Anesthetic management in intrauterine surgery to evaluate an experimental model of myelomeningocele in non human primates (*Macaca mulatta*)[1]

Anestesia em cirurgia intra-uterina para avaliar um modelo experimental de mielomeningocele em primatas não humanos (*Macaca mulatta*)

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ABSTRACT
Purpose: Evaluate the anesthetic management in intrauterine surgery to induce myelomeningocele in non human primates *Macaca mulatta*. **Methods**: A total of nine fetuses had intrauterine surgery; laminectomy was performed on them in L5 and L6. The studied variables were: maternal death, fetus death, cardiac frequency, respiratory frequency, arterial pressure, temperature, and oxygen saturation. **Results**: No maternal or fetal deaths occurred; the only variable that was reported below the normal ranges was temperature. **Conclusion**: No maternal or fetal deaths occurred; the only variable that was reported below the normal ranges was temperature.
Key words: Anesthesia. Laminectomy. Meningomyelocele. Hypothermia. *Macaca mulatta*.

RESUMO
Objetivo: Avaliar o manejo anestésico em cirurgia intra-uterina para induzir mielomeningocelo em primatas não humanos, *Macaca mulatta*. **Métodos**: Operaram-se um total de nove fetos in útero que foram submetidos à laminectomia em L5 e L6. As variáveis a estudar foram mortes maternas ou fetais, freqüência cardíaca e respiratória, pressão arterial, temperatura e saturação de oxigênio. **Resultados**: Não se apresentaram mortes maternas ou fetais, a temperatura se manteve abaixo dos 36°C, não tendo repercussões no bem-estar dos macacos. **Conclusão**: Não ocorreu nenhum óbito materno ou fetal, sendo que a única variável abaixo do normal foi a temperatura.
Descritores: Anestesia. Laminectomia. Meningomielocele. Hipotermia. *Macaca mulatta*.

[1]Research performed at Hospital General Dr. Manuel Gea González, México.

Introduction

In fetal surgery, the pregnant monkeys and their fetuses experience pain and stress; both generate homodynamic changes that could provoke hypoxia, acidosis, hypotension and brain damage, that could kill the fetus or the mother[1]. That is why an adequate anesthetic and analgesic management is important. The objective of this report is to evaluate the anesthetic management used during the intrauterine laminectomy surgery, to induce myelomeningocele in non human primates *Macaca mulatta*.

Methods

The research was done in accordance with the Mexican Official Norm NOM-062-ZOO-1999 (Technical specifications for the production, care and use of laboratory animals and was approved by the Internal Committee of Care and Use of Laboratory Animals and the CAMINA, A.C. Research Center's ethical and research commissions. Nine female monkeys *Macaca mulatta* between the ages of 4 and 10, weighing between 4500 grams and 7500 grams, and in week twelve of gestation were the subjects of this study. A total of nine fetuses had surgery, divided in three experimental groups, on all of them laminectomy in L5 and L6 was performed.

The two models used in the surgical correction in order to protect the exposure of the spinal cord to the amniotic fluid were: mesh placement in three fetuses, and suture of the skin in other three fetuses. Other three fetuses exposed to the amniotic fluid were evaluated as models, simulating myelomeningocele. A tenth fetus was evaluated as a control model to compare the normal development and maturity, both intrauterine and extrauterine.

Animal preparation

The animals were isolated from the group in which they lived 12 hours prior surgery, they had an eight hour fasting period, and were placed in stainless steel metabolic cages. The cages were special for non human primates and were in a room with controlled climate at a temperature of 23°C. They were physically restrained to administer Tiletamine-Zolazepam (Zoletil®)[2] 4mg/kg by intramuscular route, an hour before surgery.

A catheter 22 (Becton Dickinson Insytec® 0.9X25 mm) was placed in the cephalic vein, and a lukewarm 5% glucosed solution, 50 ml, was administered through a venoclyisis (Flebotek®) during 30 minutes. One hundred ml of a lukewarm 0.9% sodium chlorine solution was administered during the surgical time and postoperative. Cardiac frequency, respiratory frequency, arterial pressure, oxygen saturation and temperature were monitored with an Ohmeda Cardiocap/5® monitor. The electrodes were collocated on the thorax, to monitor the heart rate and the respiratory rate; to measure the arterial pressure we put the cuff on the inside part of the right leg thigh. Oxigen saturation was measured from the right hand thumb with the SpO2 Sensor.

Anesthetic management

Atropine, 0.02 mg/kg was administered IV, a number 3 face mask was placed, and 3 – 4 liters of oxygen with 4% of Sevoflurane was administered per minute with a vaporizer (Ohmeda®)[3]. Palpebral and deglutory reflexes were watched so intubation with and endotraqueal tube without balloon numbers 4, 4.5 or 5 could be placed when they disappeared. The size of the endotraqueal tube used depended on the individual, and a pediatric laryngoscope (Welch Allen®) with a straight blade size 2 was used to place the tube. Once the individual was intubated, the concentration of sevoflurane was reduced to 2%, staying like that for the rest of the perioperative time[4]. Before the surgery started, 10 µg of Fentanyl were administered IV. During the surgical procedure the variables were registered every 10 minutes. When the surgery was over 0.1 – 0.2 mg/kg of Butorfanol were administered IM[5]. The extubation process started with the closure of the sevofluarane, and ended with the removal of the tube when the palpebral and deglutory reflexes were present as well as leg and arm movement. They were disconnected from the monitor until the cardiac and respiratory rates, temperature, blood pressure, and oxygen saturation, were within normal rates. The last thing to be removed was the venous catheter, corroborating that no bleeding was present in order to take the individual to its individual cage to continue with its recovery.

