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Original Research Paper

SURGICAL INTRAVENOUS CATHETERISATION OF PIG

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Summary: For the single daily blood samples collection in swine blood venipuncture of the anterior caval vein is the most common method. If more frequent blood samples are needed then repeated venipuncture is less desirable, as it may cause damage to blood vessels and it could also influence the experimental results. Current methods utilized are therefore not suitable for frequent sampling due to the stress to which the pigs are exposed. To avoid the above-mentioned problems, several methods of catheterisation have been developed to allow for repeated sampling. In our experiment, an intravenous catheter was inserted into the jugular vein of a pig under general anaesthesia. No antibiotics were used perioperatively. Behaviour, appetite and body temperature were monitored twice daily. Blood samples were also taken twice daily for 5 days and were submitted to haematological and biochemical examinations. The values were compared to the results obtained from the samples, taken before the cannulation. Data were processed with a descriptive statistical method. According to the pig's behaviour, the animal did not display any clinical indications of pain or stress, provoked by the catheter. The catheter remained patent and on its primary position for the whole time of the experiment that was confirmed by an X-ray examination. The correct surgical intravenous cannnulation did not provoke any systemic inflammation process. Haematological and biochemical parameters of blood were not influenced by the catheterisation.

Key words: blood specimen collection; catheterization; *v. jugularis*; hematologic tests; swine

Introduction

Several techniques for the collection of swine blood are in current use. Venipuncture of the anterior caval vein is the most common method for obtaining blood samples, although other methods are used (1). If single daily blood samples are required, then jugular venipuncture is considered an acceptable collection protocol. If more frequent sampling is needed to asses various biomedical parameters (eg. pulsatile hormone secretion or hormonal responses to various secretagogues) in a conscious animal, then repeated venipuncture is less desirable as it increases stress and could potentially jeopardize the well-being of the animal (2). Beside it may also influence the experimental results. Moreover, it may cause damage to blood vessels, especially when frequent sampling is done (1). To avoid the above-mentioned problems, several methods of catheterisation have been developed for repeated blood sampling in pigs (3). They can involve surgical procedures under general anaesthesia (1, 3) or non-surgical cannulation of anesthetised pigs (2) or of pigs, restrained in the standing position with the use of a nylon or cotton snare placed around the snout (4, 5).

Sometimes the need to undertake biomedical research on conscious pigs also appears at our institute. The aim of our work was to insert a long-term intravascular catheter into the jugular vein of a pig without the use of antibiotics and to monitore its influence on some blood parameters.

Material and methods

A castrated male commercial slaughter pig cross of 35 kg was used in the experiment. It was stabled at the university´s pen 3 days prior to surgery to adapt to a new environment. The animal was allowed drinking water ad libitum and was fed 1 kg of a commercial feed twice daily.

Prior to the catheterisation procedure, blood was taken from vena cava cranialis to assess haematological and biochemical parameters. The investigated haematological parameters included red blood

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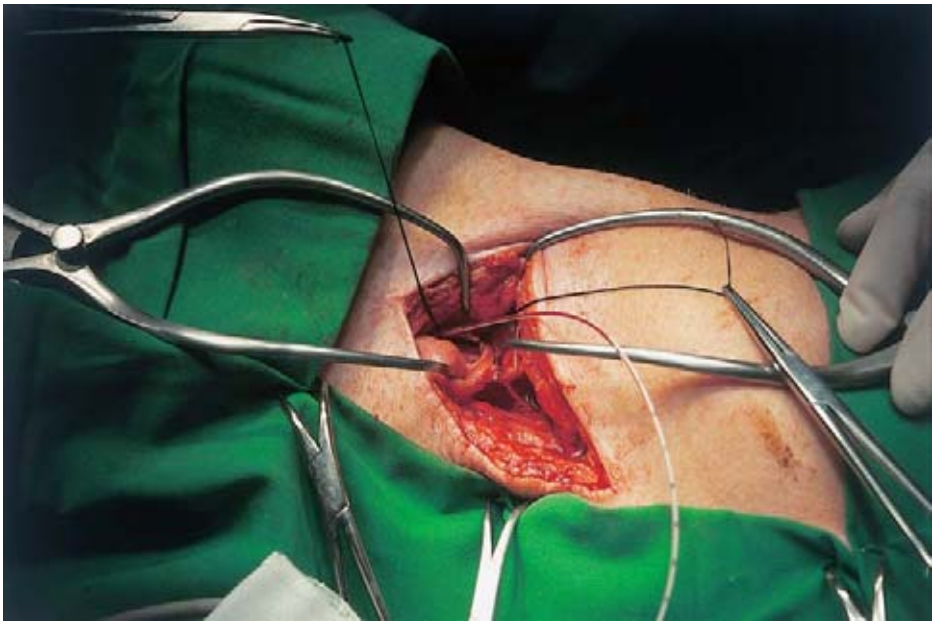

