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Gold Beads Implants for The Treatment of Canine Chronic Recurrent Otitis Externa

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Abstract : A clinical trial was performed to assess clinical efficacy and/or reduction in relapses by gold-bead implantation into acupuncture points in dogs with canine chronic recurrent otitis externa (CCROE). Forty dog-patients randomly divided into two groups were diagnosed as suffering CCROE, having intact tympanic membranes and a history of recurrences. Treatments were: control group (CG), treated with commercially available antimicrobial otic droplets, dosed twice daily for 7 days; and experimental group (GBI- gold bead implants), treated as for CG plus the insertion of 13 gold-bead implants under light anesthesia. Overall per cent assessment of composite clinical progression and progression of individual clinical signs were recorded. Bilateral chronic external otitis was diagnosed in 60% of the cases and left or right otitis in 20% of the cases each. Logistic model for repeated measures analysis showed that GBI induced a better clinical recovery as far as lesion score of some clinical signs is concerned. The overall percent cures of each group showed statistically significant difference. A McNemar analysis revealed that higher number of relapses was observed in CG patients as compared to the GBI ($P < 0.05$). In particular during these days, lesion on the pinna showed in odd ratios analysis a 7:1 ratio (recurrence CG:GBI) and ear wax/pus (4:1) from D42 to D365. It is concluded that gold-bead implantation into acupuncture points improves resolution of some clinical signs and greatly reduce relapses in CCROE affected dogs after 1 year follow-up (98.75%).

Key words **:** chronic-otitis, dog, gold-bead-implants, acupuncture.

Introduction

For dogs, it has been stated that, as far as otitis externa and otitis media is concerned, information based prospective studies that adhere to the principles of evidence-based medicine are rare (22). This view was taken as the impetus to carry out a clinical assessment of a medical procedure based on the insertion of gold beads in acupuncture points. For the basis of such a study, it is possible to define canine chronic recurrent otitis externa (CCROE) as an acute or chronic inflammation of the ear canal and/or pinnae, either unilateral or bilateral without perforation of the tympanic membrane. This form of otitis is one of the most common diseases in dogs (11). The etiologies associated with otitis external can be classified into the following: 1. Predisposing factors, such as anatomical and conformational inadequacies, excessive moisture, iatrogenic interventions and obstructions (polyps and tumors), 2. Perpetuating causes such as bacteria, yeast and otitis media, and 3. The primary causes, such as parasites, foreign bodies which can be from plants to dirt, insects, hypersensitivity and allergic diseases, keratinization disorders and autoimmune diseases (30). It is common that a primary etiology in a predisposed patient causes external otitis and perpetuating circumstances lead to chronic recurrent cases with a chronic infection (4,12).

Under normal circumstances, external ear canal has low number of resident as well as transient bacteria; but a normal healthy ear will yield some organisms such as *Staphylococcus* sp., *Streptococcus* sp., *Pseudomonas* sp., and *Proteus* sp. Once over-colonization of the ear canal occurs, the most common pathogens found include: *S. pseudintermedius, Pseudomona aeruginosa, Proteus mirabilis, Escherichia coli, Corynebacterium* spp, *Enterococcus* spp and *Streptococcus* spp. Similarly *Malassesia pachydermatis* can be found in healthy ears and when otitis sets, it is also the commonest finding with the occasional isolation of *Candida* spp (10,30). Diagnosis is based on changes of the pinna and direct and visualization of the ear canal under anesthesia, observing inflammatory changes of the ear canal (erythema, stenosis, proliferation, ulceration) or the presence of foreign bodies, masses, as well as consistency, color of exudates and patency of the tympanic membrane when possible (5). Myringotomy may be performed if fluid is present in the tympanic cavity. Cytological assessment and anaerobic/aerobic culture, as well as sensitivity testing, are recommended but often an empirical - antimicrobial therapy is initiated (5).

