

Computed tomography in diagnosing pituitary tumours in rats – the authors' own observations

PIOTR DĘBIAK, ANNA WILCZYŃSKA*, JERZY ZIĘTEK*,
ALICJA WÓJCIK*, STANISŁAW WINIARCZYK*, ŁUKASZ ADASZEK*

Department and Clinic of Animal Surgery, Laboratory of Radiology and Ultrasonography,
*Department of Epizootiology and Clinic of Infectious Diseases, Faculty of Veterinary Medicine,
University of Life Sciences in Lublin, Głęboka 30, 20-612 Lublin, Poland

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Dębiak P., Wilczyńska A., Ziętek J., Wójcik A., Winiarczyk S., Adaszek Ł.

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Summary

The aim of this study was to present cases of rats with suspected pituitary tumours and the application of computed tomography in diagnosing pituitary tumours in rats. The study included 25 animals with symptoms suggestive of pituitary tumours (ataxia, paralyse, strabismus). The animals were submitted to a detailed clinical examination and computed tomography (CT). The CT confirmed pituitary tumours in 20 out of 25 animals. Additionally, this study enabled the proof of a definite correlation between the size of the pituitary tumour and clinical symptoms observed in sick animals. This article presents the usefulness of tomography examination in diagnosing pituitary tumours in rats and it is the first paper of this kind in Poland. However, it should be borne in mind that a CT without the use of a contrast medium only allows for the identification of neoplasm in some cases. It is the contrast-computed tomography that significantly increases diagnostic chances of this method in identification and evaluation of the size, shape and contour of proliferative lesions of the brain region in rats.

Keywords: rats, pituitary tumours, computed tomography

Rats are more frequently kept by humans as companion pets. As patients of veterinary practices, they can receive medical care including preventative and therapeutic treatment. Continuous advancement of veterinary techniques and knowledge of diagnostic methods of imaging in these patients improves the process of their diagnosis and treatment, including treatment of neurological conditions.

Pituitary tumours in rats are mostly hormonally active (functioning) tumours, although sporadic cases of non-hormonally active (non-functioning) tumours may occur, which give no clinical symptoms throughout the animal's entire life. Functioning pituitary tumours grow very quickly, which is accompanied by dynamic development of clinical symptoms. Most commonly these neoplasms are diagnosed in females at the age of above 18 months, especially in unsterilised ones but they may also be found in males (15). Tumour growth depends on many factors, such as genetic preconditions, inbreeding or exposing animals to carcinogens (17, 18).

Functional pituitary tumours can secrete all hormones that are normally produced by this gland. In

the case of the most commonly diagnosed pituitary tumour – prolactinoma – the secretion of prolactin (PRL) increases. The second most prevalent is the tumour that secretes somatotropin (GH). Other types of pituitary tumours secreting hormones such as ACTH, TSH, FSH and LH are rarely found in rats, while the secretion activity may concern several of the above hormones or only one of them (18).

The nature of clinical symptoms observed in pituitary patients is strictly connected with tumour growth (15). In most patients the symptoms develop gradually but there are also cases with a sudden onset of symptoms and differential diagnosis should take into account middle ear infections, strokes, cranial injury or poisoning (9).

The exacerbation of clinical symptoms is related with the tumour mass whose growth exerts pressure on surrounding brain structures causing neurological disorders. For example, the tumour pressing on the optic chiasm causes visual impairment or loss of vision and the pressure on the brain stem causes dysphagia or respiratory problems. When pressure is placed on the cerebellum, ataxia may be observed while the pressure

on the hypothalamus most frequently causes impaired ingestion (3).

One of the most characteristic symptoms observed in pituitary tumours is tilting of the head (*torticollis*), weakening of the limbs and ingestion problems which are caused by weakened masseter muscles (3). Additionally, a circling movement or walking in circles is observed. Nystagmus and apathy are characteristic symptoms of an advanced stage tumour diagnosed at a late phase (5, 7).

The therapy for the most common pituitary tumours in rats – prolactinoma – consists of palliative treatment involving the administration of dopamine D2 receptor agonist such as bromocriptine or prolactin inhibitors such as cabergoline (2, 6).

The aim of this paper was to present case studies of 25 rats with suspected pituitary tumours.

Material and methods

Animals. The study included 25 rats (12 males and 13 females) that were the patients of the Clinic of Infectious Diseases of the Faculty of Veterinary Medicine of the University of Life Sciences in Lublin showing clinical symptoms suggestive of pituitary tumour (Tab. 1). The average age of the animals was 23.2 months (males 22 months,

females 23 months). The average body weight was 377 g (males 422,5 g, females 335 g). These rats had been kept as domestic animals and fed with a standard feed for rodents. They had no contact with other animals, irritants, or toxins.

