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DEPARTMENT OF VETERINARY PUBLIC HEALTH & EPIDEMIOLOGY
West Bengal University of Animal & Fishery Sciences
37, K. B, Sarani, Kolkata - 700 037,
West Bengal, India
Website: www.aphv.org.in.
E -mail: aphvl997@gmail.com

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"Zoonotic Potentialities of COVID-19 and significance of Veterinary Public Health"

UTPAL DAS

Supdtg. Veterinary Officer (utpaldasvet@gmail.com)

Deptt. of Health, Kolkata Municipal Corporation

Abstract

A novel coronavirus suddenly emerged in human population and escalate rapidly causing global coronavirus disease 2019 (COVID-19) pandemic. Though the origin of the associated virus SARS-CoV-2 (Severe Acute Respiratory Syndrome coronavirus 2) remains ambiguous, genetic evidence suggests that bats are a reservoir host of the virus and pangolins may be the possible intermediate. Novel coronavirus has crossed the species barrier to infect humans and other animal species. Also infected humans can expedite reverse-zoonotic transmission to animals. Considering the ever evolving interconnections among human, animals and ecosystems, zoonotic potentialities of COVID-19 is reviewed to include trans-disciplinary activities of public health veterinarians in this "Era of Zoonoses."

COVID-19 is certainly a newly emerged zoonosis; though it is not yet understood properly. A big uncertainty envelops the world since December, 2019. COVID-19 (SARS-CoV-2) is the latest addition to the seven corona viruses found in humans and experts said that all the viruses came either from bats or mice or turkey or cow or pig or cats or dogs(Ye et.al, 2020). However, pneumonitis with unknown etiology were detected in Wuhan, China in December 2019, and shortly after, a novel coronavirus (sever acute respiratory syndrome coronavirus 2[SARS-CoV-2]) was identified as the causative agent. The virus spread rapidly to other parts of China and many other countries. Despite tremendous efforts to contain the disease, the World Health Organization declared the spread as a global pandemic, referred to as coronavirus disease 2019 (COVID-19)(Chen et.al, 2020).

As Public Health Veterinarian, we have been succeeding the development of COVID-19 in order to identify and discuses the followings and hence, the present review provides a knowledge update on these points:-

- Zoonotic transmission from animal to human
- Potential risks to animals
- Intra and inter species transmission between animals
- Possible reverse zoonotic dissemination from human to animal
- Development of suitable animal models necessary for the evolution of vaccine and anti viral drugs or drug regimen.

The possible mechanisms of transmission of SARS-CoV-2 include droplet inhalation; direct contact with the person infected by the disease, direct contact with the persons exposed to the virus (animal/meat handlers), infected animals, and indirect (fomites) contact transmission (Pal, et.al., 2020).

SARS-CoV-2 initiates infection via the binding of its spike (S) protein to a specific cellular receptor. The human receptor for SARS-CoV-2 is angiotensin-converting enzyme-2 (ACE2). A bat coronavirus (bat-

CoV), Ra TG13, has been isolated from Yunnan, China and its whole genomic sequence is 96% identical to that of SARS-CoV-2. Another coronavirus was isolated from Malayan pangolins, and the whole genomic sequence of the pangolin coronavirus (pangolin-Cov) is 91.02% and 90.55% identical to that of SARS-CoV-2 and bat-CoV-RaTG13, respectively (Zhang et.al.,2020). Although the bat-CoV-RaTG13 sequence is closest to that of SARS-CoV-2 (96% similarity), the receptor-binding domain (RBD) of the pangolin-CoV S protein is more similar to that of SARS-CoV-2 than that of bat-CoV-RaTG13. Five key amino acids essential for binding to the human receptor are identical between pangolin-CoV-RaTG13 may not efficiently infect humans (Ruffel, 2020). This RBD on the S-protein is used by the SARS-CoV-2 to attach to the Angiotensin converting enzyme-2 (ACE2) receptors present on various human cells, including the cells of respiratory tract. These findings suggest that SARS-CoV-2 may have evolved from pangolin-CoV and adapted to humans via natural selection. Further studies are needed to substantiate that pangolins are an intermediate host. According to another school of thought, the potential reservoirs for SARS-CoV-2 are bats. Protein sequences alignment and phylogenetic analysis showed that turtles, pangolin, and snakes have been identified as alternative intermediate hosts for SARS-CoV-2 (Guo et. al., 2020).

With wide host adaptability, and the ability to undergo mutations and genetic recombination, CoVs may continue to pose a potential threat to public health. The microbes, with their ability to undergo genetic variations, and survive inside different hosts may evolve into a novel species as evidenced from our previous experience with the Influenza virus. The novel CoV may have evolved in a similar fashion. Studies so far have noted that there may be two types of SARS-CoV-2 strains (L, and S type) circulating throughout the world. But the good news is that the SARS-CoV-2 is comparatively stable than its predecessor SARS-CoV in terms of the mutations and the genetic variations. SARS-CoV and its elimination from the humans were noted to be due to the 29-nucleotide deletion that occurred during the human-human transmission (Tang et.al., 2020). SARS-CoV-2 has wide host adaptability that includes humans, birds, livestock, masked palm civets, mice, dogs, cats, camels, pigs, chickens, and bats, wherein they typically cause respiratory illness(Schwartz & Graham, 2020). Companion animals like Cats and Dogs are often in close contact with humans, and thus, it is important to determine their suspectibility to SARS-CoV-2. COVID-19 has been reported in two dogs in **Hong Kong** that live with COVID-19 human patients have become infected and tested positive. One of the dogs developed specific antibodies against SARS-CoV-2 and scroconverted, indicating an active infection. Canine cases of COVID-19 were also reported in the Netherlands and US (Hang & Dong wan, 2020). A family in **North Carolina U.S.A.** experienced mild COVID-19 symptoms, and their pug also showed mild symptoms with inappetence. All three family members tested SARS-CoV-2 positive, and the virus was detected in the dog. The family owned two dogs and a cat, but only one dog tested positive. In Netherlands, a COVID-19 patient owned a dog and three cats, and the dog was suffering severe breathing problems. This bulldog tested SARS-CoV-2 positive and was euthanized due to the illness. The three cats also developed specific antibodies for SARS-CoV-2. All four animals appeared to have contracted the virus from their COVID-19 owner. In contrast to dogs, cats appear to be highly susceptible to the virus. **In Belgium**, a cat living with its COVID-19 owner became clinically ill, exhibiting respiratory problems accompanied by diarrhea and vomiting. The specific viral sequence of SARS-CoV-2 was detected in the feces and gastric vomitus of the cat, and that sequence was identical to that of the cat owner, indicating the occurrence of reverse zoonotic transmission of SARS-CoV-2 from human to animal. In **Wuhan**, 102 cats were tested, and 15% of them were scroconverted, indicating SARS-CoV-2 exposure of cats from either people or other cats (Zhang et.al., 2020). An additional case of COVID-19 in cat was reported in **Hong-Kong**. This cat did not show any clinical signs, but oral, nasal, and fecal samples tested were found to be positive. In the **US**, SARS-CoV-2 positive cats have been reported in two separate areas in the state of New York (USDA, 2020a). A veterinarian tested one cat after it exhibited mild respiratory symptoms. The cat was SARS-CoV-2 positive, but no one in the household was found as