Figure 1: Cannulation of the jugular vein in pig

cells (RBC), haemoglobin (Hb), mean corpuscular volume (MCV), haematocrit (Ht), white blood cells (WBC), trombocytes (Trom.), neutrophils (Neu.), eosinophils (Eos.), basophils (Baso.), lymphocytes (Limp.), band neutrophils (BN) and monocytes (Mon.). The investigated biochemical paramethers were: bilirubin (Bil.), alanine aminotransferase (ALT), urea, iron (Fe), gama glutamyltransferase (GGT), proteins (Pro.), glutamate dehydrogenase (GLDH), potassium (K), magnesium (Mg), phosphorus (P) and creatine kinase (CK).

The investigated haematological parameters were analysed with analyser ABC VET installed at the laboratory of the Clinic for ruminants of the Veterinary faculty.

The investigated biochemical parameters were analysed with analyser COBAS MIRA.

After a 24-hour period without feed and 12 hours without water, the animal was premedicated intramuscularly with midazolame (0.15 mg/kg, Dormicum, Roche), ketamine (10 mg/kg, Bioketan, Vetoquinol) and xylazine (0.4 mg/kg, Chanazine 2%, Chanelle Pharmaceuticals). Anaesthesia was induced by intravenous administration of propofol (1-2 mg/kg, Diprivan 10mg/ml, Zeneca Pharmaceuticals Ltd.) and then maintained also by propofol (10 mg/kg/h, Diprivan 10 mg/ml, Zeneca Pharmaceuticals Ltd.). Analgesia was included by morphine (0.2 mg/kg, Morphini Hydrochloridum, 20 mg/ml, Alkaloid Skopje) and butorphanol (0.3 mg/kg, Torbugesic, Fort Dodge Animal Health).

The surgical area of the neck was shaved, disinfected and draped. Under sterile conditions a left jugular sulcus was exposed between musculus brachiocephalicus and m. sternomastoideus, near the prescapular part of the pectoral muscle so that the left external jugular vein was reached. After the vein was lifted, two vascular tapes were placed around it 3 cm apart. The jugular catheter Intramedicut 2 (16G, Sherwood medical) was inserted 10 cm proximally. Two silk ligatures were placed around the vein. Distally from the place of the insertion, the vein was ligated (Softsilk 2/0, Autosuture), while proximally the vein was ligated over the catheter (Softsilk 4/0, Autosuture) so that the catheter was fixed in the desired position. The patency of the catheter was confirmed with the application of heparinised saline (2 I.U. of heparin (Heparin razt. za inj., 25000 I.U./5 ml, Krka) in 1 ml of saline (0.9% NaCl, Braun)). The subcutis on the left neck area was chanelled dorsally. The catheter was looped in the subcutaneous plane in order to provide additional lenght for neck movements. The rest of the catheter was led externally to the back of the neck where it was fixed with silk (Softsilk 4/0, Autosuture). The neck was taped with selfgripping plaster (Vivapore 15 x 10 cm, Tosama). The incision was closed routinely; subcutis with continuous sutures, using absorbable, synthetic multifilament material (Polysorb 2/0, Autosuture), and cutis with single interrupted sutures, using silk (Softsilk 2/0, Autosuture). No antibiotics were administered.