Clinical examination. A general assessment of the condition of the animals used in the study was performed on the basis of clinical observations and data obtained during an interview. The animals were brought to veterinary practices with various problems, such as loss of appetite, movement disorders, paresis of the limbs and nystagmus (Tab. 1). Blood for biochemical and haematological analyses was collected from all rats. The haematological examination was performed using an Exigo Boule analyser (Sweden), whereas the biochemical examination was conducted using a Mindray BS-300 analyser (Poland).

All the animals were subjected to clinical examination including a simple neurological examination involving a wheelbarrow test and wrist flexion test. The wheelbarrow test was performed by raising the back of the rat's body together with its pelvic limbs so that the animal could only rest on its thoracic limbs. Next, the rats had to take a short walk on the examination table during which the movement of their thoracic limbs was observed. Normally the rat should rest on these limbs with confidence and move according to the examiner's intention. This is described as a positive result. The wrist flexion test was performed by

Tab. 1. Description of animals used in the study and their clinical signs and symptoms

Animal no.	Gender	Age	Body weight	Clinical symptoms observed	Presence of a pituitary tumour
1.	♂	1.5 years	400 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
2.	♀	2 years	350 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
3.	♀	1.5 years	400 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
4.	♀	2 years	320 g	Apathy, forelimb weakness, circling movements	confirmed
5.	♀	2 years	250 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
6.	♀	2 years	430 g	Apathy, forelimb weakness, circling movements	confirmed
7.	♂	2.5 years	620 g	Apathy, forelimb weakness, circling movements	confirmed
8.	♂	2.5 years	360 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
9.	♀	2.5 years	370 g	Apathy, circling movements, nystagmus	none
10.	♀	1.5 years	390 g	Apathy, forelimb weakness, circling movements	confirmed
11.	♂	2.5 years	420 g	Apathy, forelimb weakness, circling movements, CRD	confirmed
12.	♀	1.5 years	210 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
13.	♀	3 years	320 g	Apathy, circling movements, nystagmus	none
14.	♂	2 years	400 g	Apathy, forelimb weakness, circling movements	confirmed
15.	♂	1.5 years	350 g	Apathy, forelimb weakness, circling movements	confirmed
16.	♂	1.5 years	700 g	Apathy, forelimb weakness, circling movements	confirmed
17.	♂	6 months	280 g	Apathy, hindlimb weakness, circling movements, CRD	none
18.	♀	2 years	370 g	Apathy, forelimb weakness, circling movements, nystagmus	confirmed
19.	♀	2 years	210 g	Apathy, forelimb weakness, circling movements, CRD	confirmed
20.	♂	3 years	330 g	Apathy, forelimb weakness, circling movements	confirmed
21.	♂	9 months	350 g	Apathy, forelimb weakness, circling movements, CRD	confirmed
22.	♀	9 months	360 g	Apathy, hindlimb weakness, circling movements, nystagmus	none
23.	♀	2 years	380 g	Apathy, forelimb weakness, circling movements, pyometra	confirmed
24.	♂	2 years	400 g	Apathy, circling movements, otitis media, pyometra	none
25.	♂	1.5 years	460 g	Apathy, forelimb weakness, circling movements	confirmed

flexing a thoracic limb in a wrist joint so that the animal could rest the whole body on this joint. Normally the rat should quickly correct this unphysiological position of the limb, which is described as a positive wrist flexion test.

Tomography. Tomography was performed using a Philips MX 16 CTB device. Before the examination, the animals were sedated with an i.v. infusion of propofol at a dose of 0.1 mg/100 g of body weight.

With the patients positioned at a sternal recumbency, the head CT was performed in transverse planes using the Soft Tissue algorithm WL 40 WW 350 (Window Level, Window Width). The parameters of CT scanning were noted in a special examination protocol and they are as follows: 120 kV, 120 mA, slice width 1 mm, collimation 16*0.75, pitch 1, lamp rotation time 0.75 seconds. The scanning was conducted before and after intravenous administration of the medium (Omnipaque 300 mg I/ml; GE Healthcare Oslo, Norway) at a dose of 2 ml/kg of body weight.