positive. This cat may have contracted the coronavirus outside of the house-hold from a person with COVID-19. In another case, a cat with respiratory symptoms and living with a COVID-19 patient tested positive. These data clearly show that cats are susceptible to SARS-CoV-2 and may contract COVID-19. A controlled experiment was conducted to support the observation showing that cats are susceptible to COVID-19 (Shi et.al., 2020). The results of that experiment showed that cats are highly susceptible to SARS-CoV-2, with the viral sequence detected in the nasal turbinates, soft palate, tonsils, and small intestine. The infections led to massive lesions in their nasal passages; adolescent cats were especially vulnerable to COVID-19. Notably, the coronavirus spread from infected cats to uninfected cats via respiratory droplets. Feline cases of COVID-19 have additionally been reported in **Spain**, **France**, and **Germany** (https://www.oie.int/scientific-expertise/specific-information-and-recommendations/questions-and-answers-on-2019novel-coronavirus.). So, these companion animals are the victims of the reverse zoonosis, though rarely infected; but not infectious.

Following table represents reverse zoonoses in different animals worldwide, as reported:-

Table: COVID-19, a reverse zoonosis

Type of animal	Date	Country
Dog (Pug)	01.07.2020 (11.04.2020)	USA (North Carolina)
Tiger	06.04.2020	USA
Lion	17.04.2020	USA
Cat		USA (New York)
Cat	28.07.2020	UK
Mink	17.06.2020	Denmark
Cat	26.05.2020	Russia
Cat	13.05.2020	Germany
Cat	11.05.2020	Spain
Cat	08.06.2020	Spain
Cat	02.05.2020	France
Cat	12.05.2020	France
Mink	26.04.2020	The Netherlands
Cat	28.03.2020	Belgium
Dogs (iPom & GSD)	09.03.2020	Hong Kong

Among farm animals, minks at two breeding farms in the Netherlands showed various symptoms including respiratory illness and were found to have been infected with SARS-CoV-2. The minks were likely to have contracted the virus from farm staff, and mink farms were subsequently placed under quarantine (https://www.oie.int/scientific-expertise/specefic-information-and-recommendations/questions-and-answers-on-2019novel coronavirus/). A subsequent study suggested that an infection took place from minks to humans, and cats played a role in the spread of the virus between farms, implicating possible occurrence of **interspecies transmission** (cat to mink) and secondary zoonotic transmission (mink to human) of SARS-CoV-2. In experimental infections, pigs, chickens, and ducks remained SARS-CoV-2 negative and did not develop any clinical signs. Viral sequences were not detected in any swabs collected from virus-inoculated animals, and the animals remained sero-negative for two weeks post-infection. Therefore, pigs, chickens, and ducks seem not to be susceptible to SARS-CoV-2. Regardless, a wider range of farm animal species needs to be examined to assess the risks of SARS-CoV-2 infection and to identify possible impacts of COVID-19 on the agricultural and food supply industries.

It is logical to speculate that secondary transmission may occur from COVID-19 animals to humans, despite no direct evidence showing whether transmission from cats to humans can occur. Owning a pet cat is commonplace, thus, the possible transmission of SARS-CoV-2 between owners and cats or between cats, for example, in a veterinary hospital setting, is of concern. Experimental infections under varying conditions, wide-ranging surveillance using specific diagnostic tests, and collection of epidemiological data will help enlighten the role of companion animals in the spread of COVID-19. Among wildlife, lions and tigers are susceptible to SARS-CoV-2. Four tigers and two lions at the Bronx Zoo in **New York City**, developed clinical symptoms associated with respiratory illness, and testing confirmed as SARS-CoV-2 positive (USDA, 2020b). The big cats had been exposed to a zookeeper who was COVID-19 positive and actively shedding the virus, indicating occurrence of reverse-zoonotic transmission.

