Periodontal diseases are among the most common oral lesions observed in carnivorous animals. According to literature, they affect over 80% of dogs aged three years or older (16). Untreated gingivitis may, relatively quickly, lead to periodontitis and impairment of the restorative processes of periodontal tissues. Metabolites produced by bacteria penetrate tissue inducing inflammation. Their cytotoxicity towards periodontal tissue results from enzymatic damage, they also suppress the organism’s immunological mechanisms. The presence of inflammation causes damage to periodontal ligaments, bone resorption in the alveolar process on all interdental surfaces, and root furcations. It is harmful to epithelial attachment, gingival connective tissue, periodontium and root cementum (6, 7).

The goal of conventional periodontal treatment is to eliminate etiological factors and damaged tissue, as well as to stop the disease process. The subsequent therapeutic step is aimed at regenerating the damaged periodontal tissues and restoring its proper function. The reconstruction of damaged periodontal structures is done with the use of biomaterials (13). A number of regenerative techniques have been developed, including guided tissue regeneration (GTR) and guided bone regeneration (GBR). Considerable advances made in recent years in terms of the uses of implant materials in the treatment of periodontal diseases have greatly increased our ability to regenerate damaged tissues (19).

Rapid development in the use of various types of implant materials in periodontal reconstruction in...
In our in vivo studies microporous hydroxyapatite material of controlled homogenous pore size distribution and pore sizes within the range of a hundredth and tenth parts of a micrometer were applied. The above material was developed at the Faculty of Materials Science and Ceramics, University of Science and Metallurgy, Kraków, Poland. The initial hydroxyapatite powder was obtained by the wet chemical method. The granules and shaped blocks were obtained directly from the filter cake after its drying followed by calcination at 800°C. Fabricated material shows a high water absorption ability, which is a particularly advantageous feature in medical applications. In animal experiments microporous HAP granules (0.3-0.5 mm) and shaped blocks with ~68 vol.% porosity and average pore size 0.1 µm were applied.

The study was conducted on 40 dogs treated at the Department and Clinic of Veterinary Surgery at the University of Life Sciences in Lublin, divided into 2 groups. The first group comprised 2 sub-groups: a study group (G1) and control group (K1) with 10 dogs each. Similarly, the second group comprised a study (G2) and control (K2) sub-groups. Dogs qualified for the study were selected for systemic diseases based on medical history, clinical examination and the examination of blood parameters. Dental examinations of patients were performed under general anaesthesia. Prior to the collection, the owners of dogs qualified for the study and were informed of the purpose of the present study and gave approval for their participation.

The dominant clinical symptoms in all animals from the first group (G1, K1) included: feeding disorders and fetor ex ore. The following were evaluated: gingival index (GI), depth of periodontal pockets (PPD), clinical attachment level (CAL) and tooth mobility (TM) (Tab. 1). Based on the above parameters, the severity of periodontal inflammation was assessed (the fourth stage of PD). Prior to therapy, radiological examinations were also performed in order to determine the extent of damage to the alveolar process.

The gingival index (GI) was established after Lӧe and Solness. It was rated on the following scale: 0 – healthy gingiva (gum light pink in colour), 1 – mild inflammation – minor colour changes and slight swelling, no bleeding during probing, 2 – moderate inflammation – reddening, swelling and gingival gloss, bleeding present during probing, 3 – severe inflammation – strong reddening, swelling, ulceration and incidence of spontaneous gingival bleeding.

The examination of pocket depth using a periodontal probe and assessment of connective tissue attachment (the distance from the cemento-enamel-junction to the bottom of the pocket) was performed with the use of a Williams periodontal probe. Tests were performed in four locations at the surface of all incisors (mesial and distal labial and lingual side). The sum of all obtained values was divided by four to obtain the index value for the particular tooth. Subsequently, values obtained for individual teeth were added up and divided by the number of teeth examined. Patients qualified for the study were diagnosed with mandibular or maxillary incisor gingival
pockets of ≥ 4 mm and loss of connective tissue attachment of ≥ 1 mm. On the basis of the above, the animals were classified under one of the following three categories depending on their PPD index: 1–3 mm — physiological depth of the gingival sulcus, 3–4 mm — mild periodontitis, 4–6 mm — moderate periodontitis, 6–9 mm — severe periodontitis.

The value (in mm) obtained from the measurement of connective tissue attachment level was used as a reference value in the assessment of ongoing treatment.

Tooth mobility was measured with a probe. 4 classes of mobility were adopted: 0 — no mobility, 1 — lateral tooth mobility of 1 mm or less, 2 — lateral tooth mobility of over 1 mm, 3 — lateral and vertical tooth mobility.