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Treatment of external otitis consists of medical and surgical therapy. Medical therapy involves 1-2 week treatment with local and parenteral antibiotics often combined with antifungal agents and topical glucocorticoids that are formulated generally in otic droplets (22). With appropriate treatment, improvement is typically observed within 1-2 weeks. Animals with extensive pathological changes, recurrent clinical signs, or cases refractory to medical therapy may require surgical intervention such as total ear canal ablation combined with bulla osteotomy when complicated with otitis media (18,21). Yet, in spite of these interventions, relapses are commonly observed. With this scenario, it is not exceptional that small animal clinicians see themselves confronted with disappointed owners when failing to achieve a definitive resolution to this apparently uncomplicated health problem.

Gold implants in acupuncture points are widely used in human dentistry, surgery and medicine (2,7,24). In veterinary medicine small gold beads or small pieces of gold tread are inserted into specific acupuncture points and it has been advanced that they can be effective for many severe, otherwise degenerative conditions, such as cervical spondylomyelopathy, degenerative myelopathy, severe spondylosis, hip dysplasia, elbow or knee arthritis, and epilepsy (1,10,15,16). Yet, to others this technique offers no proven benefit over regular acupuncture and poses substantial potential for injury (27,28). Furthermore it has been suggested that, since implantation is regarded as malpractice in human medicine, a similar label should be given to this procedure in veterinary medicine.

Prompted by positive personal experiences with gold bead implantation into acupuncture points, and based on the hypothesis that gold bead implantation can speed up the recovery and/or delay or impede the recurrence of canine chronic recurrent otitis externa, a randomized clinical trail with evaluation of progress by three clinicians blinded to treatment was carried out in forty dogs affected by infectious chronic external otitis and assessing two treatments: administration of a conventional antimicrobial drugs combination as otic droplets, and the same treatment plus thirteen gold bead implants in acupuncture points.

Materials and Methods

Study population and design

A longitudinal study with regular time schedule was performed with seven days interval, an ultimate visit was done at 365 day. This study received approval by the Ethics Committee of the Postgraduate Studies Division at the *Facultad de Medicina Veterinaria* of the *Universidad Nacional Autónoma de México*. In this study, only client-owned dogs were eligible for inclusion and they were gathered from 6 veterinary hospitals around Mexico City in a two year period from 2007 to 2009. In all, forty dogs aged from 1 to 8 years old were randomly assigned, with a table of random numbers, to either of two groups. Inclusion criteria demanded a diagnosis of canine chronic recurrent otitis externa either unilateral or bilateral, which was achieved through direct otoscopic examination under tiletamine-zolacepam sedation, with an intact tympanic membrane, and a history of three or more previous episodes of the same problem. Perpetuation of primary causes was ruled out i.e., parasites, foreign bodies, and evident hypersensitivity reactions according to the dermatologic history questionnaire proposed by Rosser (30, and keratinization disorders linked to endocrinological problems. Similarly, predisposing features were absent such as anatomical conformational factors, excessive moisture, iatrogenic intervention, obstructions such as polyps, tumors. Evaluation of progress was based on the absence of the above listed primary causes and predisposing factors as well as based on recording the following signs: lesions of the pinna, hemorrhagic lesions, pruritus, presence of pus and/or ear wax, using in all these variables the following scale: zero = absence of sign, 1 = moderately present; 2 = clearly present and 3 = severe form of the giving symptom. Ulceration of the tegument was classified as present or absent; the integrity of the tympanus as with inflammation or without it, and the cerumen/pus odor was graded as present or absent. Assessment of clinical signs through observations was made by three independent clinicians blinded to treatment. Healing or degree of healing was established every 7 days until day 49 of treatment for each symptom, as follows: with lack of signs (100% cured); with noticeable progress (75% cured); with less signs than at the beginning (50% cured); with no signs of improvement or deterioration (25% cured) and worse than at the beginning (zero per cent cured). Also a composite overall lesion score was obtained weekly, using the mean percent value of all symptoms. Relapses were checked upon until D4 and an ultimate confirm was done at day 365. For ethical considerations a control-untreated group was not regarded as an option. An indicator of clinical efficacy was taken as the average of healing on all symptoms at each day of observation. Evaluation of the disease was carried out on the following days 0 (D0), 7 (D7), 14 (D14), 21 (D21), 28 (D28), 35 (D35), 42 (D42) and 49 (D49). Then an ultimate visit at day 365 (D365) was made. If zero per cent would have been detected on D7, dogs would have been graded as “not cured” recorded and excluded from the trial to pursue another treatment scheme out of the protocol. Such case did not occur.

