

Veterinaria

Novel application of using ozone for urobiomic restorative therapy with Microbiome Restorative Therapy in antibiotic resistant *E. faecium* canine urinary tract infection and to regain autonomic nerves.

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Keywords

Ozone , Microbiome Restorative, Therapy MBRT, Fecal Microbiotia Transplantation, FMT, fecal transplant, gut microbiome, Antibiotic resistance, Urine Biome Transplant, antibiotic stewardship

Abstract

Objective: The aim of this study was to examine the role of Microbiome Restorative therapy (MBRT) (Fecal as well as urine transplant) by also involving ozone therapy, ultraviolet blood therapy with hemealumen, acupuncture, homoeopathy, multi-lumen laser and nutritional support in the alteration of the antibiotic-resistant urinary infection with damaged autonomic nerves in the injured dog

Material and Methods: Ultraviolet Blood Therapy at UVBI 50 cc of O3SS and 5 cc of blood and 1 cc of hypertonic Biocean with the Hemealumen, MBRT orally was started on day one and rectal MBRT on day 5. Homoeopathy treatment with Aconite 1 M, Arnica 10 M and home on Laser Multilumen (LM), 1 Natrum Sulfate as herbal support for bladder nutraceuticals was given to the injured dog. A combination of medical ozone at the dose rate of 60 cc 37 µg/mL and ozonated saline O3SS was infused into the bladder and 60 cc. 41 µg/mL gas insufflation of the bladder as well as 80 cc of subcutaneous O3SS fluid over the pelvis and lumbar area. Aqua puncture and LM therapy with pain and neuropathy frequencies over the affected back area were given.

Results: After 6th day's observation, the total bowel and urinary infection were controlled with normal stool, clear urine and also walked normally. He was happy and acting like a puppy, no longer afraid and depressed.

Conclusion: MBRT with systemic ozone therapy had a bio-modulatory effect, reducing the characteristics of acute inflammation and helping the dog to have a normal condition...

Suggestion on how to quote this paper:

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Introduction

The variety of microbial life in the gut provides 70-85% of immunity to the body. An unhealthy gut leads to many medical symptoms in mammals. Many research studies suggest that the gut microbiome plays a necessary role in host digestion, immunity, the central nervous system, behaviour (anxiety and depression), and other metabolic disorders (Yong, 2016). The gastrointestinal system of dogs, for example, is colonized by complex members of microorganisms, namely the microbiome. The gut microbiome's foundation is built prior to life which can be affected by external factors, involving antibiotics, dietary factors, and contaminants in food and drinking water (Tashiro and Shore, 2021). MBRT has gained popularity in recent years and involves the transplanting of gut microorganisms from a healthy animal to a diseased one to recover optimal gastrointestinal health. This treatment, by far, has been used for intestine-originated disease in dogs when a common therapy does not work. On the other hand, ozone therapy constitutes mostly promotes blood circulation, peripheral and central ischemia, immunomodulatory effect, energy boost, regenerative and reparative properties, and correction of chronic oxidative stress. Ozone therapy enhances the interest in new neuroprotective strategies that may show therapeutic targets for minimizing the effects of oxidative stress (Elvis and Ekta, 2011).

This case study involves the role of MBRT with ozone therapy to cure an injurious and deadly urinary infection in a dog who was 14-month-old Male Rottweiler, hit by a car and severely comminuted fracture of vertebra L7-S1 with luxation that was repaired. After 2 months still he still had no anal tone with excoriating diarrhoea and a bleeding ulcerated tail. The atonic bladder with a non-responsive antibiotic-resistant urinary tract was infected that smelled like dead fish even after 2 months of chloramphenicol.

