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MFK PROVIDES ANALGESIA AND CARDIOVASCULAR STABILITY DURING LAPAROSCOPIC CHOLECYSTECTOMY IN SWINES

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Abstract

Laparoscopic surgery gained a lot of field in human and veterinary medicine, replacing successfully many invasive surgical techniques. Laparoscopic cholecystectomy is a reality and units must convert from open to endoscopic technique. To this end we have used the pig, whose biliary anatomy resembles the human, as a laboratory in vivo training model. This aspect has led to imagining and establishing cholecystectomy techniques in pigs that were subsequently applied to humans. We conducted the current study to address the need for a systematic investigation of anaesthetic and analgesic protocols in Landrace pigs. The goal of this study was to evaluate a partial intravenous anaesthetic (PIVA) protocol for Landrace pigs that yielded sufficient sedation for peripheral vascular catheterization, analgesia and miorelaxation for laparoscopic techniques and would have minimal cardiovascular effects while being safe for the patient. The study was carried out during several training session for surgeons on laparoscopic techniques. Sixteen pigs (weight between 15 and 30 kg, with a mean weight of 21 kg) were anaesthetized for the procedures. Pigs were randomly allocated to one of the following groups: MFK group (receiving midazolam 0.2 mg/kg/h, fentanyl 10µg/kg/h and ketamine 10µg/kg/min) or KL group (receiving ketamine 10 µg/kg/min and lidocaine 30µg/kg/min). Heart rate, respiratory rate, blood pressure and saturation of oxygen were monitored throughout the anaesthesia and recorded every 5 minutes using a vital signs monitor. Muscle relaxation was appreciated using a subjective scale. Midazolam-fentanyl-ketamine provided better analgesia and muscle relaxation, with minimal cardio-vascular effects compared to the KL protocol. Both protocols can be used in swine and ensure stable cardiovascular parameters during general anaesthesia in this species.

Key words*: laparoscopy, cholecystectomy, midazolam-fentanyl-ketamine, anesthesia*

INTRODUCTION

Laparoscopic surgery gained a lot of field in human and veterinary medicine, replacing successfully many invasive surgical techniques.

Laparoscopic cholecystectomy decreases postoperative pain, hence reducing the need for postoperative analgesia, shortens the hospital stay from 1 week to less than 24 hours, and returns the patient to full activity within 1 week (compared with 1 month after open cholecystectomy) (Tanase et al. 2015). If such techniques become standard, as seems likely, it will be necessary to develop safe and realistic learning and training models (Draghici et al. 2014).

The swine biliary tract closely resembles the human one, making this species a suitable anatomical model (J Sˇ tembı´rek et al. 2012; Kirwan et al. 2001).

Swine (*Sus scrofa*) are also common models for cardiovascular injury and intervention that largely have replaced traditional canine cardiology models (swine have similar coronary artery distribution and effective collateralized blood flow to the myocardium after coronary artery blockage) (Jan R Linkenhoker et al. 2010).

Anaesthesia and analgesia is frequently required for swine in research due to their use as preclinical models (translational research). Selection of an appropriate protocol which considers the physiologic effects of the pharmacologic agents for anesthesia/analgesia is an important aspect of designing an experiment (Smith AC et al. 2008). However, pigs are difficult to restrain and anaesthetize due to their size, temperament and resistance to

sedative drug combinations, including those with morphine (Kaiser 2006). Xylazine is an α2 agonist used for sedation of pigs. It is short acting, so it is suitable for continuous rate infusion (CRI) administration. It also has analgesic properties, provides miorelaxation, but the main side effects are related to cardiovascular depression (bradycardia and transitory hypertension), hyperglycemia and increased diuresis (Grimm et al. 2015).

Ketamine is a dissociative anaesthetic agent with a wide range of safety in swine (11-33 mg/kg IV, IM or SC). It is generally an effective restraint agent, safe cardio-respiratory (sympathomimetic effect) and analgesic (antagonist action on the N-methyl-D-aspartate (NMDA) receptors), but does not provide miorelaxation (Ajadi et al. 2008). It is best used in combination with other agents (Swindle MM 2015). Ketamine (20 mg/kg) with xylazine (2 mg/kg) has been recommended as a general anaesthetic protocol in swine for short procedures (Riebold TW et al. 1995). The addition of another analgesic agent and/or general anaesthetic is necessary to perform invasive surgery (Snjezana Golubovi 2009).

