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Plasma Concentration of Calcium, Magnesium and Phosphorus in Chinchilla with and without Tooth Overgrowth

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The aim of the study was to identify the causes underlying overgrowth of incisors in chinchillas through an analysis of selected plasma electrolyte concentrations, with particular consideration of minerals involved in the formation of osseous tissue, i.e. Ca, Mg, and P. The analysis involved 40 female standard chinchillas managed in a commercial farm system, aged 2 to 4 years, divided into two groups of 20 individuals each: D – chinchillas with incisor overgrowth and C – controls with normal dentition. Concentrations of Ca, Mg, and P were measured in blood plasma. The analysis was carried out using ICP OES (inductively coupled plasma optical emission spectrometry) by means of the Optima 2000 DV instrument (Perkin Elmer). The resulting data were analysed statistically using one-way ANOVA with Duncan's range test. The results show that abnormal metabolism of dental tissue minerals, especially Ca and P, cannot be excluded as the cause of tooth overgrowth in chinchilla.

Key words: Chinchilla, teeth, overgrowth, Ca, Mg, P plasma concentration.

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The chinchilla is a rodent with hypsodont dentition, i.e. characterised by high-crowned teeth, in which all the root teeth grow permanently. It is this type of dentition that is often affected by a pathology consisting of inadequate tooth wear and, in consequence, tooth overgrowth (KOMSTA 2008). Another symptom of the condition is that enamel is unevenly distributed over the surface of the tooth which grows throughout the life of the animal. Chinchilla enamel is the toughest and the most durable material of the dentition, resulting in reduced wear of the teeth. The surface of chinchilla teeth is carved, which helps in better grinding of hard food components. Tooth overgrowth results in sharp tooth edges and in changes within the alveoli (Fig. 1).

Enamel is an epithelial tissue devoid of cells, composed of 96.5% inorganic compounds, with the remaining part constituting water and organic matter. The tissue consists of hydroxyapatite crystals, $Ca_{10} (PO_4)_6 (OH)_2$, which form structured enamel prisms. Enamel covers dentine as a layer of varying thickness. In herbivores, enamel covers nearly the entire tooth, being distributed evenly over the whole surface of the tooth stem, or forming only enamel strips running along the tooth. The outer side of enamel is covered by an amorphous, breakable, very resistant dental cuticle, which – together with the enamel – protects the underlying dentine (KUBASIEWICZ 1974; LUTNICKI 1972; MACHOY *et al*. 2005).

The first stage of tooth overgrowth can be recognised by drooling and anorexia, followed by eye watering. Clinical examination in most cases results in a diagnosis of misplaced and elongated incisors (CROSSLEY & MIGUELEZ 2001; SULIK *et al*. 2004). Also the sharp edges of the cheek teeth injure the internal surface of the oral cavity. The next stage, visible only on radiographic images, is characterised by elongation of tooth roots which reach beyond the bone surface, osteitis, and abscesses (CROSSLEY *et al*. 1998; KOMSTA 2008).

Previous studies have demonstrated alveolar expansion visible on radiographic images (SULIK *et al*.

Fig. 1. Tooth surface of chinchillas affected by tooth overgrowth (photo M. SULIK).

Fig. 2. Changes within the alveoli. Teeth ingrowth into the eye sockets and swollen mandible (photo M. SULIK).

2007). Dental pulp obliteration and excessively mineralised osseous tissue were observed in the molars. These conditions were irreversible and treatment of afflicted animals was futile. The radiographic images were mirrored by post-mortem examination of the skulls dissected from disease-afflicted chinchillas (Fig. 2).

In chinchilla farming, rapidly developing in Poland over recent years, tooth overgrowth is becoming a common disoder. The severity of the problem has reached a level at which research should be undertaken in order to determine the causes underlying tooth overgrowth affecting farmed chinchillas.

The aim of this study was to find the causes of incisor overgrowth in chinchillas through an analysis of concentrations of selected plasma electrolytes, with particular consideration of the elements participating in osseous tissue formation, Ca, Mg, and P.

Material and Methods

*The study involved 40 standard chinchilla females aged 2 to 4 years, managed on a commercial farm, of which 20 (group D) were afflicted with incisor overgrowth, and another 20 (group K) had normal dentition.

Blood was drawn from the hearts of the chinchillas onto K_3 EDTA and centrifuged immediately upon collection. The resulting plasma was stored at -20°C until analysis. Plasma concentrations of Mg, Ca, and P were measured by means of ICP OES using the Optima 2000 DV spectrometer (Perkin Elmer).

