babies. In hydramnios however, one may have to employ some other method in assessing gestational age.

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