Acta Genet Med Gemellol 31:255–258 (1982) The Mendel Institute/Alan R. Liss, Inc.



The Lecithin/Sphingomyelin Ratio in Twin Pregnancies

R.J. Norman and S.M. Joubert

Department of Chemical Pathology, University of Natal, Congella, South Africa

The lecithin/sphingomyelin ratio showed no difference between the two amniotic sacs in 24 multiple pregnancies examined before the onset of labor. Once labor started, however, the L/S ratio was higher in the first sac compared to the second in the six pregnancies where labor had commenced and was somewhat advanced at the time of amniocentesis. Dual-sac amniocentesis is essential during labor when assessment of pulmonary maturity is required, whereas single-sac sampling is adequate before the start of labor.

Key words: Multiple pregnancy, Amniocentesis, Lecithin/Sphingomyelin ratio, Phospholipids, Lung maturity

INTRODUCTION

There is no agreement about the value of tests of pulmonary maturity in twin pregnancy. Two large series have shown concordancy between the first and second twin in respect of lecithin/sphingomyelin (L/S) ratios [9,10], but a few individual cases have been reported in which significant differences were demonstrated in the L/S ratio from the amniotic fluid or tracheal effluent in twin same pairs [3,7,8,11]. The continuing uncertainty resulting from these conflicting observations has important implications in pregnancies in which preterm labor and elective Caesarean section are more frequent than is the case with singleton gestation. This study reports on the L/S ratio in a large series of twin pregnancies, both before and after the onset of labor.

MATERIALS AND METHODS Patients

Thirty Zulu mothers with confirmed twin pregnancy were seen in the Twin Clinic at King Edward VIII Hospital, Durban, during the last four months of pregnancy. All patients had been scheduled for elective Caesarean section on the basis of previously determined criteria, including prior abdominal delivery, contracted pelvis, persistent malpresentation, and strong presumptive evidence for intrauterine growth retardation [4]. Twenty-four patients underwent Caesarean section before the onset of labor, while six mothers began their labors with membranes intact before presenting themselves to the hospital for surgery. Standard anesthesia was given [5] and amniotic fluid was obtained from the first sac before the uterus was opened. Amniocentesis from the second sac was performed under direct vision after delivery of the first twin. Intraamniotic dye was injected into the first sac in a few patients to validate the sampling procedure, but this procedure was discontinued due to the potential hazards to the fetus [2].

256 Norman and Joubert

Measurement of Lecithin/Sphingomyelin Ratio

Amniotic fluid was centrifuged at 600 g for five minutes and the supernatant extracted with chloroform-methanol (2:1). After acetone precipitation, chromatographic separation of the phospholipids was performed on silica gel plates [6], using a solvent system consisting of chloroform-methanol-glacial acetic acid and water in the ratio 50:25:7:3. The relative proportions of L and S were determined by the phosphorus content of the separated spots.

Assessment of Fetus and Placenta

Gestational age was determined by one or all of the following methods: early ultrasound, date of the last menstrual period, and external physical and neurologic scoring. The placentae were examined to exclude vascular anastamoses, and measurement of the umbilical cord blood packed cell volume was performed to exclude the twin-twin transfusion syndrome. No attempt was made to determine the zygosity of the twins by definitive techniques. Mean gestational age was 37.4 weeks (range 32–41) and mean parity 2.

Statistical Analysis

The paired T-test was used to compare twins within the same pregnancy and the unpaired T-test to compare differences between the pregnancies of different mothers.

RESULTS

Figure 1 shows the L/S ratios in twins I and II before and after the onset of labor. Before labor, there was no significant difference between twin I and twin II; subsequent to the

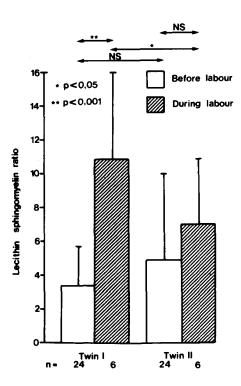


Fig. 1. Amniotic fluid lecithin sphingomyelin ratios before and after the onset of labor (Mean \pm SD).