The design of the study was descriptive, open, prospective and longitudinal. The analyzed variables were: mother mortality, fetus mortality, heart rate, respiratory rate, temperature, arterial pressure, oxygen saturation, mother's age and weight, surgical time and anesthesia. The variables were analyzed with simple frequency, maximums and minimums, mean and standard deviation.

Results

There were no reports on maternal or fetuses deaths in the nine females that were surgically operated on. The average duration of the intrauterine surgeries was 77.3 minutes (60 to 105 minutes) and the anesthetic time varied from 64 to 140, an average of 92.7 minutes (Table 1).

TABLE1 - Demographic dates of the mother, fetal surgery, surgery time, duration of anesthesia and relation between mother or fetal death in the immediately transoperatory

Case N = 9	Weight (g)	Temperature °C	Mother age (year)	Gestation time (week)	Fetal surgery	Surgery Time (minutes)	Duration of anesthesia (minute)	Mother death	Fetal death
1	6000	37.4	4	12	Closing the skin	60	85	NO	NO
2	5100	37.45	4	12	Closing the skin	80	91	NO	NO
3	7000	36.42	10	13	Correction with a silicone mesh	75	85	NO	NO
4	5735	37	10	13	Exposed	62	98	NO	NO
5	5000	36.75	10	14	Exponed	120	140	NO	NO
6	6200	37.76	8	13	Correction with a silicone mesh	72	64	NO	NO
7	5600	37.33	4	12	Exponed	62	72	NO	NO
8	6100	37.45	7	13	Closing the skin	60	65	NO	NO
9	7000	36.82	9	12	Correction with a silicone mesh	105	135	NO	NO

In this table we can observe the weight, age, temperature, from the mother and the surgical procedure that the fetus had. No reports of maternal or fetal deaths occurred during anesthetic management.

The evaluated variable means obtained during the surgical time were as follows: heart rate of 138.9 beats per minute (Figure 1), respiratory rate of 34.3 breaths per minute (Figure 2), temperature of 34.8°C (Figure 3), oxygen saturation of 96.8% (Figure 4), systolic pressure of 147.9 mmHg (Figure 5) and of diastolic pressure of 73.6 mmHg (Figure 6).

