Aerosol use of a novel disinfectant, as part of an integrated approach to preventing and treating aspergillosis in falcons in the UAE

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Introduction

Members of the family *Aspergillus* fungi have been associated with a wide range of disease syndromes in man and animals. Most prominent in this regard have been the species *A. fumigatus, A. flavus, A. clavatus, A. niger* and their associated mycotoxins. The hepatotoxic aflatoxins B1, B2, G1, G2 and the immunosuppressant Gliotoxins seem to play the biggest roles in their pathogenicity, amongst a range of dozens of fungal metabolites that exert physiological effects in animals, either alone or in synergistic association with each other, from this genus. The aflatoxins in particular are of major concern as human hepatocarcinogens. Extensive research has been conducted in lab and farm animals during the past 30-40 years after contaminated Brazilian groundnutmeal caused the death of hundreds of thousands of turkey poults ("Turkey X disease") and subsequent investigations revealed the association with aflatoxins. These fungi and their spores are ubiquitous, but strains differ widely in their ability to produce toxins, also the range of toxins produced by a particular strain under specific growth conditions (in vivo vs in vitro!) varies. Several biological, chemical and environmental factors ultimately determine the amount and combination of mycotoxins produced by toxigenic strains on a given substrate. Hot, humid micro-aerophylic conditions in association with organic material seems particularly favourable for explosive fungal growth along with the associated amounts of heterocyclic metabolites (mycotoxins) that are formed.

Furthermore, there are clear differences in the relative sensitivity of different animals to the effects of these mycotoxins, with extremely sensitive species e.g. trout hatchlings and ducklings even used as biological assays! Turkeys and ducks are the most sensitive commercial avians to mycoses and mycotoxicoses, while avian species from colder climates (penguins, Arctic waterfowl and gyrfalcons) rapidly succumb to aspergillosis under any stressful conditions, mainly as a fungal airsacullitis that progresses to a fatal, multisystemic disseminated mycosis associated with irreversible toxic damage to the parenchymatous organs. The infection may vary from chronic and insidious to acute death. The immunosuppressive effects of some of the mycotoxins predispose these birds to opportunistic bacterial infections e.g. *Pseudomonas aeruginosa*. Most veterinary attention has focused on these avian groups, but accurate, early detection of infection as well as prognostic assessment of " successfully treated " patients remain challenges to all clinicians faced with these cases. Laparoscopic examination of the trachea and caudal airsacs, improved radiological detail of soft tissues and the use of antibody detection by ELISA are all being used successfully, but many early cases still escape detection. Further refinement of the currently available ELISA systems developed by Professor Pat Redig and colleagues in the USA, both in terms of antigen and conjugate specificity and the

development of rapid, accurate toxin-based assays are logical next steps. The development of an effective vaccine remains elusive, in spite of major efforts in this direction by Poultry Research Organisations. However, new molecular techniques might be successful where conventional approaches have failed previously.

Pilot investigations by Professor Ullie Wernery from the Central Veterinary Research Laboratory, Dubai, in association with co-workers in Munich, Germany, on fungal isolates and affected organs obtained during necropsy on falcons here in the UAE have revealed high levels of certain mycotoxins and immunosuppressive gliotoxins. Further investigations exploring the importance, patterns of occurrence and counter-strategies against isolates from falcons are urgently needed to more effectively combat this very common condition. Falcon veterinarians can assist by collecting as much as possible sera from suspected , confirmed and convalescent aspergillosis cases for use in the validation of new tests/vaccines/treatment regimes.

Treatment of falcons with aspergillosis:

Early cases that are detected during routine examination procedures, many not showing any clinical signs except some green discolouration of the mutes, usually carry a fair prognosis. In the falcon hospitals in the UAE they are generally treated for 21-30 days with 10-20mg/kg itraconazole (Sporonox, Janssen-Cilag Ltd, Buckinghamshore, UK) OID, with food. During the 1999-2000 season several clinicians also experimented with an aerosol therapy in conjunction with this systemic treatment, with extremely encouraging results.

The particular design of the avian respiratory system that includes large spaces (airsacs) where air becomes humidified at body temperatures approaching 40°C and an absence of a rapid immune response due to the avascular nature of these structures, allows the establishment of fungal growth when the bird is exposed to high spore concentrations in the inhaled air. This also occurs in highly stressed/sensitive individuals when only low numbers of spores are inhaled. These fungal infections are often complicated with concurrent opportunistic bacterial infections. The delivery of antibiotics or antifungal drugs by aerosol to such affected patients has been tried many times, usually with disappointing results. This has been related to the extremely irritating nature of most antifungal agents, causing severe erosive lesions on the sensitive mucosal surfaces of the respiratory system, thus defeating the objective. Alternative explanations of such therapeutic failures usually focus on the physical nature of microdroplets needed to penetrate to the furthest recesses and diverticulae of the airsac system. Recommended optimal sizes usually vary between 5-10 microns, necessitating the use of "nebulising " systems. There are however many other variables that determine the integrity and size distribution of microdroplets in any fog, thus affecting their relative penetrating ability into the avian airsacs. Some of these include;

- Relative humidity of the inhaled air
- Surface tension/chemical makeup of the droplets

- Nozzle size, velocity of the air/compound pushed through the instrument
- Still air vs air movement

The practical realities of treatments in clinics, hospitals and farming environments dictate a pragmatic approach.

