HISTOLOGICAL EVALUATION OF PLACENTAL ARCHITECTURE IN CASES OF INTRAUTERINE GROWTH RESTRICTION

An Undergraduate Research Scholars Thesis

by

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ABSTRACT

Histological Evaluation of Placental Architecture in Cases of Intrauterine Growth Restriction

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Intrauterine growth restriction or IUGR is the setback in development arising from restricted fetal growth during gestation. This restricted growth is identified by precursors not only at parturition, but additionally during the first stages of development within the uterus. This condition when deemed severe can be related to the crucial role of the placenta for fetal development. IUGR can result from placental abnormalities in modification and transport of nutrients and placental absorption (G.Pardi et al., 2002). Environmental factors of drinking alcohol or substance abuse, or lack of nutrition from the mother during pregnancy may potentiate IUGR. The lack of proper nutritional balance in mothers, from gestation to post parturition is commonly seen within developing countries, where post-partum children continue to lack adequate nutrition and activity. Past parturition, the developmental setbacks resulting from IUGR continue far beyond neonatal and adolescence into adult health complications such as heart disease.

To define a spectrum of changes within the histological architecture of the placenta resulting from either nutrient restricted or adequate diets, unbiased stereology methods of data

collection and assessment of the sheep placenta were performed. Uniform random sampling of the stereology placental tissue images was fixed on slides and measured to calculate the total surface area and surface density. The data collected was compiled to give the 3-dimensional image of the tissue, which allowed measurement and analysis of the histologic architecture of the placenta. The data in its entirety illustrated the results of a concentrated diet versus an adequate diet on the placenta. The mean value of total placental tissue surface area in high concentrated ewe diet was greater when compared to low concentrated diet ($p < 0.05$). The mean total surface area of low concentrated ewe diet was less when compared to control ($p<0.005$). There was no significant difference between mean total surface area in high concentrated diet and control ($p>0.5$). The data supports the direct connection between placental health and maternal nutrition.

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NOMENCLATURE

IUGR	Intrauterine Growth Restrictions

CHAPTER I

INTRODUCTION

Intrauterine growth restriction or IUGR is the setback in development that arises from restricted fetal growth during gestation. This restricted growth is identified by precursors not only at parturition, but additionally seen during the first stages of development within the uterus. During gestation, the placenta plays a crucial role for the development of the fetus, providing oxygen and nutrient exchange from mother to progeny via the intertwined maternal fetal circulation. The placenta provides adequate nutritional demands placed by the rapid fetal advancements seen during pregnancy. IUGR can present as disturbance of the placental modification of transport of nutrients and placental absorption (G.Pardi et al., 2002).

By definition, a fetus deemed affected by IUGR has their birth weight or length fall 2 standard deviations below the average for their developmental stage (P. Chatelain, 2000). Concurrently, IUGR is also determined by the fetal heart rate and the speed of the umbilical artery blood flow (G.Pardi et al., 2002). In greatest severity, the circumstances of IUGR lead to fetal death. There are many suspected causes of IUGR, but the majority of factors impact placental function. Potential confounding factors of IUGR include, but are not limited to environmental factors, such as ingesting of alcohol, substance abuse, as well as the health of the mother carrying the fetus including improper nutrition during pregnancy (T.Pagano, 2014).

IUGR affects offspring not only during pregnancy, but also in neonatal life and during their adolescence years. Effects cause continued health problems into their adult years with issues such as cardiovascular and kidney disease. IUGR, and the resulting low birth weight infant is commonly seen within rapidly developing countries because mothers lack proper nutrition

during their gestation period, which are then provided a rapid diet change of a shift to high calories coupled with low activity (Crume et al., 2013).

Currently, the environment presented to mothers within developing countries leads to high instances of intrauterine growth restrictions. When a mother is chronically lacking essential nutrients before and during gestation, this leads to a state of nutritional stress in utero (Perez-Escamilla et al., 1992). This constant state of nutrient deficiency leads to compromise of the fetal development during pregnancy, in turn resulting in decreased birth weights of their children. Further, the environment of mothers seen with IUGR pregnancies raises concern on their child's postnatal growth.

The environment of the mother directly affects her child; environmental exposures to the mother during gestation can alter the uterine environment and result in low birth weight, and delayed onset of growth for her child. Surroundings, intake of nutrition, and potential disease contracted by the mother can result in post-natal delayed development. Such delayed development can result in health issues not only during the post-natal period, but onto adulthood, such as immunodeficiency. Problems of immunodeficiency can occur during lymphatic development during the 2nd and 3rd month of pregnancy, where decreased development of this system due to growth restriction in utero leads to smaller tissues development resulting in a suppressed immune system postnatally (McDade et al., 2001). In developing countries where vaccination protocols for infants are not yet established or even heard of, infants suffering complications due to IUGR are at greater risk for contraction of disease and health problems that can endanger their longevity.

This change in environment is seen not only in the child, but additionally within the placenta itself. Seeing as the placenta is the central organ of nutrient and waste exchange between mother

and child, when this organ is altered due to lack of proper nutrition, this affects the growth of the fetus.

CHAPTER II

METHODS

The data collected is the surface density and total volume from the slide images using the Nikon Elements quantitative software. Researcher attended laboratory three times a week for 1-3 hours collected data and exported collected quantitative data into Microsoft Excel. Once all images from each slide set are documented, the data was compiled into equations to calculate the surface density and total surface area.