Blood samples were collected during a 5-day period after cannulation. They were taken twice

Table 1: Results of biochemical blood examination

Day	Bil. µmol /L	ALT U/L	Urea mmol/L	Fe µmol /L	GGT U/L	Pro. g/L	Albumin g/L	GLDH U/L	K mmol/L	Mg mmol/L	P mmol/L	CK U/L
0*	2.81	46	4.68	20.1	35	64.1	25.4	1.07	4.70	0.88	2.84	502
0(3pm)	3.33	43	2.46	6.1	20	60.8	24.7	1.16	4.41	0.78	3.09	725
1(8am)	1.97	51	4.70	20.3	27	68.6	28.8	1.51	4.47	0.91	3.08	1514
1(3pm)	2.17	50	4.27	14.1	22	68.1	28.5	1.03	4.49	0.85	2.84	1419
2(8am)	2.13	45	4.41	16.7	36	64.7	26.9	1.40	5.56	0.97	3.23	1089
2(3pm)	2.05	42	2.98	13.7	35	64.7	27.2	1.25	4.43	1.04	3.28	729
3(8am)	1.77	38	3.36	23.7	37	61.7	26.0	1.22	4.92	0.97	2.86	408
3(3pm)	1.93	37	3.94	13.7	38	63.2	27.1	1.42	4.95	0.97	3.06	453
4(8am)	1.28	38	4.09	23.6	24	68.0	28.6	1.68	4.87	0.87	2.92	281
4(3pm)	0.8	37	4.27	16.4	59	64.9	27.2	3.44	5.79	1.16	4.25	312
5(8am)	1.44	31	3.85	26.0	26	62.6	25.6	1.21	4.88	0.90	2.52	239
5(3pm)	0.84	33	3.59	26.0	26	66.0	27.3	2.70	4.61	0.88	2.48	246

daily, at 8 am and at 3 pm. Two syringes were used each time. The first one was used to remove the blood, mixed with heparine, from the catheter, while the second one was used to collect the blood sample. The mixture of blood and heparine from the first syringe was then returned back to the catheter and 1 ml of heparinised saline was added. The blood samples were subjected to the same examinations as the samples, taken before the cannulation. The pig´s behaviour, appetite and body temperature were also monitored twice daily.

Data were processed with a descriptive statistical method. Based on 11 results, the mean value ± and the standard deviation were calculated for each blood parameter and the data were compared with the values, gain from the samples that were taken before the catheterisation.

On day six after catheterisation, the animal was sedated with xylazine (0.4 mg/kg) and ketamine (10 mg/kg) and the position of the catheter was checked with X-ray examination. Then euthanasia was performed by a lethal dose of euthanasia solution (T-61, Hoechst Roussel) via the indwelling catheter.

The procedure was approved by the Veterinary Administration of the Republic of Slovenia, number 323-02-16/2003, date 10.2.2003.

Results

Results of biochemical blood examination before cannulation (0*), on the day of cannulation (day 0 at 3 pm), and twice daily on days 1, 2, 3, 4 and 5 after cannulation. The time of blood sampling is given in parenthesis.

Orientational normal biochemical values in pigs, enzyme activity measured at working temperature 37° C (6):

Bil.: up to 5.64 µmol /L
CK: 50 - 500 U/L
ALT: 31 - 58 U/L
Urea: 2,33 - 6.66 mmol/L
Fe: 18,0 - 35.0 µmol /L
P: 1,81 - 3.19 mmol/L
K: 4,5 - 6.2 mmol/L
GGT: 32 - 58 U/L
Pro.: 62.0 - 82.0 g/L
Albumin: 30.0 - 40.0 g/L
GLDH: up to 1.86 U/L
Mg: 0.83 - 1.42 mmol/L

Results of haematological blood examination before cannulation (0*), on the day of cannulation (day 0 at 3 pm), and twice daily on days 1, 2, 3, 4 and 5 after cannulation. The time of blood sampling is given in parenthesis.

Orientational normal haematological values in pigs (6):

RBC: 5,0 - 8.0 x1012/L
Hb: 10,0 - 15.5 g/dl
Neu.: 28 - 52%
Limp.: 40 - 64%

Table 2: Results of haematological blood examination

Day	RBC 10^{12}/L	Hb g/dl	MCV %	Ht %	WBC 10^9/L	Trom. 10^9/L	Neu. %	Eos. %	Baso. %	Limp. %	BN %	Mon. %
0*	6.12	10.1	57	34.7	24.4	329	44	2	0	54	0	0
0(3pm)	4.62	7.6	55	25.7	15.0	272	54	0	0	46	0	0
1(8am)	5.88	9.8	57	33.3	26.3	252	44	0	0	54	0	2
1(3pm)	5.78	9.6	56	32.3	24.5	260	43	2	0	53	0	2
2(8am)	5.45	8.8	59	32.0	22.8	198	40	12	0	48	0	0
2(3pm)	5.42	8.8	58	31.5	20.4	218	57	4	0	39	0	0
3(8am)	5.14	8.3	56	28.7	19.1	242	34	8	1	57	0	0
3(3pm)	5.15	8.4	56	28.8	20.0	262	36	2	0	62	0	0
4(8am)	5.41	8.7	56	30.4	20.6	284	49	2	1	47	0	1
4(3pm)	4.95	8.1	57	28.1	25.3	260	75	2	0	23	0	0
5(8am)	4.92	8.2	56	27.8	19.1	302	27	4	0	69	0	0
5(3pm)	4.69	7.8	56	26.3	18.4	282	45	2	0	53	0	0