Methodology:

A 14-month-old Male Rottweiler was hit by a car and had a severely comminuted fracture of vertebra L7-S1 with luxation was repaired. After 2 months it still had no anal tone with excoriating diarrhoea and a bleeding ulcerated tail. The atonic bladder with a non-responsive antibiotic-resistant urinary tract infection continued that smell like dead fish even after 2 months of chloramphenicol. Two tablets of chloramphenicol (1 g/tablet) were given by mouth three times a day, with food or shortly after a meal. Gloves were worn while handling the medication to avoid rare and fatal side effects including aplastic anaemia. Side effects reported for chloramphenicol in dogs include vomiting, anorexia, and diarrhoea while serious side effects include bone marrow suppression. To monitor this, a complete blood count was recorded while canine urine, faeces or even saliva was not tested.

To recover the dog from this injurious infection MBRT with ozone therapy was utilized. On entry at MASH on 3/1/2022 Titan was in an insecure and painful condition around his tail. Both diapers had extremely smelly stools and blood was seen in the urine and stool at the bleeding tail. The treatment was done and DNA culture in the urine was examined for regular aerobic and anaerobic infection. At the first integrative exam, the same resistant bacterial strain was found to be present. After the reports were examined, the antibiotics were immediately stopped and the dog was kept on analgesics on the urecholine for three more days.

Day 1 treatment

- UVBI therapy: It was given with a dose rate of 50 cc Ozonated saline, 5 cc blood, 1 cc Hypertonic Biocean.
- Ozone therapy: Subcutaneous O₃ saline flooded over the pelvis at the rate of 150 cc approximately and distributed from L3 to the base of the tail.
- Aqua puncture with B12, B1 (60, 40 23, 57) was provided all along the sacrum and lumbar to Bau Hua, tip of tail, under tail and base of tail
- After the aqua puncture the Multi-Laser on neuropathy was given for 20 minutes over the back and infection over the bladder. The LED laser with rife frequencies was used.
- Then oral microbiome supported with nutraceuticals was given to regain the oral and gut microbiome.
- Catheter was placed within the bladder, and UA and cultures were taken for examination.
- 60 cc of ozonated saline as a flush and removed with the addition of 60 cc of O₃ gas at 41 µg /ml
- 5 cc of vitamin C and 2cc of vitamin B complex and vitamin A and D (1cc into 30 cc) was added into 2 separate pockets of O₃ saline.

Day 3 and on

- Similarly, UVBI therapy was given with the same dose rate of 50 cc ozonated saline, 5 cc blood, 1 cc Hypertonic Biocean.
- Similar treatment was performed as on day 1 till day 6.
- On Day 5 the same treatment was given with added rectal ozone 60 cc at 24 µg /ml and 50 cc of slurry of MBRT.

The following prescription was followed for the treatment:

8:58:59a Tx:
m UVBI-H 5cc/50cc/1cc Hypertonic
SQ ozone 30cc/30cc 50cc
Vitamin AD 1cc into L bolus
Vitamin B/C 2cc/4cc into L bolus
Bladder flush 60cc ozonated saline
Aquapuncture
Multi Lumen Laser 10 minutes on NeuropathyDiagnostics:
NoneSupplements:
Cranberry Comfort
MASH mix
Ultra EFA
Nutrigest
Colostrum
Ion Gut
Canine Immune Support
Canine Musculoskeletal Support
Neurotrophin PMG
Nat Sulph LM1
Arnica 200C
Thuja LM1
Thuja 200C
Ozone Oil
Bulk Fecal
Take Home Ozonated Saline

8:58:59am

Tx:

UVBI-H 5cc/50cc/1cc Hypertonic

SQ ozone 30cc/30cc 50cc

Vitamin AD 1cc into L bolus

Vitamin B/C 2cc/4cc into L bolus

Bladder flush 60cc ozonated saline

Aquapuncture

Multi Lumen Laser 10 minutes on Neuropathy

Diagnostics:

NoneSupplements:

Cranberry Comfort

MASH mix

Ultra EFA

Nutrigest

Colostrum

Ion Gut

Canine Immune Support

Canine Musculoskeletal Support

Neurotrophin PMG

Nat Sulph LM1

Arnica 200C

Thuja LM1

Thuja 200C

Ozone Oil

Bulk Fecal

Take Home Ozonated Saline

Result and Discussion:

After the Day 3 treatment following observations were noticed.