Following schematic diagram represent Zoonotic transmission of COVID-19:

Fig: Zoonotic Transmission of COVID-19
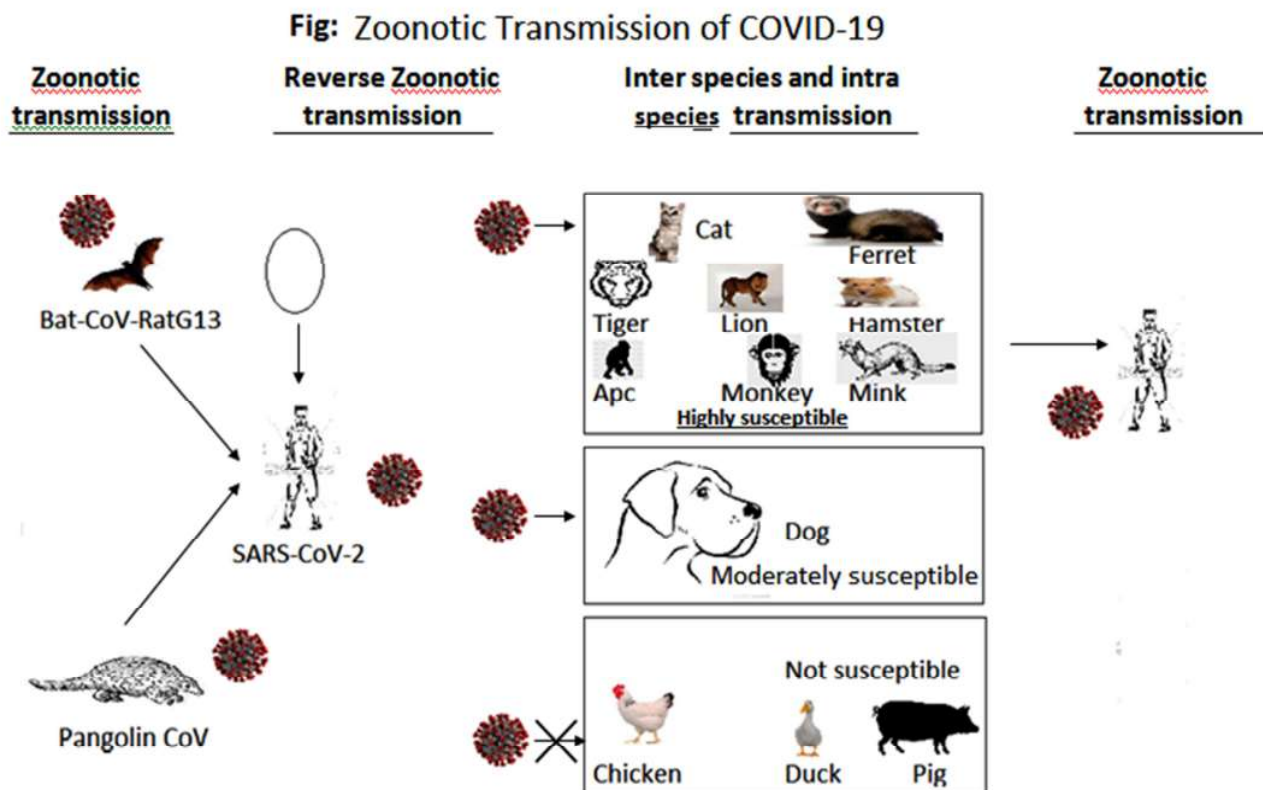
Susceptibility to COVID-19 has been evaluated in laboratory animals, companion animals, and farm animals in attempts to identify animal models of SARS-CoV-2 infection. Upon infection, susceptible animals may not show symptoms, or even if they do exhibit clinical signs, their symptoms may not match those of COVID-19 in humans. Thus, the establishment of an appropriate **animal model** is crucial. Among laboratory animals, ferrets, golden Syrian hamsters, and monkeys have been shown to be susceptible to SARS-CoV-2, and they develop clinical symptoms (Cohen, 2020). Ferrets are a good model for human influenza, particularly as they can sneeze, spreading the virus in the air. SARS-CoV-2 infects ferrets, but fatalities have not been observed (Kim et.al., 2020). The virus was detected in ferret nasal washes, saliva, urine, and faces, and airborne transmission was observed. However, viral replication in other organs was undetectable and did not lead to symptoms other than and increase in body temperature. A monkey-based study was conducted using four rhesus macaques (Munater, et.al., 2020). These monkeys were successfully infected, and viral replication occurred in the nose, pharynx, lung, and gut. The infections caused weight loss and a moderate level of interstitial pneumonia with lesions and lymphocyte infiltration occurring in the

lung, which were confirmed by H&E staining and immunohistochemical analysis. This monkey study also showed the presence of an antibody response against the virus. Using this monkey model, the efficacy of **remdesivir**, a polymerase inhibitor, was examined (Williamson et.al., 2020). Recovery in remdesivir-treated animals was significantly better than that in control animals. Only one of six treated animals had mild breathing difficulty, whereas all animals in the control group showed breathing difficulty. In treated monkeys, the viral load in the lungs was significantly lower, and lung damage was significantly milder than those in the control animals. The main protease, Mpro in SARS-CoV-2 is a viable drug target due to its role in the cleavage of the virus polypeptide. Feline infectious peritonitis, a fatal feline coronavirus infection was successfully treated with a prodrug GC-376—a dipeptide based protease inhibitor. GC 373 and GC 376 are found to be potent inhibitors of SARS-CoV-2 replication in cell culture and this research findings lays the framework for their use in human trials for the treatment of COVID-19 (Wayne et.al., 2020). Regardless of the animal model used, a significant challenge in such studies is that the experimental infection must be performed in a biosafety level 3 facility.

The high susceptibility of the Felidae family (lions, tigers, jaguars, leopards, cougars, and cheetahs) and, in particular, monkeys to COVID-19 implies a potential for outbreaks in great apes (chimpanzees, gorillas, orangutans) in zoos, animals-holding facilities, primate research centers, and national wildlife parks. Risk assessments are needed to prepare for the conservation and protection of such animals.