Treatment

Treatments were as follows: group 1 was treated with commercially available antimicrobial otic droplets formulated in an oily matrix, so each mL contains: gentamicin sulphate 3 mg; betamethasone valerate 1 mg and clotrimazol 10 mg. Dose administered was 5 droplets twice a day during 7 days. Group 2 was treated as for Group 1, plus the insertion of 13 gold bead implants 1 to 1.2 mm in diameter, under light anesthesia with tiletamine-zolacepam (2-3 mg/kg IM; Telazol, Fort Dodge, Mexico), in the acupuncture points shown in Fig 1 and described in Table 1. Insertion of the gold-beads was

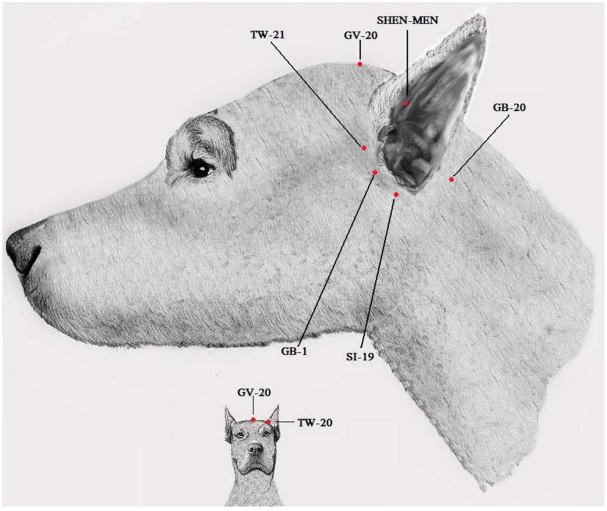

Fig 1. Location of the acupuncture points chosen to implant gold beads for the treatment of canine chronic recurrent otitis externa.

achieved by inserting a Gauge-16 hypodermic needle containing the gold-bead into the acupuncture point, and pushing the bead with a sterile wire-plunger. Thorough soap-cleaning and skin disinfection with 4% clorhexidine (Clorhexiderm Spray; DVM Pharmaceuticals, Inc., Fl, USA).

Microbiological analysis

Samples for bacteriological and fungal culture were obtained from all patients mainly from ear exudates swabs, at the beginning of the trial and on D365; hence, a total of 80 clinical samples were collected. Total transfer time to the laboratory was not more than 2 h. For aerobic and facultative strains, the specimens were cultured on blood agar (Columbia agar with 5% sheep blood), mannitol salt agar, MacConkey agar, and brain heart infusion broth. The cultures were incubated at 35ºC-37ºC for 24 to 48 h. Samples obtained for anaerobic culture were inoculated into brain heart infusion agar - supplemented with 5% sheep blood, hemin (5 µg/mL), and menadione (1 µg/mL) - and bacteroides bile esculin agar. The cultures were incubated at 35ºC-37ºC in anaerobic conditions, achieved by using a commercially available gas generator envelope for anaerobiosis. For fungal isolation, the samples were inoculated into Sabouraud glucose agar supplemented with 0.05% chloramphenicol, 0.05% cycloheximide, and Dixon's agar. They were kept at room temperature for 15 days. Bacterial species were identified by Gram morphological and biochemical characteristics, as described by Murray *et al*. (23). Fungal isolates were identified by their macroscopic and microscopic morphology, according Sidrim and Moreira (31).

The hypothesis test for parameters P binomial distribution was used to investigate significant differences between the isolation profile from the left and right ears of the same animal. Differences were considered significant at $P < 0.05$. Statistical analysis for clinical efficacy was carried out based on the repeated measures mixed model at IBM SPSS 19®, and a regression logistic model for repeated measures with the R package (26) was used to analyze each ordinal variable (25). The McNemar Change Test which is used for testing the shift in the proportion of abnormal responses from before and after treatment in the same group of dogs was used to analyze the changes between D42 and D365 with IBM SPSS 19®,.