- Urine was clearer and had less odor and
- Stools slightly formed but still incontinence

On Day 6 the stools were normal with complete control of the bladder for urination on his own and was able to walk for 2 miles.

The use of these alternative modalities helped to reduce the infection with the use of ozone and restoration of the microbiome. The other modalities helped in reducing inflammation and trauma to the tissue. Neurotransmitters originating from the microbiome helped the damaged nerves to recover from the accident. He also regained his happy mental puppy behaviours as his owners got their dog back. We know that ozone increases the nitrous oxide in the tissue and the flooding of the back and bladder help to increase NO in these areas. Some herbs can help bring NO to tissue but the direct placement of the ozone could be a factor. He was happy and acting like a puppy with no longer fear and depression and till date doing well with a normal life.

Doing Microbiome Restorative Therapy MBRT with Urobiome for the Bladder Urine transplant

Because Titan had been on antibiotics for so long the idea of doing a microbiome urine transplant became an option. In the literature, there has been no documented treatment. The owner is a nursing student and she wanted to try doing a urine transplant. She signed off on this unknown treatment. The donor was one of our microbiome donors in Vienna and she has been a donor for her entire life at eight years of age. "Touch of Life" donors have never had antibiotics, NSAIDs or pesticides, or herbicides and are only fed an organic fresh predominate plant-based diet with some free-range animal protein. A free first-morning sample of her urine was obtained at 6 a.m. A urine culture and analysis were sent out but the results did not come back for several days. We did the UVBI treatment, as done previously but this increased the volume to 80 cc 8 cc of blood and 2 cc of Hypertonic Biocean. We gave only ozone gas 60 cc within the bladder at 41 µg per milliliter to remove any biofilm within the bladder. No saline flush was done and we had the catheter remain and waited for about 15 minutes to allow the ozone to reduce the biofilm and for it to be broken down. We injected 30 cc of the donor's urine that was placed in the refrigerator after collection and before delivery. The catheter was pulled and the urine was massaged around the bladder. He was sent to the car and was told not to urinate for as long as they could possibly hold it and he held it for several hours.

New urine culture was taken after the UMBRT showed a more sensitive bacteria that could be treated with multiple antibiotics.

Status *Proteus mirabilis* - 10,000 - 50,000 CFU per ml

Isolate 1 MIC

Amoxicillin S ≤ 2

Amoxicillin-Clavulanic Acid S ≤ 2

Cephalexin S 8

Cefpodoxime S ≤ 0.25

Cefovecin S ≤ 0.5

Ceftazidime S ≤ 0.12

Ceftiofur S ≤ 1

Amikacin S ≤ 2

Gentamicin S ≤ 1

Ciprofloxacin S ≤ 0.06

Enrofloxacin S ≤ 0.12

Marbofloxacin S ≤ 0.5

Doxycycline R ≥ 16

Nitrofurantoin R 128

Chloramphenicol S 8

Trimethoprim/Sulfamethoxazole S ≤ 20

Cefotaxime S

(A)

Antibiotic Resistance for Detected Clinically Relevant Microbes

The sample was screened for the presence of antibiotic resistance genes and intrinsic resistances of clinically relevant microorganisms. For this analysis more than 90 antibiotic resistance genes were screened. The cautious use of any antibiotic drug is highly recommended. Please follow the guidelines for antimicrobial stewardship in veterinary practice.