Propofol is a sedative hypnotic agent used for induction of anesthesia in pigs due to its rapid onset of action and short elimination time. It can also be used in coinduction protocols with midazolam or ketamine. Doses of 4-5 mg propofol / kg IV in pigs that were mildly to moderately sedated and 1-3 mg propofol / kg IV in deeply sedated pigs have been used. It does not have analgesic properties and produces cardiorespiratory depression .

Fentanyl is a short acting, full μ opioid agonist. It provides analgesia, but has respiratory and cardiovascular depressant effects (increased $PaCO_2$, bradycardia), reduces gastro-intestinal motility and patients can develop tolerance or hyperalgesia following its administration especialy at high doses (Simones et al. 2016). Intravenous administration of fentanyl at 5-30 μg/kg/h in combination with paralytic agents, ketamine, and nitrous oxide has been used successfully in cardiac surgery in swine (Pascal et al. 2015).

Benzodiazepine tranquilizers are effective sedatives and central acting miorelaxants, with minimal to negligible effect on the cardiovascular and respiratory functions. Midazolam has effects lasting less than 30 minutes and may be used on a daily basis for procedures requiring sedation (Lacoste et al. 2000).

Lidocaine, a sodium channel blocker used as local anaesthetic, can be administered as a CRI to provide analgesia (Valverde et al. 2004). It causes vasodilation, has antiarrhythmic effects and gastrointestinal prokinetic properties.

The goal of this study was to evaluate a partial intravenous anaesthetic (PIVA) protocol for Landrace pigs that would yield analgesia and miorelaxation for laparoscopic techniques.

MATERIALS AND METHODS

The study was carried out during several training session for surgeons on laparoscopic techniques between 2013 and 2015. The study received ethical approval from the Ethical committee of the University of Medicine and Pharmacology Carol Davila of Bucharest.

Sixteen pigs (weight between 15 and 30 kg, with a mean weight of 21 kg) were anaesthetized for the procedures. They were housed together 24h before the procedures with free access to water and food. Food was withheld 12h before the anaesthesia started. To minimise the stress, animals were premedicated prior to being transfered to the operating room. Animals arrived sedated and were carefully protected against hypothermia by insulating with warm water filled mattress. All the patients were premedicated with xylazine 2 mg/kg and ketamine 20 mg/kg im (in the neck area). Once the pig was profoundly sedated, vascular access was secured by placing a catheter 20 – 22G in the auricular veins. All patients were preoxygenated before induction for minimum 2 minutes. Induction was done with propofol 4 mg/kg IV up to effect. Larynx was sprayed with lidocaine 2% in order to reduce the incidence of laryngospasm (ketamine maintains this reflex). Once orotracheal intubation was achieved in sternal recumbency, the PVC endotracheal tube was connected to a circle breathing system and anaesthesia was maintained with isoflurane 1.2–1.5% in oxygen. Manual ventilation was provided when dyspnea or apnea were noted. All animals had an an intravenous infusion of Ringer lactate set at 5 ml/kg/h started immediately after the orotracheal intubation.

For the laparoscopic technique, abdominal insufflation of carbon dioxide is mandatory in order to allow proper visualization and manipulation of body tissues and instruments (Fig 1). Abdominal pressure of insufflated carbon dioxide was maintained at 6-8 mmHg.

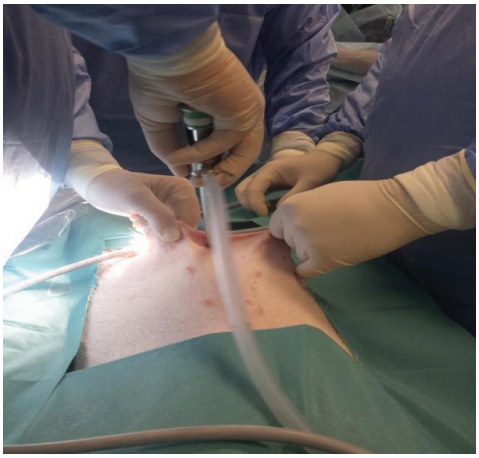

Figure 1. Abdominal insufflation with carbon dioxide

Pigs were randomly allocated to one of the following groups: MFK group (receiving midazolam 0.2 mg/kg/h, fentanyl 10 µg/kg/h and ketamine 10 µg/kg/min) (n=8) or KL group (receiving ketamine 10 µg/kg/min and lidocaine 30 µg/kg/min) (n=8).
Heart rate, respiratory rate, oscillometric blood pressure, rectal temperature and saturation of oxygen were monitored throughout the anaesthesia and recorded every 5 minutes using a vital signs monitor (Mindray MEC-1200 VET®). (Fig.2)