Results

Table 2 shows the population included in this trial, listing gender and whether the canine chronic recurrent otitis externa was bilateral or unilateral. Also in this table results from bacterial culture and fungal culture are shown. Either bacterial and/or fungal growth was obtained in all samples. The agents most frequently isolated were *S. intermedius* and *Malassesia pachydermatis*. A single microorganism was present in 27.5% of the samples. Two microorganisms were isolated in 50% of the samples and 3 or more were present in 22.5% of the samples. The most frequent association observed was *S. intermedius* + *M. pachydermatis* (54.8%). Canine chronic recurrent otitis externa was bilateral in 60% of the cases, 20% left and 20% right sided only. Although most frequently the same

Table 1. List of acupuncture points utilized for gold-bead implantation

Western Nomenclature	Chinese name	Anatomical Location
SI 19; small intestine	TONGZILIAO	On the face, anterior to the tragus and posterior to the mandibular condyloid process.
GB 1; gall bladder	SHANGGUAN	Anterior to the intertragic notch at the posterior border of the condyloid process of the mandible.
GB 20; gall bladder	FENGCHI	In the depression created between the origins of the sternocleidomastoid and trapezium muscles, at the junction of the occipital and nuchal regions.
TW 20; triple warmer	JIAO SUN	Directly above the ear apex
TW 21; triple warmer	ERMEN	With the mouth open, in the depression anterior to the supratragic notch and posterior to the mandibular condyloid process.
GV 20; governor vessel	TA-FENG-MEN	On the top of the head, middle of the natural line of the root of mane, namely at te crosspoint of the external parietal crest of the parietal bone
Wonder point	SHENMEN	At the bifurcation of crura of the antihelix

Table 2. Population of dogs included for gold bead implantation and as control animals, listing gender, type of otitis treated, as well as bacterial and fungal culture results

GROUP	GENDER	EAR	Bacterial and fungal isolation		
			Etiological agent found	40 days after treatment	1 year after treatment
1	8 M 12 F	4 Unilateral/right 4 Unilateral/left 12 bilateral	13 *Staphylococcus intermedius,* 8 *Malassezia pachydermitis,* 4 *Proteus mirabilis,* 3 *Streptococcus* sp, 3 *Staphylococcus aureus,* 3 *Candida albicans,* 2 *Pseudomonas* sp, 2 *Trichophyton mentagrophytes* 2 *Serratia liquefaciens* 1 *Klebsiella pneumonia,* 1 *Shigella sp* 1 *Acinetobacter* sp 1 *Aspergillus* sp	5 *Staphylococcus intermedius* 1 *Streptococcus* sp, 1 *Pseudomonas* sp 1 *Proteus mirabili* 1 *Malassezia pachydermitis* 1 *Staphylococcus aureus,*	7 *Malassezia pachydermitis,* 7 *Staphylococcus intermedius,* 4 *Staphylococcus aureus* 2 *Streptococcus* sp, 1 *Proteus mirabilis,* 1 *Trichophyton mentagrophytes,* 1 *Pseudomonas* sp
2	9 M 11 F	4 Unilateral/right 5 Unilateral/left 11 bilateral	11 *Staphylococcus intermedius,* 6 *Malassezia pachydermitis,* 6 *Proteus mirabilis,* 4 *Staphylococcus aureus,* 3 *Streptococcus* sp, 3 *Candida albicans,* 3 *Pseudomonas* sp, 2 *Trichophyton mentagrophytes* 2 *Serratia liquefaciens* 2 *Klebsiella pneumonia,* 1 *Shigella sp* 1 *Acinetobacter* sp 1 *Aspergillus* sp	4 *Staphylococcus intermedius* 1 *Streptococcus* sp, 1 *Pseudomonas* sp 1 *Proteus mirabilis* 1 *Staphylococcus aureus,*	4 *Malassezia pachydermitis,* 3 *Staphylococcus intermedius,* 3 *Staphylococcus aureus* 1 *Proteus mirabilis,* 1 *Trichophyton mentagrophytes,* 1 *Pseudomonas* sp

species of microorganisms were isolated from both ears, some differences were observed in the association pattern between the right and left ears in 3 of the 40 animals (7%) Results from anaerobic culture were negative for all the samples studied.