In the evaluation of the lesions multiplanar reconstructions of images were used in transverse, sagittal and dorsal planes based on soft tissue algorithm and brain algorithm. In the tomograms obtained the signs of tumour lesion were assessed without using contrast medium, and during the contrast examination the lesions were measured, including the length, width and height, the shape and contours of the tumour edges.

Results and discussion

Clinical observation. The haematological examination revealed leucocytosis in 7 rats, anaemia in 4 rats and thrombocytopaenia in 11 rats. The biochemical examination revealed an increase of alanine aminotransferase (ALT) and alkaline phosphatase (AP) activity in 3 and 8 subjects respectively, the increase of aspartate aminotransferase (AST) activity in 4 rats and the increase of bilirubin concentration in 3 rats (Tab. 2).

Tomography results. Out of 25 subjects that underwent tomography, in 20 of them a tumour was found in the region of the pituitary gland (Tab. 1). In five other animals clinical symptoms, such as circling movement or nystagmus, were related to the middle ear inflammation (otitis media).

Based on the size of the tumour the animals were divided into three groups: rats with tumours of the size of more than 10 mm in at least one dimension (7 subjects, 35%), rats with tumours of the size of more than 8 mm in at least one dimension (6 subjects, 30%) and rats with tumours of the size of less than 8 mm in all dimensions (7 subjects, 35%).

Different clinical symptoms were found in rats with and without tumours and in rats with various sizes of tumours (Tab. 3).

Tab. 2. Haematological and biochemical test results of the rats

Animal no.	RBC × 10 ¹² /l	Hb g/dl	WBC 10 ⁹ /l	PLT 10 ⁹ /l	AST (U/l)	ALT (U/l)	ALP (U/l)	Total bilirubin (mg/dl)
1.	8.22	12.1	12.1	620	75	18	57	0.4
2.	9.34	11.3	10.2	840	92	20	62	0.2
3.	8.34	14.2	8.3	1000	84	28	75	0.6
4.	10.30	12.4	4.3	920	120	45	104	0.8
5.	8.28	14.3	6.2	720	131	63	108	0.3
6.	7.23	8.3	14.3	630	93	43	93	0.2
7.	6.45	11.2	11.1	780	85	94	63	0.2
8.	7.25	9.4	10.3	920	72	62	102	0.3
9.	11.34	11.3	8.4	1100	112	46	110	0.2
10.	8.68	12.6	6.3	1200	90	138	118	0.4
11.	7.35	13.8	8.2	630	85	58	56	0.5
12.	6.26	14.2	5.1	530	76	69	58	0.5
13.	7.58	12.6	4.3	320	108	58	105	0.3
14.	8.37	16.3	6.8	830	134	98	94	0.2
15.	9.64	18.2	7.6	620	68	26	78	0.4
16.	10.54	12.5	5.8	530	72	116	89	0.1
17.	8.23	13.2	12.4	920	80	68	45	0.2
18.	9.58	14.6	14.6	830	72	73	120	0.4
19.	6.45	16.3	16.2	980	131	47	45	0.4
20.	8.23	12.6	13.2	1200	125	68	56	0.5
21.	9.23	13.8	5.4	1100	180	87	78	0.2
22.	6.34	14.6	4.3	730	58	67	27	0.4
23.	5.36	12.9	5.8	820	86	45	111	0.2
24.	9.34	16.2	6.2	950	130	92	103	0.6
25.	8.25	14.2	12.6	860	98	38	96	0.2

Tab. 3. Analysis of clinical symptoms observed expressed in percentages

Clinical symptom	Tumour of more than 10 mm at least in one dimension (7 rats)	Tumour of more than 8 mm at least in one dimension (6 rats)	Tumour of less than 8 mm in all dimensions (7 rats)	No tumour (5 rats)
Apathy	100%	100%	100%	100%
Reduced mobility	100%	100%	100%	100%
Circling movements	100%	100%	100%	100%
Forelimbs paresis	100%	100%	14.3%	0%
Nystagmus	86%	66%	0%	60%
Negative wheelbarrow test (thoracic limbs weakness)	100%	100%	14.3%	0%
Negative wrist flexion test (no straightening of the wrist after flexion)	86%	50%	14.3%	0%

In the group of animals in which a pituitary tumour was not confirmed by CT, as opposed to animals with pituitary tumours, the function of thoracic limbs was preserved (negative wheelbarrow test) similarly to the majority (almost 86%) of rats in which CT revealed the pituitary tumour of the size of less than 8 mm in all