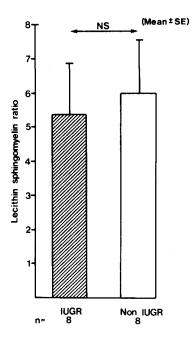


Fig. 2. Amniotic fluid lecithin sphingomyelin ratios in twin pregnancies with one growth-retarded (IUGR) and one non-growth retarded (NON-IUGR) fetus.

onset of uterine contractions, however, there was a highly significant rise in the L/S ratio in the amniotic fluid of the first twin whether compared to the group not in labor or to twin II in the same pregnancy. Figure 2 shows the eight patients with growth retardation (less than the 10th percentile for gestational age) before the start of labor. There was no difference in the L/S ratios of amniotic fluid from growth-retarded as opposed to non-growth-retarded fetuses.

DISCUSSION

Results reported in this study demonstrate that there is no difference in the L/S ratio of the amniotic fluid between the first and second twin in multiple pregnancy before the onset of labor and confirm the observations made by Spellacy et al [10]. These authors performed dual-sac amniocentesis in 14 women with twin pregnancies and reported a correlation coefficient of 0.86 for paired samples assessed for the L/S ratio. The present studies and the findings of Spellacy et al support the concept that factors such as the position in the uterus, fetal growth, weight, and sex do not significantly affect the L/S ratio of the late-gestation twin conceptus before labor begins. For ethical reasons sequential samples of amniotic fluid cannot be withdrawn from the same patient during pregnancy. Hence no conclusion can be drawn about differential rates of pulmonary maturation in the two fetuses; the findings relate to the situation that pertains near the end of pregnancy.

258 Norman and Joubert

Once labor begins, there is a significant increase in the amniotic fluid L/S ratio of the first twin. Increases in the amniotic fluid L/S ratios are generally interpreted as reflecting increased production and/or release of surfactant from the fetal lung. It may, however, be argued that the physical stress on the leading twin after labor commences exerts such an effect as to result in greater releases of lung phospholipid to amniotic fluid. If this hypothesis were correct, proportional rises in both L and S would be expected without a change in the L/S ratio. Hence it seems more likely that the increased L/S ratio observed is the result of increased de novo synthesis of surfactant, and there is good reason to believe that this phenomenon is related to increased glucocorticoid stimulation [1].

The practical implications of observations reported here are obvious. Single-sac amniocentesis of one fetus is sufficient to predict the pulmonary maturity of both fetuses before the onset of labor; on the other hand, once labor commences, a mature L/S ratio in the first sac is not a guarantee of pulmonary maturity in the second fetus.

Acknowledgments. This work was supported by a grant from the South African Medical Research Council to the Preclinical Diagnostic Research Group. We acknowledge the cooperation of the Department of Obstetrics and Gynecology.

REFERENCES

- 1. Ballard PL (1980): Hormonal influences during fetal lung development. In Ciba Foundation Symposium 78; "Metabolic Activities of the Lung." Amsterdam: Excerpta Medica, pp 251-274.
- Cowett RM, Hakanson DO, Kocon RW, Oh W (1976): Untoward neonatal effect of intra-amniotic administration of methylene blue. Obstet Gynecol (Suppl)48:74-76.
- Lemons JA, Jaffe RB (1973): Amniotic fluid lecithin/sphingomyelin ratio in the diagnosis of hyaline membrane disease. Am J Obstet Gynecol 115:233-237.
- Marivate M, Norman RJ (1982): Twins. In "Obstetric Problems in the Developing World." Clin Obstet Gynecol 9:723-744.
- 5. Norman RJ, Bredenkamp BLF, Joubert SM, Beetar C (1981): Fetal prostaglandin levels in twin pregnancies. Prostaglandins Med 6:309-316.
- Norman RJ, Joubert SM (1982): Koutine measurement of phosphatidyl glycerol in amniotic fluid. S Afr Med J 61:839–841.
- Obladen M, Gluck L (1977): RDS and tracheal phospholipid composition in twins: Independent of gestational age. J Pediatr 90:799-802.
- 8. Olson RW (1977): L/S ratio in twin pregnancy. Wisc Med J 76:116-117.
- 9. Sims CD, Cowan DB, Parkinson CE (1978): The lecithin/sphingomyelin (L/S) ratio in twin pregnancies. Br J Obstet Gynecol 83:447-451.
- Spellacy WN, Cruz AC, Buhi WC, Birk SA (1977): Amniotic fluid L/S ratio in twin gestation. Obstet Gynecol 50:68-70.
- 11. Weller PH, Gupta J, Jenkins PA, Baum JD (1976): Pharyngeal lecithin/sphingomyelin ratio in newborn infants. Lancet 1:12-15.

Correspondence: R.J. Norman, Department of Chemical Pathology, Faculty of Medicine, University of Natal, P.O. Box 17039, Congella 4013, Durban, South Africa.