The most frequent clinical signs recorded were lesion on the pinna (72%), hemorrhagic lesions (87%), and ulceration of the tegument (80%), ear wax/pus (73%), and pruritus (100%). Treatment success was statistically superior in GBI (P = 0.002) with an overall per cent degree of healing of 98.75% as compared to 68.24% in CG. The overall composite degree of healing along the length of this study in both groups is shown in Fig 2, where significant improvement until day 365 (OR = 13 CI_{95} = 10, 17 P = 0.0001) is observed for both groups, with a slightly greater and faster efficacy in dogs of the GBI.

When clinical signs were analyzed one by one, superior success in lesions on the pinna (OR[1] = 14.74, P = 0.001), hemorrhagic lesions (OR = 4.75, P = 0.02) pruritus (OR = 10; P = 0.001), and ear wax/ pus (OR = 4.33; P = 0.003), were obtained in dogs of the GBI. Statistical analysis showed no significant differences in odd ratios when assessing, ulceration of the tegument (OR = 0.46, P = 0.23) and integrity of the tympanus (OR = 2.7, P = 0.17). Based on the McNemar Change Test recurrence of lesions started to show a pattern on day 35 as compared to D42 but only for patients in CG.

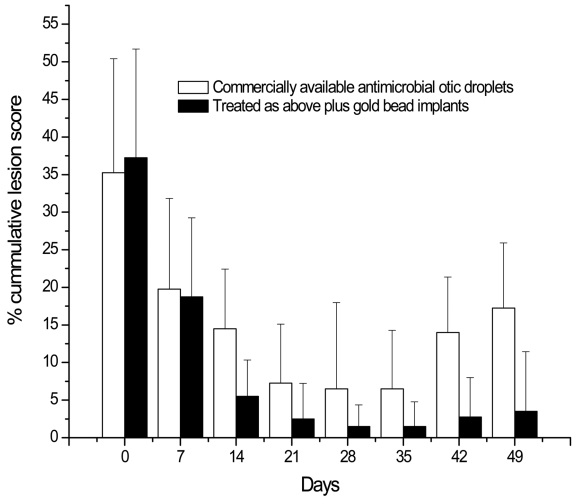

Fig 2. Frequency bars + 1SD of the the cumulative lesion score progression of cases of canine chronic recurrent otitis externa treated either with commercially available antimicrobial droplets only (empty bars) or adding gold bead implants (black bars) to the same treatment.

[1]Overall OR

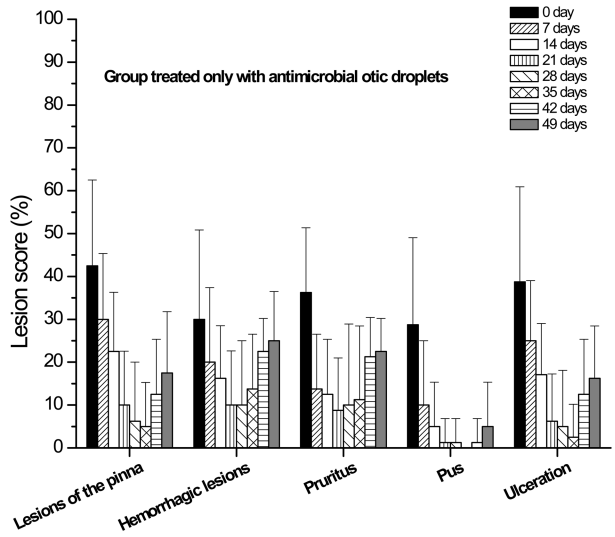

Fig 3. Frequency bars of all signs of canine chronic recurrent otitis externa in the group treated only with commercially available antimicrobial - antiinflammatory otic droplets.