Drug Class*	Antibiotics	Enterococcus faecium (Ei)	Proteus mirabilis (Pi)	Suggested Dose†	Drug Delivery
1st	Colistin	NDR	F	15 mg/kg, q 12 hrs	IV, SC
	Colistin	NDR	F	4-20 mg/kg, q 8 hrs	PO
	Colistin	NDR	F	22 mg/kg, q 12 hrs	PO
	Colistin	NDR	F	22 mg/kg, q 8 hrs	PO
	Colistin	NDR	F	15 mg/kg, q 12 hrs	IV, SC
	Penicillin G	F	NDR	8-10 mg/kg, q 8 hrs	PO
	Penicillin G	F	NDR	22 mg/kg, q 8 hrs	IV
	Amoxicillin	NDR	NDR	22 mg/kg, q 8 hrs	IV, SC
	Amoxicillin	NDR	F	22 mg/kg, q 8 hrs	PO
	Clonazepam	NDR	NDR	12-75 mg/kg, q 12 hrs	PO
	Clonazepam	NDR	NDR	6 mg/kg, q 24 hrs	IV, SC
	Valproic acid	NDR	NDR	---	NPT/Topical Use
	Neomycin	NDR	NDR	---	Topical Use
	Chloramphenicol	NDR	NDR	5-5 mg/kg, q 12 hrs	PO
	Lincomycin	NDR	NDR	10-20 mg/kg, q 24 hrs	PO
	Doxycycline	NDR	NDR	5 mg/kg, q 12 hrs	PO
	Moxycycline	NDR	NDR	10 mg/kg, q 12 hrs	PO
	Tetracycline	NDR	NDR	20 mg/kg, q 12 hrs	PO
	Sulfamonomide	NDR	NDR	30 mg/kg, q 12 hrs	PO
	2nd	Tetrahydrothiopyridopyrimidinone	NDR	NDR	10-20 mg/kg, q 24 hrs
Metronidazole		NDR	NDR	10 mg/kg, q 8 hrs	IV
Carbapenem		NDR	U	8 mg/kg, once	SC
Carbapenem		NDR	U	3 mg/kg, q 24 hrs	PO
Carbapenem		NDR	U	2-2 mg/kg, q 24 hrs	SC
Tiamulin		NDR	U	---	Topical Use
Adjuvanted		NDR	NDR	5 mg/kg, q 12 hrs	PO
Chloramphenicol		NDR	U	2.5-7.5 mg/kg, q 24 hrs	PO
Chloramphenicol		NDR	U	20 mg/kg, q 12 hrs	PO
Fluorfenicol		NDR	NDR	20 mg/kg, q 12 hrs	PO
3rd	Amikacin	NDR	NDR	15 mg/kg, q 24 hrs	IV, SC
	Rifampin	NDR	NDR	5-10 mg/kg, q 12 hrs	PO
	Rifampin	NDR	NDR	20 or 200 mg/kg, q 8 hrs	PO
	Levofloxacin	NDR	F	10-30 mg/kg, q 24 hrs	IV/PO
	Marfloxacin	NDR	U	2-75-5.5 mg/kg, q 24 hrs	PO
	Phadofloxacin	NDR	F	2-2 mg/kg, q 24 hrs	PO
	Enoxacin	NDR	U	5 mg/kg, q 24 hrs	PO
	Ciprofloxacin	NDR	U	---	Topical Use
	Ciprofloxacin	NDR	U	3-30 mg/kg, q 6-8 hrs	IV
	Mupirocin	NDR	NDR	4-4 mg/kg, q 24 hrs	PO
Nidazolan	NDR	NDR	4-4 mg/kg, q 24 hrs	PO	
Colistin	NDR	F	8 mg/kg, q 24 hrs	PO	
Neomycin	NDR	U	2.5 mg, q 4 hrs	IV	
Piperasilin/Tazobactam	F	NDR	80 mg/kg, once, q 8 hrs	IV	

This table lists antibiotic sensitivities/resistances for the indicated bacteria based on detection of specific antibiotic resistance genes and naturally occurring, or intrinsic, resistance to specific antibiotics previously identified for that organism.

Abbreviation Keys:

- NDR Not Recommended (Due to either Resistance Genes Detected, Intrinsic Resistance, or < 10% Effectiveness in Antibiogram Studies)
- F Poor Performance (< 50% Effectiveness in Antibiogram Studies)
- U Fair Performance (< 75% Effectiveness in Antibiogram Studies)
- F Good Performance (> 75% Effectiveness in Antibiogram Studies)
- NDR No Antibiotic Resistance Detected Based on the MIDDOG Antibiotic Target Panel
- No Info
- PO Oral, by mouth
- IV Intravenous Injection
- SC Subcutaneous Injection
- TE Topical Use
-
- † Dose may vary based on patient species and/or type of infection. Reference at: www.msdvet.com/antibiotics.
- U Variable bioavailability in canine patients.
- † Contra-indicated in canine patients.

