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GUINEA PIG FETUS DOES NOT CHANGE ITS PRESENTATION DURING SECOND HALF OF GESTATION

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Abstract: Objectives: The aims of this paper are: 1. To investigate spontaneous changes in fetal presentation during the second half of gestation in guinea pig fetus, 2. to observe provoked changes of fetal presentation, by passive positioning fetus into an upside-down position and "sitting" position. Eight fetuses from singleton pregnancies were included. Experimental design: Ultrasound examinations (Toshiba Nemio SSA-550A apparatus) were started on the 26^{th} day of gestation (GD). Each fetus was examined 2-3 times in each 5-day interval until the end of gestation. First the spontaneous orientation of fetus was determined by tracking along the longitudinal and transversal axes of the fetus with the ultrasound probe. Then the fetus was brought into a head-down position relative to gravity for 5 minutes. The examination was repeated with the fetus in the head-up position. Results: In the period from the $26^{th} - 30^{th}$ GD in 7 of 21 observations spontaneous changes of presentation were observed. From the 31^{st} GD all 133 observations were negative regarding spontaneous changes of presentation and situs. The chi-squared test showed that difference in spontaneous changes of presentation and situs before versus after 30^{th} day of gestation had a statistical significance ($\chi^2 = 25.16$ p <0.05). Frequency of ultrasound examinations before and after 30^{th} day of gestation was not statistically significant (Fisher exact chi square p = 0.46, p > 0.05). Fetuses were brought into a head-down and head-up 154 times. In none of these attempts did a fetus change presentation during the five minutes of observation. Conclusion: Guinea pig fetuses do not spontaneously or at provocation change presentation after 30^{th} GD.

Key words: fetal presentation; gestation; guinea pig; ultrasound

Introduction

Etiology of fetal presentation in mammals has not been fully elucidated yet (1,2). Adequate presentation at birth is significant since anterior presentation in herd animals enables physiological delivery. Posterior presentation is accompanied by an inadequate dilatation of the birth channel, and consequently, a difficult delivery (3). In a previous paper, it was postulated that fetal presentation is a

consequence of postural development (1). In herd animals, locomotor-postural development occurs during the second half of gestation. In the anterior presentation, the cranial part of the body is above the caudal part. The hind legs are the source of the main propulsive force. When the fetus has its hind legs below its cranial part, it has an optimal mobility. In up-side down position the fetus has more difficulties to move (1).

Guinea pig are precocial rodents that have full locomotion at birth. In previous studies with guinea pigs it was shown that a reflex of turning from supination to pronation can be induced in the guinea pig fetus and that its intrauterine behavior may be gravity-dependent (4). For guinea pigs, there is no data about spontaneous or provoked changes in fetal presentation during gestation.

The aim of this paper is to investigate spontaneous changes in fetal presentation and situs during the second half of gestation. In addition, a secondary aim is to determine the existence of active turning of the fetus through 180 degrees, from its passive positioning into an upside-down position. It is assumed that in the second half of gestation a fetus that is passively positioned in an upside-down position will turn around though 180 degrees.

Material and method

Experimental animals

The study subjects were albino guinea pigs (Cavia porcellus) obtained from the Department of Biochemistry, Faculty of Medicine, Clinical Center of Vojvodina, Novi Sad. The experiments with animals were approved by the Ethics Committee of the University of Novi Sad No 04-29/62). The guinea pigs were kept in 400Wx1000Lx300H mm plastic containers in a harem system: two females and one male. Pregnant females were moved to 300Wx300Lx300Hmm plastic containers after the 60th day of gestation, where they were kept separate until delivery, and afterwards for the first 15 days with the offspring. The animals had a standard commercial pellet diet and ad libitum water enriched with vitamin C (30 mg/100 ml water). Artificial cycles with 12 hours of light (08:00-20:00) and 12 hours of dark were provided. The room temperature was maintained at 22 ± 2°C. The air was recirculated 10 times per hour.

Inspection of vaginal introitus was performed daily, and the day of vaginal membrane perforation was taken as the first day of gestation. Individual guinea pigs were identified by yellow patterns on their backs. Eight pregnant guinea pigs with one fetus were included. The number of fetuses was determined by ultrasound examination. Pregnant females were shaved before examination; the shaving of the abdominal region was made during a short-term inhalatory ether narcosis on the 25th day of gestation.

Ultrasound examination

Ultrasound examinations were started on the 26th day of gestation, until when the first movements of the guinea pig fetus usually occurs (5). During gestation that usually lasts for 66 days each fetus was examined 2-3 times in each 5-day interval (gestation days 26-30, 31-35, 36-40, 41-45, 46-50, 51-55, 56-60, ≥61). Immediately before the examination the pregnant guinea pigs were supported in a supine position on a 15x30cm board using plastic strips with clasps, fastened over the thoracic area and both hind legs. The strips were pulled through holes in the board near the body of the animal and fastened on the other side of the board.