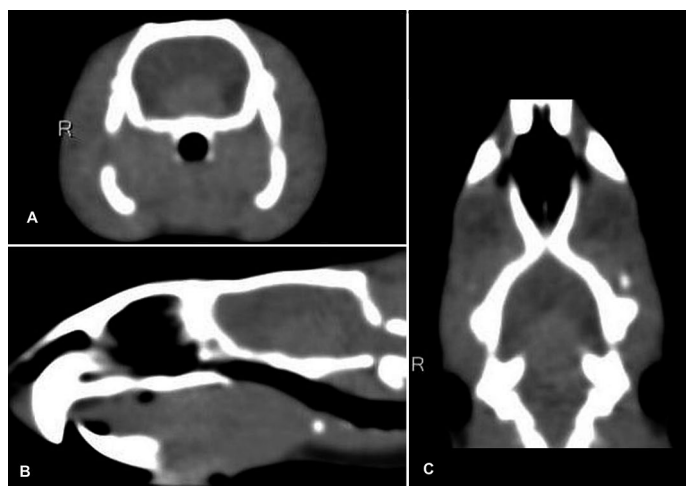


Fig. 1.1. Slightly hyperdense 50-55 HU pituitary tumour on the tomograms prior to administration of contrast in transverse plane (A) and reconstructed sagittal (B) and dorsal (C) CT images

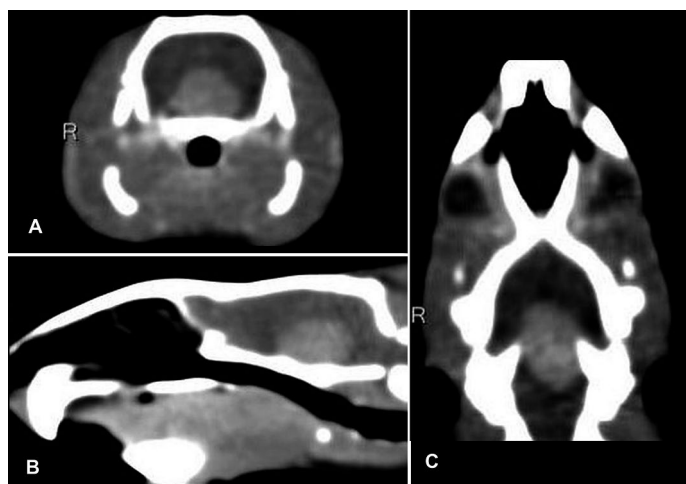


Fig 1.2. Post-contrast CT images in the same patient show a homogeneously contrast enhancing of the large pituitary tumour

dimensions. The wrist flexion test results were similar.

Neither in the rats with the pituitary tumour of the size of less than 8 mm in all dimensions or in 34% and 14% of rats with medium and large tumours respectively nystagmus was found.

In the authors' own study maximum sizes of pituitary tumour

found on tomography were 13.6 × 11.1 × 7.5 mm (length × width × height) and minimum 4.5 × 3.8 × 2 mm. It was found that at the examination stage before the contrast medium was administered the lesions in the brain region were not visible in 50% of cases (10 animals), they remained isodense to the brain tissue. In the next 10 cases tomograms revealed the presence of moderately hyperdense (up to approx. 50-55 HU) in comparison with healthy brain tissue (approx. 24-35 HU) tumour-like lesions in the pituitary region with obliterated contours (Fig. 1.1).

In the contrast examination tumour tissue was intensely enhanced up to approximately 90-120 HU in 7 cases, while in the next 3 subjects the tumour enhancement was around 89-90 HU. Normal brain tissue is enhanced up to approx. 33-40 HU after contrast administration (Fig. 1.1., Fig. 2).

It was proved that the contrast CT enabled a more detailed evaluation of the shape and contour of the tumour edges. In most cases (in 18 rats) the shape of hypodense lesions in the contrast examination was asymmetrical and irregular, while in 2 of the animals the shape of