A combination of ecological disturbance, deforestation like landscape changes, human behaviors, and public health factors contributes to the frequency of contacts between humans and wildlife, and such contacts pose a risk of exposure to transboundary animal viruses. Moreover, it is alarming that 75% of the 132 emerging human infections over the past 12 years have been caused by pathogens originating from animals and have the potential to create global problems (Das, 2019). As, we are living in the "Era of Zoonoses", the application of knowledge of 'Clinical Veterinary Public Health' is of immense importance (Das, 2017). It may be also mentioned that trained sniffer dogs are known for their extraordinary capabilities and skills with strong sense of smell. These sniffer dogs are used successfully to detect COVID-19 cases in Dubai and Helsinki airport. Hence, dogs can be trained to sniff out COVID-19 in patients for early warning measures (John Hopkin Univ., 2020) as a tool of Applied Veterinary Public Health (AVPH). Data and studies showed that detection of presumed COVID-19 cases achieved approximately 92% in overall accuracy. Also, the emergence of COVID-19 resulted in physical, mental and social human health hazard despite of economic and political problems; These mental or psychological and social health hazards often be mitigated by "Therapeutic Veterinary Public Health" (Das, 2015). the viruses can easily mutate, switch hosts, and adapt to a new host. In this period within the COVID-19 pandemic, societies and human behaviors are quickly changing, and to accommodate these changes, the customary roles of public health Veterinarians should unfold accordingly. This pandemic is like a world war between the virus and the science; except in this case, the whole world is on the same side and this war can be won by complete global collaboration collectively. COVID-19 pandemic perhaps acts as a catalyst to initiate One Health Surveillance (OHS) in many countries. Public Health Veterinarians should have significant roles in maintaining healthy ecosystems and protecting animals and humans from emerging and transboundary infections. Such roles should be based on the One Health framework, the application of which can reduce economic impacts on the livestock industry and food supply.

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ACUTE PANCREATITIS: AN UNADDRESSED CRITICAL CLINICAL CONDITION IN DOGS

Shivangi Sharma[1,*], Amita Tiwari[1], Devendra Gupta[1] and Rahul Sharma[2]

[1]Department of Veterinary Medicine, [2]Department of Animal Nutrition
College of Veterinary Science and Animal Husbandry, NDVSU, Jabalpur-482 001, MP, INDIA
*spshivi094@gmail.com

Introduction

Pancreatitis (inflammation of pancreas) develops when there is excessive activation of trypsin and other pancreatic proteases within the pancreas, which overwhelms local safeguards within the acinar cell. Acute pancreatitis refers to an acute inflammatory process of the pancreas, usually accompanied by abdominal pain and elevations of serum pancreatic enzymes in blood or urine. This syndrome is usually a discrete episode, which may cause varying degrees of injury to the pancreas, and adjacent and distant organs.

Canine acute pancreatitis is a relatively frequent disorder encountered in veterinary practice. While pancreatitis is generally regarded as a significant illness in dogs, its true incidence is unknown, since many dogs have subclinical or mild disease and recover within a few days without specific treatment. Patients presented to veterinarians with pancreatitis usually have the more severe form and their disease may result in death if their condition is not recognised and treated in time. Based on the patient's condition, it is classified as mild, moderate or severe, and non - fatal or fatal. Histopathological criteria for AP include pancreatic oedema and necrosis, infiltration of mononuclear and polymorphonuclear cells, peri-pancreatic fat necrosis and thrombosis, but without permanent disruption of the pancreatic architecture.

Acute pancreatitis (AP) is considered to be a reversible condition unless the initial triggers of disease persist, in which case chronic or recurrent inflammation develops **(Stevens _et al._ 2004).** AP is characterised histologically by neutrophilic inflammation or necrosis of the pancreatic or peri-pancreatic area with no fibrosis or exocrine atrophy present **(Kalli _et al._ 2009).** Clinically the differentiation between acute necrotising pancreatitis, AP without necrosis and chronic active AP (inflammation and fibrosis from a previous insult) is difficult. Therefore, veterinary clinical nomenclature usually relates to the severity and longevity of clinical signs rather than to the histological characteristics. The term mild AP should be used when there is no multisystem failure, whilst severe AP should refer to the presence of multisystem failure or development of complications that require a higher intensity of treatment.

Each pancreatic lobule is composed mainly of acinar cells that synthesize the digestive enzymes in the form of pro-enzymes and store them in zymogen granules. The pancreas of the dog usually has two ducts by which secretions are transported from the organ to the descending duodenum. The pancreas also contains endocrine tissue, the islets of Langerhans, accounting for one to two percent of the gland. The main function of the exocrine pancreas is the secretion by the acinar cells of a fluid rich in digestive enzymes involved in the initial degradation of proteins, lipids, and polysaccharides **(Kalli _et al.,_ 2009).**

Pathophysiology- Multiple and controversial pathogenic theories

Obesity has been suggested as a risk factor for the disease, and the disease has been reported as less severe when experimentally induced in lean dogs. Hyperlipidaemia, often grossly recognised

in patients with acute pancreatitis, may be the consequence of abdominal fat necrosis, or may potentially induce the disease. In the case of Minature Schnauzers, there is a possible link between the familial hyperlipidaemia recognised in the breed and the apparent increased incidence of pancreatitis.

Hypercalcaemia has also been recognised as a cause of pancreatitis in dogs. This may be spontaneous (malignancy, hyperparathyroidism, renal failure, hypoadrenocorticism, granulomatous disease, destructive bone disease), or may be iatrogenic (vitamin D toxicity).

Reflux of duodenal juice into the pancreatic duct can cause pancreatitis, but rarely occurs in a normal patient because of the anatomy of the duodenal papilla, which has a muscular sphincter and is also covered by a special piece of mucosa. Obstruction of the pancreatic ducts typically results in pancreatic atrophy and fibrosis.