(B)

Supplemental Data

Total Bacteria Composition

Charts below depict the relative abundance of all detected bacterial species. Each color represents a different bacterial species. The larger the colored segment is, the more abundant that species is in the specimen.



The table below lists top 8 bacterial species detected within the limit of detection. The absolute and relative abundances of each species is shown.

Species Detected	Percentage (%)	Cells per Sample
Enterococcus faecium [1]	53.39	8,300,000
Proteus mirabilis [2]	46.6	7,200,000

Total Fungal Composition

No Fungi Detected:

Images represents output of Medical Examinations (A) The antibiotic resistance for detected clinically relevant microbes and (B) The total bacteria composition.

So a very disturbing part of this case was the use of chloramphenicol. The owner was told to avoid touching the pills and handling it with gloves. No one ever warned her about the urine and the feces that are full of chloramphenicol. Her three-year-old son was barefoot and he walked through the dog's urine and got head to toe body rashes and was taken to the emergency room having difficulty breathing. So this is a public health issue. We need to understand that if there are other ways of treating infections that aren't putting people at risk we need to be discussing those options. The clinical fact that this dog regained its bladder control and its anal control with normal stools and normal urination after six days has to be looked at and examined by conventional medicine. We have wonderful complementary and alternative tools like medical

ozone, aquapuncture, homeopathy, Laser, and Microbiome Restorative Therapy (MBRT) and we need to be able to share these with universities and colleagues.



Images of the son who got infected from dog's urine.

It is crucial that we protect the microbiome and at the same time be able to stop infection and support the immune system. For too many years indiscriminate use of antibiotics, pesticides, herbicides, drugs and chemicals have destroyed and altered the internal ecosystems of humans and animals. It is good stewardship of medical care to give bodies' a healthier way to reduce infection, inflammation and allow a healing process.

Conclusion

Bringing together the need to respect that antibiotics are developing severe resistance and putting thousands of human lives at risk (animals too) as well as the antibiotics should be provided only when required and we need to look at this case and talk about it.

We were able to control the infection with alternatives and may never need to use such a strong antibiotic that puts families at risk. By using these alternatives, we were able to regain almost total neurological weakness and damage. We were able to recover this confident loving dog back to mental health. With MBRT and its role at rebooting the gut microbiome we could see the restorative action in days. There may be cases that so many veterinarians are struggling with such issues they would benefit from the experience of this case. The novel idea of restoring the bladder and its urinary microbiome has real implications in all health. The one done on Titan may have been the first one ever done and turned out to be miraculous. It appeared to be safe without any side effects.

We give blood transfusions all the time and no one has done full blood cultures on every sample and worried that it could cause some unknown disease. It is screened in humans for HIV and hepatitis but not from a cancer that could occur at a later date.

The AVMA, AAHA and AAEP have in July 2022 joined together to bring this important change in the way we use antimicrobials. We have alternatives and this is why we need to understand how these will help us do a better job caring for our patients and protecting people from antibiotic resistance that kills over 700,000 humans annually in the world. There are many alternatives that can help rescue and stop an infection. Getting some training and exposure to good techniques will be helpful. Some of the alternatives used : Herbal medicine, Medical Ozone, Laser different classes, Ultraviolet Blood therapy, Homeopathy, photobiomodulation, Manuka Honey, rife frequencies. Even simple things like extensive soaking and flushing in warm water, sea salt, Epsom salts have some value. Directly jump to antibiotics because that has always been your tool. The times and conditions have changed and we need better stewardship.