Samples for spectrometric assays were mineralised with 65% HNO_3 (Suprapur, Merck) in a high-pressure microwave mineraliser (Anton Paar), equipped with a system of permanent pressure and temperature control in each of the quartz mineralisation vessels. We used the ICP Multielement Standard Solutions IV (Merck). In order to standardise nebulisation conditions of the standard and the sample solutions, the standard solutions were replenished with an addition of the mineralising acids at concentrations equal to those in the analysed samples.

The analysis was carried out in argon plasma by the internal standard method, through the addition of yttrium (5 mgY/l) into the standard and sample solutions, in order to further mineralise potentially occurring disturbances in nebulisation, as well as against any other physical disruptions.

The emitted radiation intensity was measured at element-specific and recommended wavelengths, choosing the axial optical path (along the plasma).

Table 1

Mineral composition of enamel (MACHOY *et al*. 2005)

Organic and inorganic compounds forming enamel	Share in enamel (in %)
Organic compounds	3.5
Calcium phosphate	89.0
Calcium carbonate	4.0
Calcium fluoride	2.0
Magnesium phosphate	1.5

*The experiments were appeared by the local Ethics Committee in Szczecin, Permission No 11/05.

Table 2

Blood plasma concentrations of Mg, Ca, and P, as well as Ca:P ratio in healthy (C) and tooth overgrowth afflicted (D) chinchillas

Animal no.	Mg mmol/l		Ca mmol/l		P mmol/l		Ca/P	
	C	D	C	D	C	D	C	D
1	0.82	1.16	1.54	2.94	0.0714	0.0697	21.57	42.18
2	1.33	1.35	2.69	2.82	0.0793	0.0665	33.92	42.41
3	1.29	1.16	2.37	2.82	0.0749	0.0636	31.64	44.34
4	1.15	1.36	2.24	2.62	0.0785	0.0688	28.54	38.08
5	1.38	1.32	2.77	2.62	0.0846	0.0665	32.74	39.40
6	1.23	1.05	2.50	2.33	0.0772	0.0720	32.38	32.36
7	1.19	1.61	2.59	2.79	0.0788	0.0717	32.87	38.91
8	1.16	1.11	2.02	2.39	0.0788	0.0746	25.63	32.04
9	1.43	1.29	2.62	2.41	0.0788	0.0739	33.25	32.61
10	1.33	1.42	2.29	2.84	0.0851	0.0775	26.91	36.65
11	1.13	1.24	2.05	2.92	0.0736	0.0714	27.85	40.90
12	0.79	1.42	1.43	2.64	0.0810	0.0746	17.65	35.39
13	1.15	1.18	2.38	2.87	0.0765	0.0672	31.11	42.71
14	1.08	1.11	2.38	2.94	0.0791	0.0730	30.09	40.27
15	1.30	1.20	2.82	3.07	0.0878	0.0833	32.12	36.85
16	1.30	1.53	2.46	2.82	0.0856	0.0743	28.74	37.95
17	1.33	0.94	2.64	2.50	0.0891	0.0694	29.63	36.02
18	1.37	1.31	1.97	2.64	0.0836	0.0785	23.56	33.63
19	1.14	1.35	2.16	2.77	0.0794	0.0743	27.20	37.28
20	1.21	1.01	2.57	3.02	0.0785	0.0714	32.74	42.30
X	1.21	1.26	**2.32*** **	**2.74*** **	**0.0801*** **	**0.0721*** **	28.96	38.00
±SD	0.17	0.17	0.38	0.21	0.0046	0.0046		

***Differences significant at $P \leq 0.01$

The results were analysed using a one-way ANOVA with the Duncan range test (STATISICA.pl software package).

Results

Table 2 presents blood plasma concentrations of selected elements in healthy control and tooth overgrowth afflicted chinchillas. Calcium-phosphorus ratios for the analysed plasma are included.

Blood plasma magnesium concentration in the healthy chinchillas ranged between 0.79 and 1.43 mmol/l, with a mean of 1.21 mmol/l, whereas in the chinchillas suffering from dental disease it ranged from 0.94 to 1.61 mmol/l, remaining within the normal values.

Plasma calcium concentration in the healthy chinchillas ranged between 1.43 and 2.77 mmol/l, with a mean of 2.32 mmol/l, whereas in the chinchillas with dental disease it ranged from 2.33 to 3.07 mmol/l, with a mean of 2.74 mmol/l.