We have used commercial "Foggers" suitable for the disinfection of rooms, incubators hospital wards, etc, that produce a wide range of microdroplet sizes and rapidly create a "standing fog " under any environmental conditions. This system of aerosol delivery using very low concentrations of a novel disinfectant compound has also been used to treat, as well as prevent, a wide range of respiratory conditions in many species. These include respiratory conditions in poultry (including prevention of Newcastle Disease Virus transmission during outbreaks) and exotic birds (including falcons), pneumonia syndromes in pigs, "Kennel Cough Syndrome " in dogs (and contaminated hospital wards), "Snuffles Syndrome" in cats (and contaminated catteries/hospital wards). Also treated include a wide range of other medical applications e.g. contaminated wound irrigation, refractive fungal dermatoses, etc.

This compound, named F10, was formulated in the UK for disinfection within pharmaceutical manufacture plants, particularly aseptic fill areas (intravenous drips etc). Manufacture started in South Africa in 1994 and since then F10 has been tested against every significant/index animal/human pathogen. It is used in animal production and food manufacture as well as pharmaceutical manufacturing. It is a complete spectrum virucidal, bactericidal, fungicidal and sporicidal, but aldehyde free compound of six main synergistic active ingredients. The ingredients are comprised of a new Quaternary Ammonium Compound (QAC) plus a new Biguanidine Complex; it is classified by Registration Authorities as such , but the exact formula remains for obvious reasons a commercial secret. It drew veterinary attention as it not only outperformed other disinfectants available during efficacy testing, over a range of temperatures and in the presence of moderate organic material, but is did so at extremely low concentrations, short contact times, without any corrosive effects on infrastructure, metal alloy nozzles or any tissue irritation on workers and animals. Due to this combination of characteristics, it suddenly became feasible to regularly disinfect animal environments in their presence as a standard practice, lowering the environmental pathogen challenge significantly, **with absolutely no negative side-effects**. Many veterinarians in South Africa and neighbouring countries have now used this approach with extremely positive results. F10 is registered in South Africa (Reg No. G2781), New Zealand (MAF Reg No.H1280) and also passed all registration requirements in the UK (MAFF Reg No. outstanding, awaiting the completion of the administrative process).

I have used it in innumerable clinical situations in several countries (African and Arabian) since 1994 and was particularly encouraged with results obtained with "fogging" (not to be confused with "fumigation", the dangerous and outdated practice of using formalin plus potassium permanganate to produce formaldehyde gas) of poultry, ostriches and exotic birds suffering from complicated bacterial and fungal respiratory infections.

Individual patients have been treated in home-made Perspex boxes fitted with human nebulisers for 20-30 minutes at a time, 2-3 times a day, with a solution containing 0.2 % F10 Superconcentrate. Larger numbers of birds, especially if hooded or otherwise kept in the dark, have been treated by this " cold fogging" (in contrast to hot/steam fogging as commonly used in the disinfection of abbatoirs) with commercial portable Foggers in designated rooms. Air-conditioners are switched off for the 30-40 minutes needed to empty the 5 litre container of the model that we use, once or twice a day.

Gyrfalcons undergoing predictable stress periods (initial manning/training, returning from initial training in the desert) have undergone this treatment during the 1999/2000 season in our facilities in Dubai, with no side- effects, and none of the usual previous pattern of falcons sick or dying from aspergillosis. Early diagnosed cases have made remarkably quick recoveries with this treatment regime, but advanced cases, especially those from Russia, seemed to be beyond any recovery. Unlike experiences during previous years with aspergillosis in gyrfalcons, very few individual falcons were lost to this disease and we have started an integrated preventative programme as the next step.

Preventative medicine is in essence **Managing Risk**. Critical, predictable events or situations are identified and the associated health risks lowered as much as possible. In abattoirs, food processing plants this approach is known as "**Hazard Analysis Critical Control Point** (HACCP)" and has become the norm in modern export graded facilities. "**Integrated Pest Control**" is a term commonly used in agricultural pest animal control, usually referring to rodents or insects. The destruction of nests and prevention of breeding/multiplication, prevention of access to food sources, as well as the direct destruction of animals/insects through targeted poisoning etc are also essential parts to such programs.

A similar paradigm is needed in the control of Aspergillosis by managing the environment to make it as "fungus-unfriendly" as possible. This can be achieved by regular fogging with F10 of all chambers with newly arrived birds, new fledglings even while these are growing feathers and producing huge volumes of feather and dry faecal dust (perfect for bacterial and fungal growth!), as well as the hunting birds during training. Moulting chambers in the old style, covered with palm leaves to provide shade, are veritable "fungal gardens" during the extremely hot and humid conditions during summer in the UAE, and wherever possible should be replaced using more modern, inorganic materials like shadecloth netting or even brick and/or sandwich-panel customised buildings. The air conditioned boxes, A/C filters and feeding ledges in such facilities are regularly sprayed (backpack/knapsack sprayers), washed and fogged with F10 during summer thus inhibiting unchecked fungal growth and significantly lowering numbers of viable fungal spores and other pathogens in the falcons environment.

Please Note: While I am happy to discuss practical issues and personal experiences regarding the use of F10 or any other medication/ treatment protocol with my veterinary

colleagues all detailed enquiry's regarding test results or commercial aspects should be communicated directly with the manufacturers.

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