All indexes were measured at the onset of treatment and then during subsequent checkups. The result of dental examinations are presented in Table 1.

Groups G1 and K1 were determined by dogs diagnosed, on the basis of the GI index, with moderate or severe periodontitis, considerable inflammation of periodontal tissues, presence of 4-9 mm gingival pockets and significant loss of connective tissue attachment. Due to gingival recession and alveolar bone atrophy in the course of periodontitis, the patients also suffered from maxillary or mandibular incisors mobility (mobility class 2 and 3). All animals underwent dental treatment. The therapy included removal of tartar and alveolar bone atrophy in the course of periodontitis, the incidence of gingivitis prior to treatment and after the therapy, both in the study group and the control group. To verify normal distribution of the numeric data, the Shapiro-Wilk test was used. For the comparison between the particular groups, the t-Student test was used.

Statistical analysis. A statistical analysis was conducted to establish whether any significant correlations could be observed between the gingival pocket depth index and the incidence of gingivitis prior to treatment and after the therapy, both in the study group and the control group. To verify normal distribution of the numeric data, the Shapiro-Wilk test was used. For the comparison between the particular groups, the t-Student test was used.

Results and discussion

Eight weeks after dental treatment and the introduction of hydroxyapatite granules into gingival pockets, Group 1 animals displayed significant shallowing of gingival pockets. The mean depth of pockets was significantly reduced in all animals in this group (by 3-4 mm) when compared to the original measurement (T-Student level of p < 0.0001). The results of the statistical analysis are presented in Fig. 1. Mean values of gingival index (GI) and clinical attachment level (CAL) were significantly reduced (p < 0.0001). Considerable regeneration of connective tissue attachment and improved tooth stability was observed in clinical study which allowed the avoidance of tooth extraction (Tab. 1).

In Group 2, following canines extraction and implantation with hydroxyapatite blocks, a considerable acceleration of bone cavity healing was observed. Dental imaging performed after 4 months from biomaterial implantation revealed that 50% of the implant had been reconstructed and replaced with bone tissue (Fig. 6). Full osteointegration was observed after 8-12 months from the treatment.

In control group K1, after 8 weeks a slight reduction of periodontal pocket depth was observed, and in these weeks after the dental treatment, patients started coming back to the Clinic due to a relapse of periodontitis, which in most cases necessitated extraction of unstable incisors.
In the analysis of the PPD index after hydroxyapatite treatment in the study group (G1), as compared to the K1 control group results without the use of biomaterial, significantly lower means were observed in the study group (Fig. 2).

Statistical data confirm the viability of the study as the observed periodontal pocket depth was significantly reduced after hydroxyapatite treatment (Group I), as compared to the values reported in the control group (K1) – Tab. 2.

In control group K2, where maxillary canines extraction had been necessary, the lack of biomaterial implantation lead to complications in the form of oro-nasal fistulas, which developed in 60% of the cases and required further intensive treatment. In the remaining 20% of the cases, the bone healed correctly, however, dental imagery revealed incomplete filling of the post-extraction cavities with bone tissue.

The purpose of performing reconstructive procedures with the use of biomaterials is in human and veterinary medicine alike to recreate the integrity of damaged tissue and restore its proper function. Reconstructive materials most commonly used in dentistry include autogenic bone grafts and alloplastic materials (8). Among the latter, hydroxyapatite, used in human as well as veterinary dentistry, has become a model compound reflecting the bone mineral phase. It is considered to be a leading biomaterial in bone surgery due to its high biocompatibility, osteoconductivity and osteoinductivity as well as capacity to form chemical bonds with living tissue (9, 12). Dense hydroxyapatite materials are not resorbable and bone reconstruction takes place around the material particles (22). In porous forms, they undergo biodegradation and are gradually replaced with regenerating bone. In dental surgery, hydroxyapatite is typically used in the form of lamellas, granules or blocks of various shapes and sizes. The size of HAp granules and the diameter of pores in the range of macro and microporosity impact the effects of bone reconstruction. The smaller the granules, the greater the area of interaction formed and the faster the material’s resorption (Fig. 2, 3). The minimum size of open pores allowing for the formation of a biological bond between the implant and the bone is 100 µm (10). Porosity of the implant material also facilitates the conduction of osteoblasts, oxygen perfusion, and angiogenesis in the newly formed bone tissue. If not enough space is left, bone tissue growth becomes impossible (25). Hydroxyapatite induces the production of growth factors and populations of osteogenic cells, and by ensuring a favourable chemical environment, it facilitates the formation of new bone (11).