Classification of pancreatitis
Pancreatitis is divided into four main groups
(1) Acute
(2) Relapsing acute (with clinical and biological restitution of the gland)
(3) Chronic relapsing (chronic pancreatitis with acute exacerbations)
(4) Chronic (with anatomical and/or functional residual damage to the gland) - This is defined as a continuing inflammatory disease of the pancreas, characterised by irreversible morphological change, and typically causing pain and/or permanent loss of function **(Sarner and Cotton, 1983).**
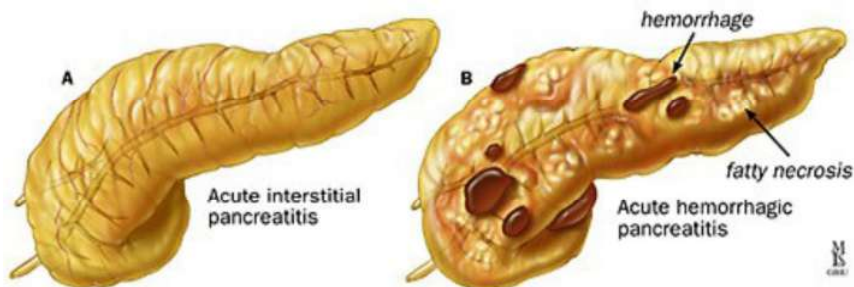
Figure. A, Acute interstitial pancreatitis; B, acute hemorrhagic pancreatitis.

Etiology
A number of factors are thought to contribute to acute pancreatitis in dogs which can be summarized as-
1. **Diet-** Obese dogs are at greater risk for pancreatitis, and recent consumption of a fatty meal is a common finding. Hypertriglyceridemia may induce pancreatitis through the toxic effects of fatty acids (generated by pancreatic lipase) on the pancreas. This may explain the development of pancreatitis after a fatty meal.
2. **Drugs-** Many drugs can potentially cause pancreatitis, but a few are more commonly associated with the disease. These drugs include seizure medications, such as potassium bromide; chemotherapy drugs, such as L-asparaginase and azathioprine; and antibiotics, such as tetracycline and the sulfonamides.
3. **Duodenal fluid reflux-** Duodenal contents (active pancreatic enzymes, bacteria and bile) reflux into the pancreatic ducts contributes to the development of AP
4. Trimethoprim-sulfamethoxazole has been thought to cause an immune mediated pancreatitis in dogs.
5. While steroids are a suspected cause of pancreatitis, no clinical evidence exists to support that claim. Steroids cause an elevation in serum lipase, but no associated pancreatic lesions

are found on biopsy. It is thought that steroids induce a lipase isoenzyme that may be of hepatic origin

6. A variety of diseases and metabolic disorders have also been shown to cause pancreatitis. In dogs with hepatobiliary disease and inflammatory bowel disease, it is possible for the inflammatory process to extend into the pancreas. Any surrounding inflammation— as in the case of neoplasia, for example—can cause pancreatic duct obstruction.

7. Other metabolic diseases such as **hypercalcaemia** and immune mediated disease, have also been implicated as causes of pancreatitis. Infectious etiologies have been described in cats; however, this is not a common cause of canine pancreatitis.

8. Some toxins have been associated with the disease, including cholinesterase inhibitors or cholinergics. The theory is that they stimulate hypersecretion. Other toxins include zinc and scorpion venom.

9. Other potential causes of pancreatitis include ischaemia (associated with shock, hypotension, or venous occlusion), infectious agents (uncommon, but viral, parasitic and mycoplasmal causes have been suggested such as babesia), or potentially immune mediated disease

Clinical Presentation of pancreatitis

. The clinical signs will vary with the severity of the disease process. In more severe cases common clinical signs may include anorexia, vomiting, weakness, abdominal pain, diarrhoea and obtundation. In some severe cases there may be more systemic clinical signs including fever, tachypnoea or dyspnoea, or signs of shock. Abdominal pain has not been reported as frequently in canine patients as in human patients. The abdominal pain may be manifest by the assumption of an unusual posture (**prayer position**), pain response upon physical examination. Some dogs may have respiratory distress, icterus, dehydration or signs of a bleeding disorder. Cardiac arrhythmias may also be apparent. Some severely affected dogs may be hypothermic.
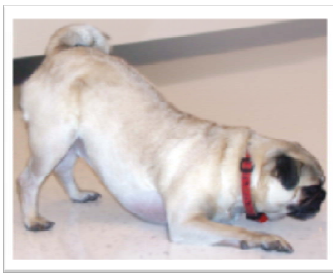
Dog exhibiting signs of cranial abdominal pain by adopting the "praying" posture, with head down and paws outstretched (Source- Internet)

Due to the vague clinical signs of AP, differential diagnosis should include all the conditions causing acute abdomen syndrome:

- Acute enteritis or gastroenteritis (*Parvo*-virus, syndrome of acute gastroenteritis)
- Exacerbation of infl ammatory bowel disease.
- Intestinal obstruction, particularly due to foreign bodies or intussusceptions
- Peritonitis
- Acute renal failure
- Acute hepatitis/acute hepatic failure
- Pyometra
- Ruptured abdominal organs
- Acute prostatitis

Complications of pancreatitis

The most commonly seen complications of AP are diabetes mellitus, diabetic ketoacidosis, intestinal obstruction, bile duct obstruction, renal failure and pulmonary oedema; rare complications of acute pancreatitis include pleural effusion, pancreatic abscess and pseudocyst formation, cardiac arrhythmia, disseminated intravascular coagulation (DIC), bacteraemia and acute respiratory distress (ARD).

Diagnosis of pancreatitis

The diagnosis usually depends on the analysis of history, clinical presentation, biochemical abnormalities, and imaging study and histopathology results
When examining the diagnosis, it is important to remember that none of the hematologic or serum chemistry changes will specifically indicate pancreatitis. Because the clinical signs vary for this disease, a complete history, physical examination and diagnostic workup are needed to distinguish pancreatic disease from the myriad of other disorders that may result in the same clinical signs. The diagnosis of acute pancreatitis requires two of the following three features:
(1) Abdominal pain consistent with acute pancreatitis (acute onset of a persistent, severe, epigastric pain often radiating to the back)
(2) Serum lipase activity (or amylase activity) at least three times greater than the upper limit of normal
(3) Characteristic findings of acute pancreatitis on transabdominal ultrasonography **(Theoni 2012)**

Serologic markers

Two serologic markers are available for pancreatic enzymes trypsin-like immunoreactivity (TLI) and pancreatic lipase immunoreactivity (PLI). Both are exclusively pancreatic in origin. While not perfect, these markers are currently the most sensitive and specific available for pancreatitis.