Ultrasound examinations were performed with a Toshiba Nemio SSA-550A apparatus with a 6-11 Hz linear probe. Pregnant females were brought into supination with the board on which they were fastened to. The orientation of fetus was determined by tracking along the longitudinal and transversal axes of the fetus with the ultrasound probe. The position of the fetus was determined on the basis of the positions of its head, spine, heart, forelimbs and hind legs (Image 1). Then the board was rotated until the fetus was brought into a head-down position relative to gravity and any changes in fetal presentation were observed for 5 minutes. The same subjects were used as controls. After a 2-minute pause, the examination was repeated with the fetus in the head-up position.

Statistical analysis

Statistical tests were completed using the SPSS software (version 21, 2012, IBM, Armonk, NY, USA). Results with a p - value of p<0.05 was accepted to be statistically significant.

Results

Spontaneous changes of presentation

Table 1 - summarizes data related to fetal presentation and situs at the beginning of ultrasound examination, with pregnant females in supination. A transversal lie with the head on the right side was the most frequent situs. At the last examination before delivery there was one anterior and one posterior presentation of the

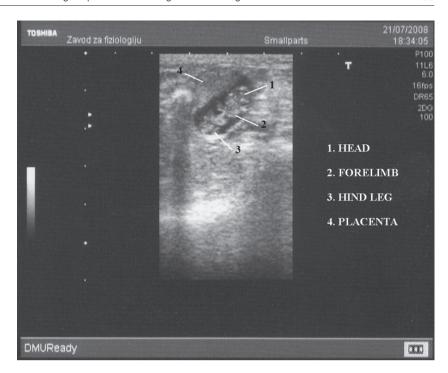


Figure 1: 29-days old guinea pig fetus

Table 1: Fetal presentation and situs at the beginning of ultrasound examination, with pregnant females in supination

| Days of gestation | Longitudinal lie No (%) | | Transverse lie No (%) | | | | Total |
|-------------------|-------------------------|------------|-----------------------|-------------|-----------|------|--------|
| | AP | PP | HL | HR | SP | USDP | No (%) |
| 26-30 | 5 (23.80%) | 7 (33.33%) | 3 (14.28%) | 6 (28.56%) | 0 | 0 | 21 |
| 31-35 | 4 (17.39%) | 6 (26.08%) | 3 (13.04%) | 8 (34.74%) | 2 (8.69%) | 0 | 23 |
| 36-40 | 6 (28.57%) | 5 (23.80%) | 4 (19.04%) | 6 (28.57%) | 0 | 0 | 21 |
| 41-45 | 5 (31.25%) | 4 (25%) | 1 (6.25%) | 6 (37.50%) | 0 | 0 | 16 |
| 46-50 | 4 (23.52%) | 4 (23.52%) | 3 (17.64%) | 6 (35.29%) | 0 | 0 | 17 |
| 51-55 | 3 (16.66%) | 2 (11.11%) | 5 (27.77%) | 8 (44.44%) | 0 | 0 | 18 |
| 56-60 | 5 (26.31%) | 3 (15.78%) | 2 (10.52%) | 9 (47.36%) | 0 | 0 | 19 |
| ≥ 61 | 2 (10.52%) | 3(15.78%) | 4 (21.04%) | 10 (52.63%) | 0 | 0 | 19 |

fetus, as well as six transverse lies; in 4 cases with the head on the right side and in two cases with the head on the left side.

In the period from the 26th to the 30th day of gestation a total of seven changes in presentation and situs (in five fetuses) were observed, that indicate turning around the longitudinal axis of the body for 180°. From the 31st day of gestation none of the fetuses in our sample changed their body orientation by 180°. They oscillate around one position for 45° during whole second period of gestation. This means that no fetus made a

spontaneous turn around its longitudinal axis. The chi-squared test showed that difference in changes of presentation and situs before vs after 30^{th} day of gestation had a statistical significance of x^2 = 25.16 p <0.05. The strength of these association is very strong (phi = -0.77, Cramer's V = 0.77, p < 0.001). Frequency of ultrasound examinations before and after 30^{th} day of gestation was not statistically significant (Fisher exact chi square p = 0.46, p > 0.05). Oscillations around one position by 45° were present in all experimental animals during the whole examined period. There is no

statistically significant difference in frequency of oscillations around one position for 45° between each examined period of pregnancy (Fisher exact chi square p = 0.46, p > 0.05).

Provoked changes in presentation

Fetuses were brought into a head-down position for a total of 154 times and in none of these attempts did a fetus through 180° degrees during the five minutes of observation. The fetuses did not change their presentation. The same results were obtained when fetuses were positioned in a head-up position after a 2-minute pause.