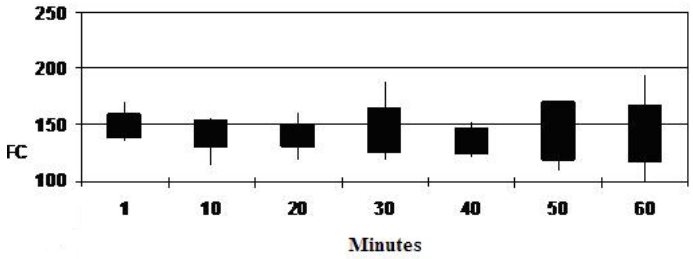

FIGURE 1 - Cardiac Frequency during transoperatory

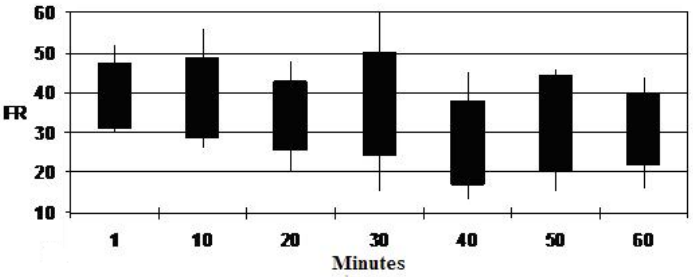

FIGURE 2 - Respiratory Frequency during transoperatory

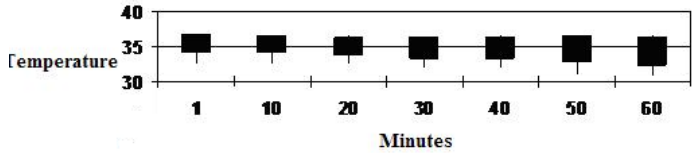

FIGURE 3 - Temperature during transoperatory

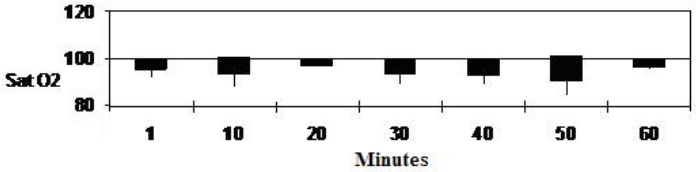

FIGURE 4 - Oxygen Saturation during transoperatory

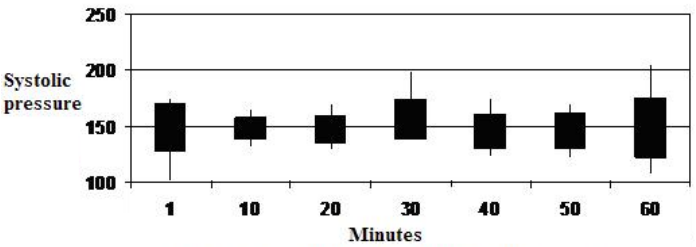

FIGURE 5 - Systolic Pressure during transoperatory

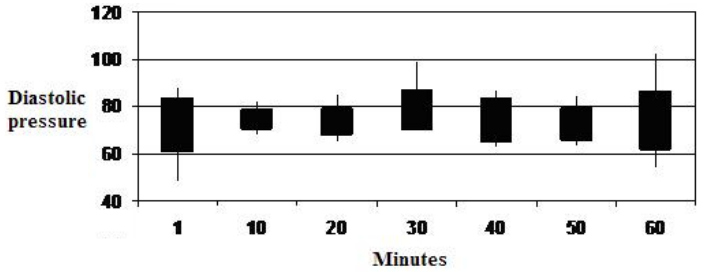

FIGURE 6 - Diastolic Pressure during transoperatory

Discussion

In the last years there has been diverse research in animals, like the one performed by Heffez *et al.*[6,7], in rats, Pedreira *et al.*[8]. in rabbits, Meuli *et al.*[9] in sheep, Pedreira *et al.*[10] in ovine fetuses and Michjeda[11] in non human primates *Macaca mulatta*; creating intrauterine surgical models to reproduce different diseases and perform its correction. Also George and Fuh[12], describe the use of non-human primates (*Macaca mulatta*), as models for the surgical correction of diseases for the neural tube. These have allowed the intrauterine correction of different pathologies in humans, such as myelomeningocele, sacrococcygeal teratoma, obstruction of the urinary tract and lung adenomatoide disease[13,14].

In the anesthetic management in non human primates done by Harrison *et al.*[15], they used Ketamine for immobilizing the pregnant monkey, and the anesthesia was maintained with a mixture of 60% nitrous oxide, oxygen and 0.23 to 1% of halothane, with a total flood of 1 liter per minute. Temperature was regulated trough heating pads and stand lamp. Neither maternal nor fetus deaths were reported in the study. In our research, the chemical restrain of the pregnant monkeys was done with Tiletamine-Zolazepam, this combination is a dissociate anesthetic and tranquilizer with minimal cardiovascular effects, and more powerful than ketamine[16]. In the induction of the anesthesia a mixture of 4% sevoflurane with 3 liters of oxygen per minute was used allowing the intubation process without any difficulties. Being an inhalational halogene anesthetic useful for induction, sevoflurane is used in concentrations of 2 - 4%. It does not irritate the respiratory tract, and it shows a rapid recovery[17]. Fentanyl was used for the surgical analgesia which is a synthetic opiate analgesic. Due to the fact that it is more liposoluble than morphine, it reduces the risk of respiratory depression[18].

In intrauterine surgery of non human primates, Johnson-Delaney[19] obtained cardiac rates between 100 and 150 beats per minute; Wolfensohn and Lloyd[20] obtained 120 - 180 and Gonder *et al.*[21], 149 - 207. Johnson-Delaney[19] registered respiratory rates of 40 to 65 breaths per minute, Wolfensohn and Lloyd[20] of 32 to 50, and Hrapkiewicz *et al.*[22] of 10 to 25. The minimum and maximum values that we obtained were: cardiac rate of 98 to 200 beats per minute and respiratory rate of 11 to 60 breaths per minute.

Johnson-Delaney[19] reports data of blood pressure, with a systolic pressure mean of 125 mmHg and a diastolic pressure mean of 75 mmHg. In our study the means obtained of systolic pressure was 147.9 and 73.6 of diastolic pressure; all of these data is similar to those published Hrapkiewicz *et al.*[22].

The temperature reported by Johnson-Delaney[19], Wolfensohn and Lloyd[20], and Hrapkiewicz *et al.*[22] in the

intrauterine surgical management of non human primates were maintained between 36 and 40°C. The normal temperature of a pregnant Rhesus monkey is of 36 to 40°C, in the case of the nine pregnant females used in the study, they had an average temperature of 34.8°C during the surgical time. Anesthesia time varied from 65 to 135 minutes, with an average of 93.6 ± 27 minutes. In the females that we observed that the temperature decreased, there was an increase in surgical time. In this study, we tried to keep temperature through intravenous lukewarm solutions, but it was not enough to avoid hypothermia.

Conclusion

In the present study we describe the anesthetic management in intrauterine surgery to induce myelomeningocele in non human primates *Macaca mulatta*. No maternal or fetal deaths occurred; the only variable that was reported below the normal ranges was temperature.

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