Trypsinogen activation peptide

During the inappropriate activation of trypsinogen to trypsin, TAP is released into the pancrease where it may diffuse into the intravascular or peritoneal space. Trypsin activation peptide (TAP) may be measured in the serum or urine of patients clinically suspected of having pancreatitis but its lability and limited availability limit its usefulness

Biochemical panel

Several biochemical changes, in addition to amylase and lipase elevations, can occur in patients with pancreatitis. Liver enzyme activities can be increased because of hepatocellular damage, whether from local pancreatic inflammation, the transport of pancreatic enzymes in the lymphatics, concurrent multiple organ failure or decreased hepatic perfusion due to hypovolemia. Serum biochemistry in dogs with acute pancreatitis includes-

- **Azotemia**- It may be pre-renal or renal.
- **Hepatic enzymes**- Alkaline phosphatase (ALKP) may be 2-15 times the normal and alanine amino-transferase (ALT) may be 2-5 times the normal value in dogs suffering from acute pancreatitis **(Watson, 2004).**
- `Hyperbilirubinaemia-` There may be 2-5 time increase in bilirubin in dogs.
- `Hyperglycemia and hypoglycaemia-` Hyperglycemia is a common finding in dogs with pancreatitis. This may be due to stress hyperglycemia or concurrent diabetes mellitus. Hyperglycemia is not a diagnostic marker for pancreatitis, but may be suggestive. Hypoglycaemia may also be seen in severe pancreatitis due to concurrent liver disease or sepsis.

- `Hypocalcaemia-` Hypocalcaemia occurs from low albumin, intracellular shifts of calcium and deposition of calcium in saponified peri-pancreatic fat.
- **Hypercholesterolemia-** commonly identified in dog with acute pancreatitis.
- **Hyperlipemia-** it may be associated with diseases such as endocrinopathies and hepatic lipidosis that may affect dogs **(Mansfield *et al*., 2001).**
- `Hypoalbuminaemia-` It may be a consequence of systemic inflammation.
- `Hypoproteinaemia-` may be a result of dehydration.
- **Hyperamylasemia and hyperlipasemia** are both associated with pancreatitis. Any decrease in glomerular filtration rate (GFR) or renal function will result in elevated amylase and lipase values, often as high as 2.5 to 3 times the normal values. It is important to understand that elevated amylase and lipase are not diagnostic for pancreatitis, and that normal values do not rule out pancreatitis. Elevated results should always be interpreted in light of concurrent dehydration or renal disease.

Hematological panel

The complete blood count results in pancreatitis are non-specific and depend upon the severity of disease. One of the more common findings in dogs with severe pancreatitis is neutrophilia with a left shift, but a stress leukogram with only mild neutrophilia and no left shift may occur in mild cases.

Sepsis and severe systemic inflammation can result in neutropenia with a degenerative left shift. If DIC is present, then platelets may be decreased. Dogs with dehydration will have elevated hematocrits and total red blood cell numbers.

Imaging studies

The most commonly used imaging techniques in veterinary practices are radiography and ultra-sonography.

Radiography

Abdominal radiographs have limited use in diagnosing pancreatitis, but they are helpful in ruling out other causes of vomiting, such as gastrointestinal foreign bodies. However, a few radiographic changes may be suggestive of, but not diagnostic for, pancreatitis.
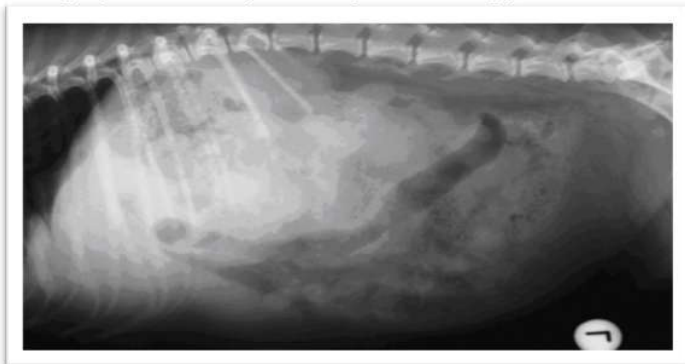
Lateral abdominal radiograph of a dog with pancreatitis. There is a mass effect in the cranial abdomen, caudal to the stomach, as well as ventral displacement of the gas-filled descending duodenum **(Kalli *et al.*2009)**

Ultrasonography

Ultrasonography is more sensitive than radiography in diagnosing pancreatitis. However, finding the pancreas and accurately evaluating it can be difficult. Some of the more common ultrasonographic findings are an enlarged pancreas, hypoechoic mass effect to the pancreas, hyperechoic peri-pancreatic fat and fluid accumulation around the pancreas.
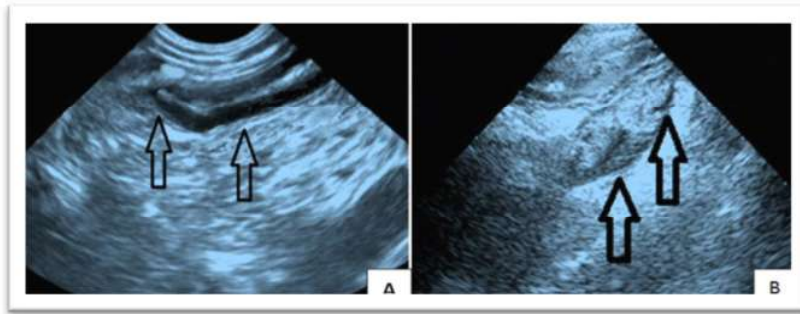
Ultrasound examination: A. Regular normal dog pancreas, contour and no visible nodules.
B. Pancreatitis of a dog, appear as hypo echoic nodules. 2.5 cm, changes thickening of the pancreas.
(Rahmoun and Fares, 2018)