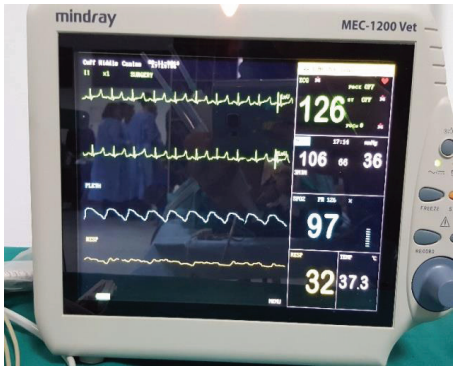

Figure 2. Vital signs monitor Mindray MEC-1200 VET®

Rescue analgesia was provided with ketamine 5 mg/kg IV if heart rate and systolic blood pressure increased by 20% compared to the baseline.
The surgeons were asked to assess subjectively the degree of miorelaxation during the procedure using a 1 to 5 scale were 1 was no miorelaxation and 5 – maximum degree of miorelaxation.
The duration of anaesthesia ranged between 210 and 240 minutes.
All patients were euthanized at the end of the procedures using 7.5% potassium chloride.
Statistical analysis used a one way ANOVA and the results are expressed in mean ± standard deviation.
A value of $p < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSIONS

Administration of ketamine 20 mg/kg and xylazine 2 mg/kg resulted in adequate sedation of the pigs. Spontaneous breathing was maintained. Vascular access was obtained easily in the auricular vein. Pigs were transported from the housing den to the experimental lab without incidents. This was a 5 minutes distance trip. The ketamine doses reported previously for swine cover a wide interval (11 – 33 mg/kg IV or IM). We chose an intermediate dose taking into account the addition of an α2 agonist that provides sedation itself (xylazine).
After the bolus injection of propofol, followed by lidocaine sprayed on the larynx, laryngeal and pharyngeal reflexes disappeared, and all pigs were intubated without noteworthy complications. Orotracheal intubation was performed with the pig positioned in sternal recumbency, using a stylet and only after a minimum of 2 minutes facial mask preoxygenation (Fig 3).
No complications were encountered for this stage of the protocol and the intubation was achieved in less than 1 minute for all the patients.
This comes into agreement with the results reported by Theisen et al. (2009) that recommended the ventrodorsal position as the first choice for providing a smooth and fast airway in laboratory pigs.

Figure 3. Orotracheal intubation

Mean values for oscillometric blood pressure, heart rate, respiratory rate, rectal temperature and saturation of oxygen are presented in table 1.

Table 1. Mean values of physiological parameters monitored during anaesthesia for the MFK and LK groups and p values calculated between the two groups

Parameter	MFK group	LK group	P value
Sistolic blood pressure (mm Hg)	116.6	98.3	0.007
Mean blood pressure (mmHg)	82	75.6	0.055
Diastolic blood pressure (mm Hg)	61.3	52.3	0.078
Heart rate (bpm)	75.6	95.3	0.022
Respiratory rate (rpm)	5	9	0.017
Temperature (^{o}C)	35.2	35.7	0.63
Saturation of oxygen (%)	98.3	97.5	0.74

There was a significant statistical difference between the two groups for the heart rate and sistolic blood pressure (see table 1).
This parameters are commonly used as indicators of nociception during general anaesthesia. At the same time, we must not forget that they can both be influenced by the drugs administered during the anaesthetic protocol: fentanyl CRI induces bradycardia, while ketamine, midazolam and lidocaine maintain heart rate. The higher heart rate for the LK group should not be interpreted as a sign of nociception for the duration of the whole anaesthesia as this is the mean value calculated for the 210 – 240 minutes of surgical procedure. Instead, the trend in this parameters was monitored. Rescue analgesia was provided when there was a 20% increase in one of the two parameters mentioned above. No gross movement was recorded as response to surgical stimulation. The pigs in group MFK received 9 doses of rescue analgesia, while group LK received 15 ($p = 0.031$). The significant statistical difference in the requirement for rescue analgesia allows us to conclude that MFK provided better analgesia for this type of surgical procedure.
There were no significant statistical difference between the two groups regarding mean blood pressure, diastolic blood pressure, oxygen saturation and temperature values. Episodes of hypotension appeared equally in the two groups, most of the times correlated with surgical maneuvers that elicited vagal reflex. Treatment was cesation of the maneuver for a couple of minutes and if that was not enough to rstore blood pressure values, HES 6% 10 ml/kg in 10 minutes was administered. The colloid boluses were repeated as needed in order to restore a mean blood pressure over 60 mmHg.