WBC: 10.0 - 20.0 x109/L
Ht: 32 - 47%
MCV: 32 - 47%
Trom.: 250 - 600 x109/L
Mon.: 2 - 8%
Baso.: 0 - 2%
Eos.: 1 - 8%
BN: 0 - 4%

During the trial the pig behaved normally, having a good appetite, and had normal body temperature. No clinical manifestations of pain or stress due to the catheter or repeated blood collections were noticed. The catheter remained on its primary position and patent for the whole time of the experiment.

Discussion

Intravenous catheterisation of the pig was performed under general anaesthesia due to the fact that the cannulation of the jugular vein with the use of classical means of restraint can only be done in 15 to 20 minutes (5). This represents 15 to 20 minutes of stress for the animal and can lead to the alteration of some blood parameters. Conscious pigs, restrained by a snare, may interrupt cannulation efforts by making undesirable movements. It could also prove more hazardous for the investigators than working with anesthetized animals (4).

According to the pig's behaviour, the animal did not appear to be stressed by the catheter. Irritation or itching in the neck area were not evident. The catheter remained on its primary position for the whole time of the experiment, what was confirmed by the X-ray examination prior to the euthanasia of the animal. The fixation of the catheter did not have to be too tight and had to allow normal passage of the blood so that the blood collection was possible. The vein and the catheter remained patent for all 5 days of the experiment. The blockage of the vein or the obstruction of the catheter due to the thrombosis and phlebitis were reported (1).

The correct surgical intravenous cannnulation did not provoke any systemic inflammation process that was confirmed by monitoring blood parameters. The investigated blood parameters, with the exception of L and CK, were within the limits of the reference values. Initially elevated values of L and CK were at the end of the experiment within the limits of the reference values. Although the values of the investigated blood parameters were, according to the initial values, changing during the experiment (Tables 1 and 2), their mean values, considering standard deviation, did not deviate from the borders of the ref-

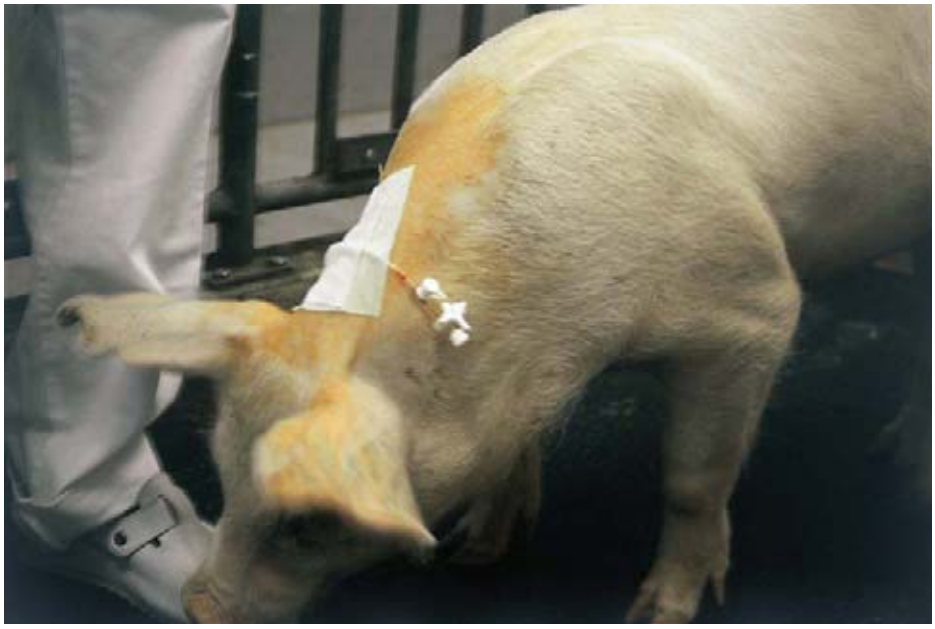

Figure 2: Pig with the intravenous catheter, two days after the cannulation.