Biopsy/histopathology

The last diagnostic test to consider is biopsy with histopathology. This remains the gold standard for the diagnosis of pancreatitis. Biopsy can be performed surgically, or a fine-needle aspirate can be taken with ultrasound guidance. Aspiration may help rule out the presence of pancreatic neoplasia. Surgical biopsy of the pancreas does not induce pancreatic inflammation or necrosis if the blood supply is not disrupted **(Center, 2004).**

Treatment

General treatment principles involve replacing fluid losses, maintaining hydrostatic pressure, controlling nausea and providing pain relief. Specific interventions recently advocated in human medicine include the use of neurokinin-1 antagonists for analgesia and early interventional feeding.

Fluid therapy

Intravenous fluids are the mainstay of therapy for pancreatitis. Initially fluids should correct dehydration over the first 12–24 hours, while also meeting maintenance needs. The fluid rate should be adjusted frequently to account for ongoing losses (e.g., vomiting, diarrhea, ascites) and to correct fluid, electrolyte, and acid-base imbalances. If needed, colloidal support can be given in the form of fresh frozen plasma, hetastarch, or dextrans (10–20 mL/kg/day).

Plasma

Plasma will provide α-macroglobulins to scavenge activated proteases within the serum; it also provides clotting factors and is indicated if there is evidence of disseminated intravascular coagulation (DIC). Purported benefits of plasma transfusion in treatment of AP include replacement of circulating α-macroglobulins, coagulation factors and anti-inflammatory factors **(Mansfield and T. Beths; 2015)**

Analgesia

Pain is the common clinical sign of acute panceatitis and analgesic therapy should be considered for abdominal pain in every animal with suspected or confirmed pancreatitis. Intravenous or subcutaneous opioids are typically used but alternatively, intra-peritoneal infusions of lidocaine or bupivacaine mixed with sterile saline can be administered. Options for outpatient pain control include fentanyl patch, tramadol or butorphanol. Pethidine, fentanyl or butorphanol do not increase the pressure in the bile duct to the same extent as seen with morphine and can therefore be used. Fentanyl, a synthetic opioid 100 times as potent as morphine, can be administered iv and transdermally (fentanyl patches). Fentanyl has a profound negative effect on gastrointestinal motility, and so is seldom used in the management of AP.

Suggested doses for analgesic agents for dogs with acute pancreatitis

Drug	Dose	Route	Frequency
Fentanyl	2-10 µg/kg/h	IV	CRI
Morphine	0.1-1 mg/kg	IV, IM	Prn
Butorphanol	0.1-1 mg/kg	SQ	q6h
Hydromorphone	0.1-0.2 mg/kg	IV, IM, SC	q6-8h
Methadone	0.1-0.5 mg/kg	IV, IM, SC	q6-8h
Ketamine*	10-20 µg/kg/min	IV	CRI
Lidocaine*	30-50 µg/kg/min	IV	CRI

(IV- Intravenous, IM- Intramuscular, SC- Subcutaneous, CRI- Continuous Rate Infusion)

Anti emetics

In dogs affected with pancreatitis, vomiting is both centrally and peripherally mediated. Maropitant, an effective anti emetic agent blocks both centrally and peripherally mediated emesis by blocking neurokinin1 (NK1) receptor and substance.

Suggested doses for anti emetics for dogs with acute pancreatitis

Drug	Dose	Route	Frequency
Chlorpromazine	0.2-0.5 mg/kg	IV, IM, SC	q8h
Metoclopramide	1-2 mg/kg 0.1-0.4 mg/kg	IV IM, SC	CRI q24h q8h
Ondansetron	0.1 mg/kg	IV	q8-12h
Maropitant	1 mg/kg	SC, IV	q24h

Gastric acid reduction

Proton pump inhibitors are the preferable agents to increase the gastric Ph for gastric mucosal health by preventing the development of gastric mucosal ulcerations, also increased gastric Ph decreases exocrine pancreatic stimulation.

Corticosteroids

Corticosteroids may exert multiple positive benefits in AP by inhibiting the release of pro-inflammatory mediators, decreasing sequestration of neutrophils in the pulmonary vasculature, as well as reducing adhesion of primed neutrophils to the endothelial surface of pulmonary vasculature, pulmonary vascular permeability, and release of elastase and free radicals from adherent neutrophils.

Conventional treatment summary

• Parenteral administration of electrolyte solutions (44-66 ml/ kg + % dehydration x B.W. x 1000) ml
• Correction of metabolic acidosis (bicarbonate 1-2 mmol/kg IV)
• Correction of hypokalaemia (Scott's scale)
• Antiemetic agents (metoclopramide @ 0.2 - 0.4 mg/kg SC every 6 to 8 hours or 1 mg/kg/day by continuous intravenous infusion, ondansentron @0.05 mg/kg IV every 8 to 12 hours, maropitant @1mg/kg SC every 24 hours)
• H2 blockers (ranitidine @ 2-4 mg/kg IV every 12 hours)
• Antibiotics (enrofloxacin @ 2.5-5 mg/kg SC every 12 hours, trimethoprim-sulphathiazine @15 mg/kg IV every 12 hours) *
• Plasma or blood transfusion (10-20 ml/kg B.W.)
• Analgaesics (meperidine hydrochloride @5-10 mg/kg IM or SC every 2 to 4 hours, butorphanol @0.2 - 0.4 mg/kg SC every 6 hours, morphine @ 0.5-2 mg/kg SC or IM every 3-4 hours, diadermic phentanyl patches)

* The routine use of antibiotics in canine AP is not recommended since infectious complications are rather uncommon. However, in cases with evidence of pancreatic infection or in cases of AP failing to respond to supportive measures, antibiotic use is justified.