Phosphorus concentration in the plasma of the healthy animals ranged between 0.0714 and 0.0891 mmol/l, with a mean of 0.0801 mmol/l, while in chinchillas affected by dental disorders it ranged from 0.0636 to 0.0833 mmol/l, with a mean value of 0.0721 mmol/l.

Discussion

The magnesium concentration level is considered normal if it remains within the range of 0.74-1.23 mmol/l (ŚMIELEWSKA-ŁOŚ 2000). In the group of afflicted chinchillas, a slightly increased level of magnesium was observed. Magnesium function is strictly coupled with the function of calcium and phosphorus (JAROSZ *et al*. 2004). It is essential for the mineralisation process and growth of bones and enamel. Hypermagnesemia may result from renal failure in conjunction with excessive magnesium intake (ANGIELSKI *et al*. 1996). An excess of this element may lead to disorders in the cardiac conductive system, reduced blood pressure, torpor, muscular-neural weakness, and hypocalcaemia. Hypocalcaemia, on the other hand, results in osteomalacia and osteoporosis (ANGIELSKI *et al*. 1996; ARNOLD & GAENGLER 2007).

The observations presented here have revealed a highly significant difference in the plasma calcium concentration between the afflicted chinchillas and the healthy controls. There is very little data in the literature on the biochemical parameters of healthy chinchilla blood. Plasma concentration of minerals has been best described for the guinea pig, a species similar to the chinchilla (SULIK *et al*. 2007). The normal plasma concentration of calcium ranges from 2.08 to 3.0 mmol/l (ŚMIELEWSKA-ŁOŚ 2000). The results observed in this study do not exceed those cited in the reference; however, in animals afflicted with dental disease this value was close to the upper limit of the reference range. An elevated calcium concentration may have resulted from hyperparathyroidism, or from an overdose of supplemented vitamin D (HERNANDEZ-FERNANDEZ & PELAEZ-COMPOMANES 2003; BARANOWSKI *et al*. 2008). An elevated calcium concentration may lead to anorexia and, consequently, a lower food intake; this leads to reduced mastication, which in consequence prevents the dental surfaces from wearing (CROSSLEY 2001; LIESGANG *et al*. 2007; SONE *et al*. 2005).

It has been accepted that the plasma phosphorus concentration in guinea pig normally remains within the range of 0.97 to 2.46 mmol/l (WINNICKA 1997). In the presented study, a deficiency of this element was found in both analysed groups. Phosphorus deficiency in the afflicted group was higher compared with the healthy controls, and the differences were significant. Apart from calcium, phosphorus represents a basic structural component of bone and teeth (SONE *et al*. 2005; SULIK *et al*. 2007). A drop in phosphate concentration may be related to excessive excretion or transfer to cells, as well as to an insufficient dietary supply of phosphates; it may also be due to hyperparathyroidism and deficiency of vitamin D which determines phosphorus absorption (ANGIELSKI *et al*. 1996; ARNOLD 2007). The deficiency is manifested through a number of symptoms, including anorexia, ossification disorders (osteomalacia, bone demineralisation, osteoporosis) or tooth decay (CROSSLEY 2001; JAROS *et al*. 2004).

Observations carried out in a chinchilla reserve in Chile (SOTO 1993) have revealed that the natural diet of the chinchilla is very diversified depending on the season of the year. In summer, wild chinchillas graze basically on grasses, whereas in winter mostly on seeds and fruit. Roots and bark are not preferred food items. The natural diet of wild chinchillas is rich in dry plant components of the vegetation growing in the higher parts of the Andes. The plant elements eaten by chinchillas are coarse and thus facilitate tooth wear. Moreover, the plants are soiled with tiny quartz particles, which additionally increase tooth abrasion.

Application of a monodiet in the form of balanced pellets on a modern commercial farm probably does not satisfy the animals' requirements for minerals. The pellets are also too soft, and the seeds used for their production are too finely ground, which does not induce the animal to gnaw the food intensively (REITER 2008). This is another factor that may lead to overgrowth of the chinchilla's ever growing teeth.

In conclusions:

1. Abnormal metabolism of dental tissue elements, especially Ca, and P, cannot be excluded as a cause of tooth overgrowth in chinchilla.

2. Environmental factors that are related to chinchilla on-farm management may be of importance, especially a diet that does not provide a sufficient amount of coarse material needed for tooth wear, or does not meet the requirements for minerals.

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