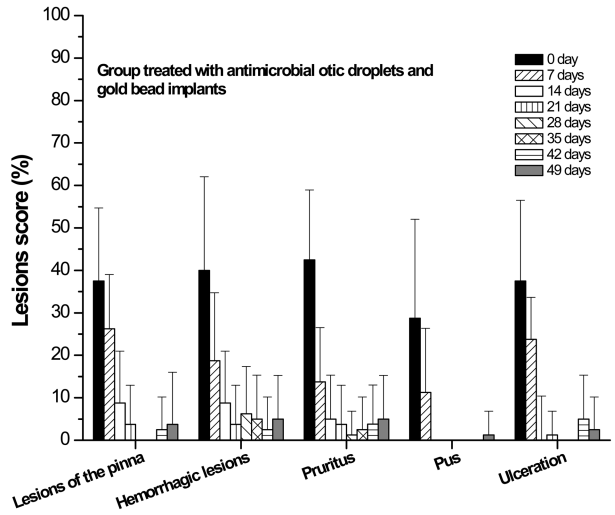

Fig 4. Frequency bars of all signs of canine chronic recurrent otitis externa in the group treated with commercially available antimicrobial - antiinflammatory otic droplets plus gold bead implantation in 13 acupuncture points.

This was also observed from D42 to D365 as shown in Figs 3 and 4, where progress of individual signs is depicted. In particular during these days lesion on the pinna showed a 7:1 ratio (recurrence CG:GBI) and ear wax/pus (4:1) from D42 to D365, at 365 day almost all dogs was remitted in ulceration of the tegument and integrity of the tympanus lesions in both groups.

Discussion

Validity of a clinical trial relies on stringent methodology and statistical design to avoid bias interpretation of data. In this study, an attempt was made to avoid such problems through close monitoring of individual clinical progress, and using proven statistical methodology (25). Data from longitudinal studies in general, and from clinical trials in particular, are prone to incompleteness. For example, geographical mobility of patients, owner's willingness to cooperate with this study and interference with other health issues such as changes in diet, other infections and so forth, prevented this study to have a longer follow-up. However, the statistical analysis here elected, uses an on-line software package, known as "R" (26). This method provides consistent estimates of the model parameters, providing that missing data occurred completely at random. The algorithm used for parameter estimation has been shown to be efficient and more robust than other generalized estimating equation methods for a limited range of data, as estimation of the correlation parameter is based on a complete case analysis (25). However, an evident experimental weakness of this study is the lack of a true negative control group, as well as a group using only gold beads for implantation. Both approaches were not possible due to reluctance of owners to be included in such groups. Future assessments of this technique may consider the inclusion of a group treated only with gold bead implantation, thus avoiding possible artefacts.

The use of gold bead implants has proven to be effective for the treatment of other conditions mainly cervical spondylomyelopathy and musculoskeletal disorders (8), also for pain management in hip-join arthritis (15). Conversely, negative results have also been published (27). Differences may be ascribed to methodology and more particularly to statistical analysis, rather than to gold bead implants *per se.* In this study, at first there appears to be no outstanding beneficial effect in the group treated with antibacterial droplets plus gold bead implants (GBI), as compared to only using antibacterial droplets (CG). Yet, if a careful analysis of individual symptoms is carried out differences in favor of using gold bead implants can be detected, offering a faster healing as shown in Figs 3 and 4. But particularly important is the lack of recurrence in the GBI that can be observed towards the end of this trial at 365 day as compared to a pattern of recurrence on day 35 of the CG. In this clinical trial, no predisposing or perpetuating factor for CCROE could be pin-pointed in any patient of either group. It is then tempting to speculate that an undetermined predisposing factor may be present; for example, individual susceptibility to ear infections due to immune incompetence in that particular body area. This is not in complete agreement with Rosser (30) whose referral practice ascribes as primary cause for CCROE hypersensitivity and allergic diseases. Such a difference may deserve further analysis if other possible causes i.e., the nutritional status.