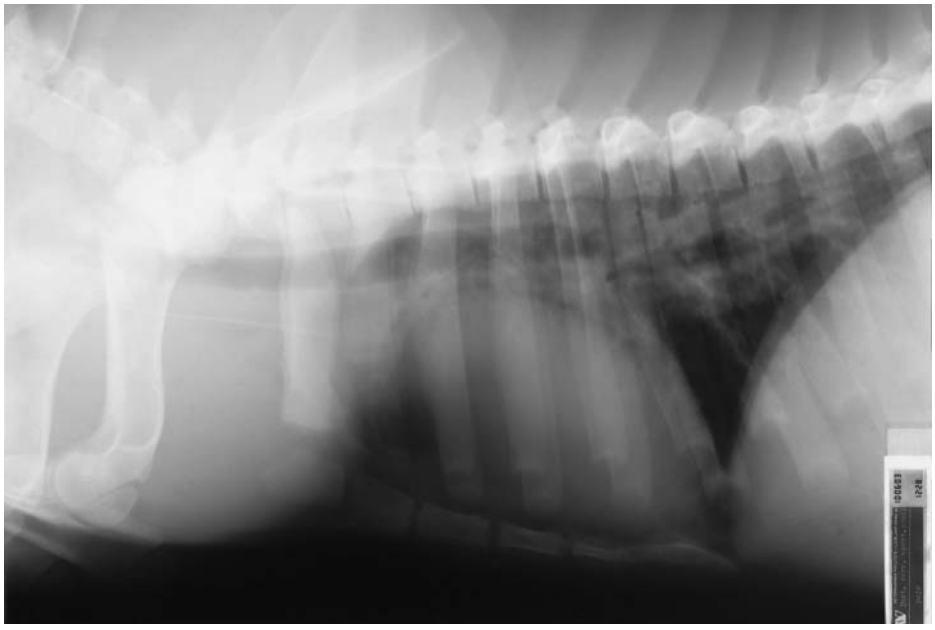

Figure 3: X-ray conformation of the intravenous catheter's position.

erence values. Pijpers et al.(1) reported the changes in rectal temperature, WBC counts, serum Zn and Fe levels, that may provide essential information about the reactions of defence mechanisms against infections. These effects of surgery and anaesthesia did not last longer then two days.

In our experiment, catheterisation did not have influence on the haematological and biochemical parameters of blood, however an adequate sample of animals should be tested in order to confirm these results.

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KIRURŠKO VSTAVLJANJE VENSKEGA KATETRA PRAŠIČU

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Povzetek: Pri enkratnem odvzemu krvi prašičem se le-ta najpogosteje jemlje iz sprednje velike dovodnice (vena cava cranialis) z iglo. Pri pogostejšem jemanju pa ta način ni primeren, saj lahko povzroči poškodbe krvnih žil in vpliva na rezultate preiskav. Poleg omenjenega so metode, ki jih navadno uporabljamo za odvzem krvi prašičem, za večkratno jemanje v kratkem času neprimerne zaradi stresa, ki so mu živali pri tem izpostavljene. Da bi se izognili tem težavam, so razvili različne metode kateterizacije. V našem poskusu smo prašiču v splošni anesteziji vstavili intravenski kateter v jugularno veno. Antibiotikov nismo uporabljali. Po vstavitvi katetra smo dvakrat dnevno spremljali obnašanje, ješčnost in telesno temperaturo prašiča. Pet dni smo dvakrat dnevno prašiču jemali kri in opravili hematološke in biokemične preiskave. Dobljene vrednosti smo primerjali z vrednostmi hematoloških in biokemičnih preiskav krvi, odvzete pred vstavitvijo katetra. Podatke smo obdelali s pomočjo deskriptivne statistične metode. Po obnašanju prašiča smo sklepali, da kateter zanj ni bil moteč. Ves čas poskusa je kateter ostal prehoden in na mestu, kjer smo ga vstavili, kar smo potrdili z rentgenskim slikanjem prašiča. Kirurško vstavljanje katetra ni povzročilo sistemskega vnetnega procesa. Kateterizacija ni vplivala na vrednosti biokemičnih in hematoloških parametrov.

Ključne besede: krvni vzorec, zbiranje; kateterizacija; *vena jugularis*; hematološki testi; prašiči