The equations used are taken from the *Unbiased Stereology: A Concise Guide* by Peter Mouton, where the surface density is calculated using the following: $S_v = 2*\sum I/\sum L$, where $\sum I$ is the total number of line intersections across all sections of the image, and $\sum L$ is the total length of the line used (800 µm) by all sections. Once the surface density of the slides is calculated, that value is used to calculate the total surface area using the following: Total $S = S_v * V_{ref}$, where S_v is the calculated surface density, and V_{ref} is the reference volume of the placentome volume of the Ewes. Once the data in its entirety is complied, the data of the placental architecture was statistically analyzed utilizing paired T test (P(T<=t) two-tail) on the basis of its restriction versus adequate data during gestation, seen in the mean total surface area.

CHAPTER III

RESULTS

The data collected and displayed in Figure 1 illustrates the surface density and total surface area of both the maternal and fetal tissue taken from ewes used in study. The ewe is identified by their given number, and the designated tissue sample is labeled by either “Mat Car” for maternal caruncle or “Fet Cot” for fetal cotyledon. Descriptive data expressed as mean and standard error below. Data was analyzed utilizing pair T- two tailed test (P(T<=t) two-tail) in order to determine the statistical variance between the mean total surface area of control diet and restricted diet (50%) with a high and low subpopulation. Mean total surface area resulted in experimentation (mean±SEM): 560.5±39.06 μm^2, 239.4±35.50 μm^2 and 395.4±48.80 μm^2 for control, low and high respectively. Figure 1 depicts the mean total surface area of the control, low and high sample sets.

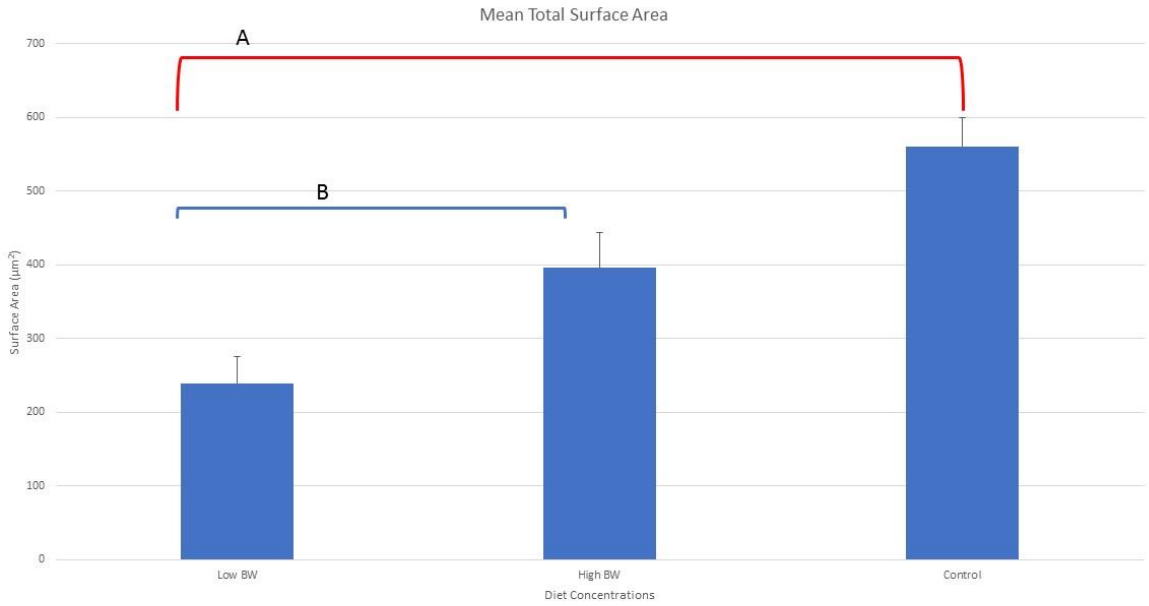

Figure 1: Mean total surface area of control, low and high subgroups. A: Mean total surface area of high resulted greater surface area when compared to low ($p < 0.05$) B: Mean total surface area of low less than when compared to control ($p<0.005$). Results indicated that mean total surface area does not differ between high and control ($p>0.5$).

CHAPTER IV

CONCLUSION

IUGR is known to hinder the placental methods of nutrient transport and absorption (G.Pardi et al., 2002). Nutritional stress in utero then follows in regards to lack of nutrient transport and absorption in cases of malnutrition of a mother chronically lacking nutrient uptake during time of gestation. (Perez-Escamilla et al., 1992). This continuous circumstance of nutrient deficit causes compromise of the fetal advancement in pregnancy, subsequently in reduced birth weights of progeny. Compiled data from microscopic slides of placentome tissues resulted in the calculated surface density, giving rise to the total surface area of analyzed tissue.

The mean total surface area of the control, low and high illustrated the reduced surface area of the placenta with a restricted diet. Ewes with a 50% restricted diet and low birthweight fetuses had tissues with less surface area compared to high and controls. Control group and high groups did not display any difference in the mean total surface area. Data resulted in decreased total surface area in 50% low group when compared to both control and high, giving rise to a hypothesis that there is restricted placental development in a subpopulation of mothers lacking proper nutrition uptake.

Data indicated mean total surface area of placenta of low birthweight fetuses and resulted in statistical variance when compared to that of the control, suggesting that the placental surface area is greatly reduced when given restricted diet. Additionally, when comparing mean total surface area of high versus low, mean total surface area of low was significantly lower than that of high, illustrating the reduced surface area of placenta due to diet restriction in a subpopulation of individuals. Restricted growth seen in the low birthweight individuals of the 50% restricted

diet can attest to placental disturbances of nutrient transport from maternal to fetal tissue. In turn, restricted placental development leads to delayed fetal development and growth, signifying signs of IUGR for fetus.

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