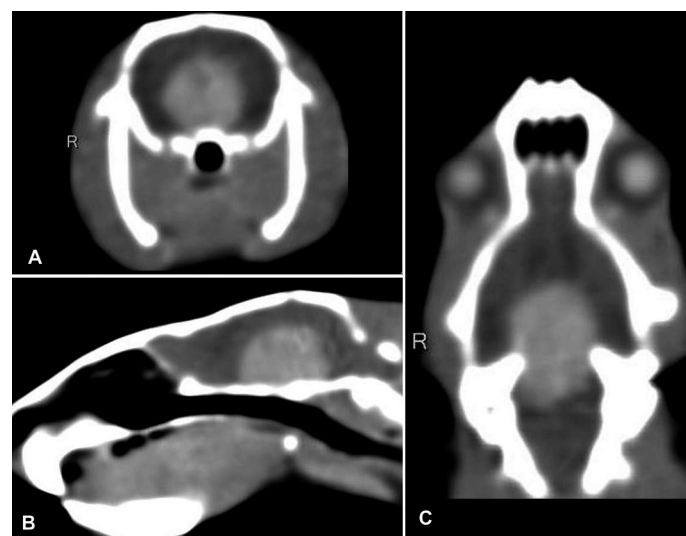


Fig. 2. Post-contrast transverse CT image (A), sagittaly (B) and dorsally (C) reconstructed images show a large, well circumscribed and homogeneously contrast enhancing (100 HU) of the pituitary tumour

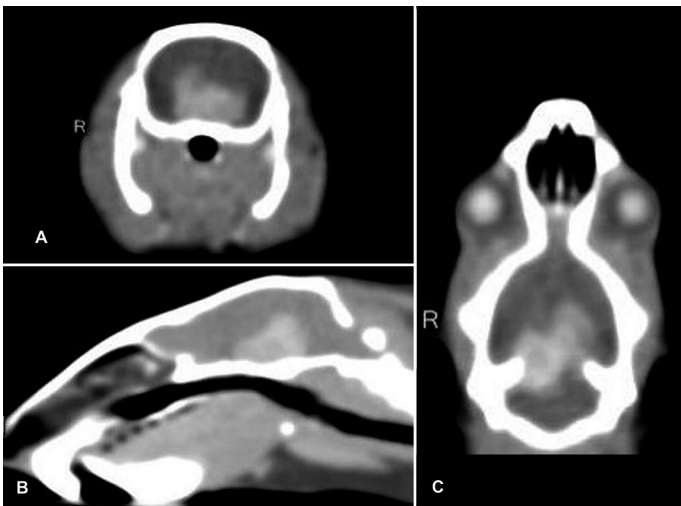


Fig. 3. Post-contrast CT images show a round, irregular margined, poorly circumscribed and nonhomogeneously contrast enhancing mass of the tumour pituitary

the pituitary tumour was oval and symmetrical. Post-contrast tomograms revealed a regular, smooth edge of the tumour in the pituitary region in 5 rats and irregular and partly obliterated contour of the tumour edges in 15 rats (Fig. 3).

Pituitary tumours are among the 5 most common intracranial neoplasms (14, 16). Despite the fact that in most cases these neoplasms are mild, most frequently they lead to animal death (2). Pituitary tumours are difficult to diagnose and in many cases they are only diagnosed in a post-mortem examination or at an advanced stage of the disease (12, 13).

Diagnosis of pituitary tumours in rats solely on the basis of clinical examination results is difficult and carries a huge risk of error. If the tumours are relatively small, clinical symptoms may suggest other conditions, such as middle ear inflammation. While making a diagnosis one may take into account the efficacy of treatment (*ex juvantibus* diagnosis) – in the case of otitis media the administration of bromocriptine and cabergoline will have no effect (1).

To make a final diagnosis, it is recommended that tomography and magnetic resonance are performed on patients (6). Unfortunately, due to the limited availability of the equipment, the high cost of the examination and because the patient needs to be specially prepared (placing a permanent venous line, premedication), these examinations are not commonly available, they are rarely performed and only in large centres. Moreover, there is not much information in the literature on this topic (19). However, the usefulness of these examinations in diagnosing only the tumours or establishing and comparing the outcomes of therapy is invaluable (4). What should also be pointed out is the safety of performing tomography examination in rats. No side effects were observed in any of the patients that underwent computed tomography.

The results of the authors' own study confirm the observations of other investigators who showed a high correlation between the use of tomography in the evalu-

ation of changes in the pituitary tumour lesions and the results obtained in the post mortem anatomopathological examination (4). It proves that CT may and should be used to evaluate the efficacy of treatment.

This article presents the usefulness of tomography examination in diagnosing pituitary tumours in rats and it is the first paper of its kind in Poland. However, one should also bear in mind that CT has certain limitations. Tomography without the use of a contrast medium allows for the identification of neoplasm only in some of the cases. It is the contrast-computed tomography that significantly increases the diagnostic chances of this method in the identification and evaluation of the size, shape and contour of proliferative lesions in the brain region in rats.

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Corresponding author: Assist. Prof. Łukasz Adaszek, ul. Głęboka 30, 20-612 Lublin, Poland; e-mail: ukaszek0@wp.pl