For example for over 25 years I have NOT used antibiotics for dentals even with dogs with heart conditions. Using homeopathy like Arnica, Silica, Calendula and Hypericum take dogs out of pain and stop infections. Herbal mixtures with Echinacea, green tea will also be good. But the best one I have seen so far is ozone therapy. Used by human Biological Dentists around the world, it solves the problem that needs to be cure for our patients. Something for pain, infection, physical flushing of the pockets and debris, reducing the infected biofilm, helping the gums to become more oxygenated and therefore a faster healing response. By not having that information before, you are not judicious and being irresponsible by prescribing an antibiotic. But ozone does have those medical abilities of destroying bacteria, spirochetes, microorganism as the site and then floods the area with pure O₂ and the healthy tissue is restored to a better level of wellness. So let's see what else it can do for quality care.

An added reason why it worked

1. Using Aconite for shock using Nat Sulf to deal with the injury to the spinal cord could help as a homoeopathic remedy.
2. Flooding the fascia with ozone lifting up the fibers in the beaten up tissue over the surgical site by ozonated saline, this probably allowed the acupuncture meridians to bring the nerve sensations down to the tail allowing it to move and therefore opening up stagnation within that meridian. The tail wagging after the second acupuncture treatment most likely opened up channels to the distal meridians.
3. Did the ozone bring the mitochondria activity that increases the nitric oxide at the location where the ozone was flooded.
4. Using the conception vessel the tip of the remaining part of the Rottweiler's tail as well as bringing the whole daily circulation to his back was part of the aqua puncture treatment.
5. We placed the catheter within the bladder and initially flushed it with ozone to try to break up any other crystals that were there that went in a ceiling and were withdrawn and ozone gas at a higher micrograms per ml was placed within the bladder repeatedly.

6. Giving B12 in the acupuncture points helps that point be stimulated for a longer amount of time.
7. Using alternative modalities with all positive support for each other may have been a reason why the system was able to reboot and start nerve regeneration because it was not suppressed anymore with pain meds, antibiotics and other drugs.
8. The owner was continuing to give subcutaneous ozonated saline over the back in the interim time between our appointments.
9. The mental components of Titan came back as he had his gut repaired in the gut brain connection started to work again.

Funding:

Self-funding.

Conflict of Interest

Private practitioner at Main Street Animal Services of Hopkinton www.mashvet.com. No conflict of interest exists.

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References

1. Bonforte, G., Bellasi, A., Riva, H., Ferradini, M., Arrighi, E., Groppi, G., Franzini, M. (2013). Ozone therapy: a potential adjunct approach to lower urinary tract infection? A case series report. *Giornale Italiano di Nefrologia: Organo Ufficiale Della Societa Italiana di Nefrologia*, 30(4), gin-30.
2. Dai, T., Vrahas, M. S., Murray, C. K., Hamblin, M. R. (2012). Ultraviolet C irradiation: an alternative antimicrobial approach to localized infections?. *Expert review of anti-infective therapy*, 10(2), 185-195.
3. Elvis, A. M., Ekta, J. S. (2011). Ozone therapy: A clinical review. *Journal of natural science, biology, and medicine*, 2(1), 66.
4. Hamblin, M. R. (2017). Mechanisms and applications of the anti-inflammatory effects of photobiomodulation. *AIMS biophysics*, 4(3), 337.
5. Hamblin, M. R. (2017). Ultraviolet irradiation of blood: "the cure that time forgot"? *Ultraviolet Light in Human Health, Diseases and Environment*, 295-309.
6. Masan, J., Sramka, M., Rabarova, D. (2021). The possibilities of using the effects of ozone therapy in neurology. *Neuroendocrinol. Lett*, 42, 13-21.
7. Roman, M. (2015). Micro-biome restorative therapy: successful treatment of dogs and cats with fecal transplants. *J Am Holistic Veterinary Med Assoc*, 38, 8-12.
8. Tashiro, H., Shore, S. A. (2021). The gut microbiome and ozone-induced airway hyperresponsiveness. Mechanisms and therapeutic prospects. *American journal of respiratory cell and molecular biology*, 64(3), 283-291.
9. Yong E. 2016. *I Contain Multitudes: The microbes within us and a grander view of life*. Random House.