Based on these studies, a direct deleterious effect of gold bead implants on bacteria or fungi seems unlikely. Predictably, the microbial agents most frequently isolated were *S. intermedius* and *M. pachydermatis* before and after treatment, and bacteriological cure was not obtained in any case.

This feature plus the fact that there were high cure rates in both groups reveals that, antimicrobial drugs are important for the treatment of CCROE, but lower or lack or recurrence in GBI may reflect a change in the individual susceptibility to infection rather than a direct effect of the gold beads on the etiological agents. Persistence of etiological agents in dogs, otherwise clinically free of CCROE, is in agreement with the known existence of resident and transient bacteria and yeast found in normal ears (30) and is also consistent with the lack of congruity when culturing and assessing sensitivity patterns of samples obtained from the external ear canal and tympanic cavity (4,5,30). Nevertheless, it has been stated that topical therapy is key to the successful resolution of otitis externa which is essentially a surface infection. Systemic antimicrobials are unlikely to achieve therapeutic concentrations within the fluid and waxy exudates of the external canals in which the infectious organisms are harbored (22).

This study was not structured to determine the mechanism of action of gold bead implants to enhance clinical efficacy, and there appears to be no complete explanations in formal literature. In this respect, apart from the perspective of health and disease as explained by acupuncture which may be further analyzed elsewhere (3,14,17), no sound explanation has yet been universally accepted. However, based on the assumption that gold ions diffuse into the surrounding tissue and mimic, on a local scale, the treatment with gold-containing drugs used for arthritic conditions (6), the following actions can be credited: after implantation, local macrophages and other inflammatory cells attach themselves to the metallic gold surface (19). This attachment is mediated by activation of the complement system as C3 adsorbs to the implant surface. The C3 forms complexes with complement factor B or factor H resulting in the formation of C3b or iC3B respectively. These latter complement fractions are ligands for macrophages surface receptors. Additionally, fibronectin and vitronectin absorb to the locally implanted surface of the gold bead and act as a ligand for the macrophages through RGD-integrin receptor domains (proteins that contain the Arg-Gly-Asp). These and perhaps other ligands are the primarily interactions between gold surface and macrophages and other inflammatory cells. It is also known that the inflammatory cells produce an ultra-thin layer, a dissolution membrane, within which the necessary chemistry for liberation of gold ions is found. This is 10-100 nm thick biolayer membrane essential for the dissolution of metal implants and particles which cannot be phagocytosed (19). The inflammatory cells (e.g. macrophages, other neutrophils) release cyanide compounds (for example aurocynide = $Au(CN)2^{-}$) into the dissolution membrane (2,13), and this stable ions, inhibit the lysosomal enzymes of inflammatory cells in the synovial tissue and decreases the number of inflammatory cells *in situ*. Aurocynide inhibits antigen processing and suppresses NF-kappa B-binding activity and I-kappa B-kinase activation, and in turn reduce the production of pro-inflammatory cytokines (20). It has been theorized also that gold implants emit a minute positive electrical charge that neutralizes a negative electrical charge of the point, producing analgesia and preventing further arthritic changes at the joint (9,32). Consequently, better improvements in the GBI in lesions on the pinna (OR = 4.75, P = 0.02), ear wax/ pus (OR = 10; P = 0.001), and pruritus (OR = 4.33, P = 0.003), anti-inflammatory actions, appear to be consistent with the so far proposed mechanism of action, and may explain the lack of a recurrence pattern that started to show on day 35 for the GBI.

In veterinary medicine the overall consensus is that gold bead implants do not cause apparent side effects. Contraindications to the use of this procedure include cancer or bone infection at the implant sites and it must be considered that gold bead implants may compromise the quality of computerized scans. However, long term follow up studies are not available. In contrast, it has been stated in humans that implants may activate mast cells, cause contact dermatitis, and gold-induced myelotoxicity (29). Hence a longer follow up study is necessary to assess long term effects of gold implants and long-term frequency of recurrences of CCROE. However, based on theses results, it is our view that benefits of implanting gold beads in acupuncture points, for the treatment of CCROE, outweigh possible risks.

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