Intermittent positive pressure ventilation was needed in 6 animals from the MFK group and in 2 animals in the LK group. This was probably due to depressant effect of fentanyl on the respiratory center, but this effects can be overcome by supplementing ventilation of the patients.
Fentanyl CRI alone or in combination with ketamine and lidocaine has been shown to have a sparing effect on the minimal alveolar concentration (MAC) of isoflurane in dogs (Aguado et al. 2011; Simoes et al. 2016). It is reasonable to assume that the same effect will be seen in swine. Kleine et al. (2015) looked at the effect of midazolam on isoflurane MAC in pigs. They found no effect of midazolam on the isoflurane MAC, but in their study midazolam was administered as a single dose and not as a continuous rate infusion. Lidocaine has a significant sparing effect on the isoflurane MAC in dogs only at higher infusion rates (100 µg/kg/min) than the one we used in our protocol (Aguado et al. 2011). We assume that for the protocols we used the major effects on

MAC would be due to fentanyl and ketamine. We did not measure the MAC in our patients, but we monitored the clinical responses to surgical stimulation and correlated them with the vaporizer setting. For the MFK group, the mean setting for the isoflurane vaporizer was 1.2% and this allowed surgery without increase in heart rate and systolic blood pressure. For the KL group, the mean value setting for the isoflurane vaporizer was 1.4%. There was a strong statistical difference between the 2 groups ($p < 0.001$). This confirms our hypothesis and comes in agreement with the studies mentioned above.

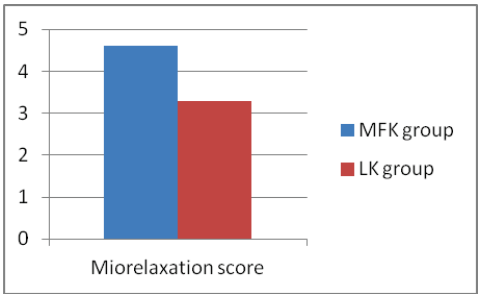

Figure 4. Miorelaxation scores subjectively assessed by the surgeons.

CRI doses for midazolam in pigs were previously reported at 1.14 to 2.71 mg/kg/h. The doses used in this study was reduced to a fifth of the minimal reported one, while ketamine and fentanyl were used at the common previously reported infusion rates (Kaiser et al., 2007). Still , MFK provided good miorelaxation (miorelaxation score 4.7) (Fig.4.). There was a significant statistical difference between the two groups ($p = 0.034$), still both protocols ensured good conditions for surgery. Other studies reported similar results, even after single intramuscular administration of midazolam-ketamine (Clutton et al. 1997).

Our study has several limitations. One is the use of oscillometric blood pressure measurement instead of the invasive arterial measurement that is considered the golden standard for this parameter. This was due to the lack of equipment in the laboratory where the study was conducted. Still, the method is accepted for routine monitoring of the blood pressure and physiological range values were determined for some breeds of swine, including after sedation with midazolam (Goodrich et al. 2001).

The second limitation of our study is the euthanasia of the patients at the end of the surgeries. This did not allow us to assess the quality of recovery and the possible occurrence of life-threatening events at this time. The recovery period is an essential time frame for any anesthesia and patient, also for experimental patients as this can interfere with the results of the experimental protocol and can introduce statistical errors in the results.

The third limitation of our study was the lack of monitoring of the end-tidal expired carbon dioxide ($EtCO_2$). This is required in order to monitor the ventilation of the patient during general anaesthesia. It is used also to indirectly monitor the cardiac function as CO_2 levels are positively correlated to the cardiac output. Also, due to abdominal insufflation of CO_2 during laparoscopic technique, hypercapnia can be encountered. A solution to this would have been the arterial blood gas analysis, but that was also not possible due to the same technical limitations of the laboratory used for the surgical training sessions.

CONCLUSIONS

MFK provided better analgesia and increased miorelaxation during laparoscopic cholecistectomy, while LK provided a better stability in terms of ventilation. Both protocols can be used in swine and ensure stable cardiovascular parameters during general anaesthesia in this species.

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