Discussion

The results of this study indicate that there are two periods of gestation in the guinea pig with regard to its situs and presentation. After the occurrence of the first movements on gestation days 25 and 26, the fetus is capable of passive or active turning around its longitudinal axis for 180° degrees only for a few days. After that, throughout the second half of gestation, its intrauterine position is fixed and it is no longer capable of turning around its longer axis. This finding is contrary to other mammalian species in which postural development occurs prenatally. The incidence of different presentations and lies during gestations with one fetus in human species (6,7) and herd precocial mammals (8-10) indicates the existence of three stages. The beginning of the first stage is characterized by equal proportions of longitudinal and transverse lies with equal proportions of breech and cephalic presentations within the longitudinal lie. During this stage, there is an increasing incidence of longitudinal lie with a proportional decrease in transverse lie. In the second stage, which occurs in the second half of gestation, a transverse lie is almost completely absent, whereas in the longitudinal lie there is an increasing incidence of anterior (cephalic) presentation and a proportional decrease in posterior (breech) presentation. By the end of this stage, around 95% of fetuses are in a longitudinal lie with cephalic presentation. In the third stage, during the last weeks of gestation, there is a further mild increase in the incidence of longitudinal lie with cephalic presentation (2,11). Published data show that in humans and herd mammals there is spontaneous turning around the longer axis in the second half of gestation (2,9). Studies with exteriorization of guinea pig fetuses have shown that turning around the shorter axis occurs when gestation is terminated around the 60th day. Taking an upright position and maintaining balance occurs around the 63rd day of gestation (12). In a study dealing with intrauterine induction of righting reflex from supination to pronation, turning to one side was present already around the 40th day of gestation, and it was the most frequent after the 60th day of gestation (4). Despite the presence of postural reactions in the guinea pig fetus in the second half of gestation and its ability to turn around the shorter body axis, turning around the longer body axis could not be induced by positioning the fetus in an upside-down position. In a sheep fetus, in the period of an exclusive increase in the incidence of anterior presentation, it is possible to induce the fetus to turn around its longer axis by positioning it in a head-down position (2). Therefore, it is likely that the turning around the longitudinal body axis in the guinea pig fetus is not possible due to physical characteristics i.e. lack of space. The guinea pig fetus is therefore not a suitable and adequate experimental model for confirmation of the concept that presentation of the human and herd mammals fetuses is influenced by their intrauterine postural development.

A limitation of the present study is that it was not able to register changes in fetal position greater than 180° degrees. A 270° degree turn would have been registered as a 90° degree turn. The absence of erratic changes in fetal position indicates that this problem does not affect the results of the study. Oscillations of 45° from the basic position are probably caused by changes in the position of the internal organs.

In order to confirm or discard the hypothesis on the influence of postural development on presentation of the fetus, it is necessary to advance the experimental model of the sheep fetus. A problem with the sheep fetus as an experimental model is that the fetus assumes a transverse lie when the pregnant female is positioned into a sitting position. This manipulation with the pregnant female is performed in order to provoke a fetus in anterior presentation to assume a head-down position and turn for 180° degrees. In order to overcome this problem, investigators used external pressure with their palms on the

abdominal walls of pregnant females (2). However, this is an inappropriate method because of external, physical stimulation of fetal movements. It is necessary to surgically fix the horn of the uterus (hornopexia) to accomplish stable longitudinal situs of the fetus prior to studying its reaction to changes of gravity vector in the intrauterine environment.

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PLODOVI MORSKIH PRAŠIČKOV NE SPREMENIJO LEGE V DRUGI POLOVICI BREJOSTI

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Povzetek: Namen prispevka je: 1. raziskati spontane spremembe v legi plodu v drugi polovici brejosti pri plodovih morskih prašičkov; 2. opazovati vzpodbujene spremembe lege plodov v položaj od zgoraj-navzdol in v »položaju sedenja«.V raziskavo je bilo vključenih osem plodov iz ene brejosti. Z ultrazvočnimi pregledi (aparat Toshiba Nemio SSA-550A) smo pričeli 26. dan brejosti. Vsak plod je bil pregledan 2-3-krat v 5-dnevnih razmikih do konca brejosti. Najprej je bila ugotovljena spontana orientacija plodu z določanjem njegove vzdolžne in prečne osi z ultrazvočno sondo. Potem je bil plod za 5 minut obrnjen v položaj z glavo navzdol, sorazmerno s težo. Pregled plodu je bil ponovljen v položaju z glavo navzgor. V obdobju od 26. do 30. dneva brejosti smo v 7 od 21 opazovanj opazili spontane spremembe lege plodu. Od 31. dneva brejosti dalje pri vseh 133 opazovanjih ni bilo opaziti spontanih sprememb lege plodu. Analiza Hi-kvadrat je pokazala, da je razlika v spontanih spremembah lege plodu pred in po 30. dnevu brejosti statistično značilna ($\chi^2 = 25,16$ p <0,05). Pogostnost ultrazvočnih preiskav pred in po 30. dnevu brejosti ni bila statistično značilna (Fisherjev natančni Hi-kvadrat test p = 0,46). Plodovi so bili postavljeni v položaj z glavo navzdol in navzgor 154-krat. V nobenem od teh poskusov ni prišlo do spontane spremembe lege plodu med petminutnim opazovanjem. Ti rezultati torej kažejo, da plodovi morskih prašičkov ne spremenijo lege spontano ali po provokaciji po 30. dnevu brejosti.

Ključne besede: sprememba lege plodu; brejost; morski prašiček; ultrazvok