In human dentistry it is common to use biomaterials in the treatment of bone defects, a technique known as guided bone regeneration (GBR). Hydroxyapatite used as a filling for bone cavities forms a matrix for the production of new bone tissue. It has been demonstrated to be non-immunogenic and non-toxic. After implantation,

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean</th>
<th>SD</th>
<th>n</th>
<th>t</th>
<th>p-Value</th>
</tr>
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<td>0.974337</td>
<td>10</td>
<td>2.13267</td>
<td>0.046976</td>
</tr>
<tr>
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<td></td>
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</table>
a porous structure is formed in place of the bone cavity, which facilitates the process of tissue regeneration and limits gingival recession (3). Due to its osteoconductivity, it provides a perfect base for osteogenic cell adhesion, which helps the formation of new bone (7, 18). HAp stimulates more active tissue regeneration when compared to a purely natural process (5).

In veterinary medicine, bone reconstruction with the use of biomaterials remains under clinical investigation. Song et al. (20) demonstrated in their research a higher resistance to femoral bone damage in dogs implanted with a prosthesis in the form of a synthetic femoral bone coated with hydroxyapatite versus autogenic bone grafts introduced into the medullary cavity. Other researchers also confirmed that a hydroxyapatite coated prosthesis may improve the efficiency of bone tissue growth and ensure a good connection with the natural bone (1).

In their study of hydroxyapatite, Lee et al. (14) demonstrated its effectiveness as a substitute for an autogenic shinbone graft in rabbits. It was used in the form of blocks and powder. The researchers concluded that hydroxyapatite facilitated consolidation with the host bone tissue.

In the context of veterinary dentistry, a commonly observed type of bone damage (vertical) occurs along the root and towards the crown of the tooth, which is the reason why oronasal fistulas are so common in dogs (most often located in the vicinity of the canines on the palatal side). Fistulas are also formed as a result of periapical lesions or extraction of maxillary canines. The healing of a post-extraction cavity is not dissimilar to the processes of new tissue formation observed in instances of long bone fractures (4). Tissue regeneration affects the bone edges of the wound as well as the inner surfaces of alveolar walls. It involves a combination of intra and extra alveolar processes of reformation and resorption of bone tissue. Connective tissue proliferates into the alveolus, in effect reducing the height of the alveolar process edge. It is relatively rare for the cavity to be completely filled with properly formed bone tissue (18). The use
of biomaterials offers a solution to this problem. Under optimum implantation conditions in the area of cell embedding, the cells’ ability to reorganize and form new tissue is greatly facilitated. In our study, complications of this type were observed in control group K2 animals following maxillary canines extraction, where in some cases the post-extraction cavity was still not fully filled with bone after 1 year from the procedure. Meanwhile in Group 2 animals, where microporous hydroxyapatite bioceramics was used in combination with antibiotics, proper implant in-healing was observed and the fistula closed within as little as 2 weeks. Complete filling of the post-extraction cavity with properly structured bone tissue was observed within 8-12 months from the procedure.

The clinical potential of bone substitute materials allows for their widespread use in various areas of human dentistry. Apart from treatment of bone damage, hydroxyapatite has become a common material used in periodontal regeneration related to the presence of bone pockets and tooth furcations. Its application allows the improvement of tooth stability and radiologically confirmed reduction of bone pocket depth (22).

In one of the study groups (Group 1), the implantation of microporous hydroxyapatite granules soaked with gentamicin into ginvial pockets significantly accelerated the healing of periodontal tissues and reduction of ginvial pockets. Local administration of the antibiotic allowed for a focused therapeutic concentration. Satisfactory results of the treatment could be observed as early as eight weeks after the procedure. The restorative qualities of hydroxyapatite facilitated significant shallowing of ginvial pockets, healing of the implantation areas, and stabilising mobile teeth. No such results were observed in the control group (K1) subjected to scaling combined with general antibiotic treatment. In this group, the ginvial pockets were only slightly shallower and many patients returned to the clinic after a short period of time with symptoms of periodontitis.

In clinical cases of oral inflammation in animals, an important therapeutic objective is to ensure regeneration of the periodontium and bone with the possible use of biomaterials, including microporous hydroxyapatite bioceramics. In dogs with periodontopathology, the use of above hydroxyapatite accelerates the healing process and saturated with gentamycin limits the development of inflammation. Clinical evaluation confirms that hydroxyapatite enhances regeneration of tooth support tissues and restoration of their proper function. In dogs, it has also been used as a filling for post-extraction cavities. It speeds up the reconstruction of bone tissue. The performed study confirms the validity of using microporous granules and shaped hydroxyapatite blocks in veterinary dentistry as filling for facio-maxillary bone defects and bone pockets. The economical aspect of the same is also noteworthy. The cost of the biomaterial is significantly lower than that of other available implantation materials, which further adds to its attractiveness in the context of veterinary medicine.

References

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