Nutrition

The vomiting patient can be held NPO (fasting) for 24–48 hours with subsequent gradual reintroduction of a low-fat diet when vomiting subsides. While NPO does provide a "rest" for the pancreas, most veterinary patients have been anorexic for >48 hours at the time of presentation, thus further withholding of nutrition is likely detrimental. Based on the medical consensus, it is recommended that dogs with mild AP (no systemic complications) be fasted until they are able to eat voluntarily, unless they have reached 5 days of anorexia (including the pre-hospital period), in which case enteral feeding should be initiated.

There is no current recommendation for the type of food to be administered in this acute setting. Although studies have shown no difference in dietary fat content on pancreatic secretion in healthy dog avoidance of high-fat diets in dogs with AP is logical as many of the animals are hyperlipidaemic.

Prognosis

The prognosis is variable with the severity of the disease, and can be difficult to predict. Dogs with fulminating acute disease may still die despite aggressive therapy, whereas others may survive. Mild pancreatitis often responds to symptomatic treatment and has a good prognosis, whereas severe pancreatitis requires aggressive therapy and prognosis is guarded. Early diagnosis and treatment of the disease and the absence of systemic complications are factors that result in a better outcome.

Conclusion

Patient prognosis is guarded in many cases of pancreatitis. However, rapid diagnosis and implementation of appropriate therapy early in the course of disease will reduce patient morbidity and mortality.

Acute pancreatitis has been intensively studied for centuries. Many causes of acute pancreatitis have been discovered, but its pathogenic theories are multiple and controversial. The true nature of acute pancreatitis still remains to be elucidated. The causes of acute pancreatitis are various, and its mechanism is common. Once the hypothesis is confirmed, traditional therapeutic strategies against acute pancreatitis may be improved, and decompression of pancreatic duct pressure should be advocated in the treatment.

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Status of Brucellosis in India: A review

Maansi and A. K. Upadhyay
Department of Veterinary Public Health and Epidemiology
College of Veterinary and Animal Sciences
G. B. Pant University of Agriculture and Technology, Pantnagar
Uttarakhand-263145

Abstract

Brucellosis is one of the most important diseases coming from dairy animals to human in India. Although, brucellosis is a disease affecting a wide range of animal species including the major food-producing animals as cattle, sheep, goats, pigs. Other species such as bison, buffalo, camels, dogs, horses, reindeer and yaks are also affected and act as a significant local source of infection in some regions. Infections in marine animals (dolphins, porpoises and seals) have further escalated the chances of transmission of the disease in other susceptible hosts. Till date, none of the prevention and control measures have been helpful in the eradication /elimination of this disease. With the initiation of' National Animal Disease Control' programme launched recently which aims at controlling this disease through proper vaccination, one can hope to curtail the problem to some extent.

Introduction

Ten host specific species *Brucella* are: *B. abortus* (cattle), *B. canis* (canids), *B. ceti* (cetaceans), *B. melitensis* (sheep and goats), *B. microti* (*Microtus arvalis*), *B. neotomae* (*Neotoma lepida*), *B. ovis* (sheep), *B. pinipedialis* (pinnipeds), *B. suis* (pigs) and *B. inopinata* (isolated from a human patient who had undergone breast implant)(Whatmore, 2009 and Minharro *et al*.,2013). Transmission of *Brucella* among animals mainly occurs through contact following an abortion. Contaminated pasture or animal houses are responsible for the spread of the organisms through ingestion. Inhalation, conjunctival inoculation, skin contamination and udder inoculation from infected milking cups account as other modes. In calves, pooled colostrums for feeding newborn calves serve as a source. Artificial insemination procedures transmit the disease but sexual transmission plays a little role in bovines. The sharing of male breeding stock also promotes transfer of infection between farms. Sexual transmission probably plays a greater role in the transmission of *B. melitensis* in sheep and goats along with *B. suis* in swine and *B. canis* in dogs. Other risk factors involves commingling of different flocks and herds, unscreened animal purchase entry into the farm, transhumance of summer grazing, mingling of animals at fairs and closed space animal housing. As the disease is zoonotic in nature, transmission to humans takes place by eating or drinking unpasteurized/raw dairy products, inhalation and can also enter wounds in the skin/mucous membranes through contact with infected animals. Ingestion of raw milk and occupational exposure are the key modes. Person-to-person spread of brucellosis is extremely rare. Infected breast feeding mothers may also transmit the infection to their infants. Sexual

transmission has been rarely reported. While uncommon, transmission may also occur via tissue transplantation or blood transfusions.

Brucellosis is of wide economic concern too as it causes huge economic losses. In India, brucellosis in livestock is responsible for a estimated loss of US $3.4 billion per year out of which cattle and buffalo accounted for 95.6% of total losses due to abortions, temporary infertility and sterility in adult animals (Singh *et al*, 2015).

Indian scenario

India is a land of diversity with a wide range of physical features. From cold mountains to arid deserts, vast plains, hot and humid plateau and wide sea shores and tropical islands, the physical features of India cover every terrain. The country has largest livestock numbers in the world. The total livestock population consisting of Cattle, Buffalo, Sheep, Goat, Pig, Horses & Ponies, Mules, Donkeys, Camels, Mithun and Yak is approximately 512.05 million according to 19th Livestock Census (2012). One of the primary aims of livestock development programme undertaken by the Government of India is to increase milk and meat production through sustainable disease control programmes.

Epidemiological investigation of brucellosis generally relies upon the sero-prevalence studies. Animals with history of reproductive failure and abortion are generally screened for brucellosis by the Rose Bengal plate test (RBPT), serum tube agglutination test (SAT) and enzyme linked immunosorbent assay (ELISA) kits. Bovine brucellosis is endemic in all the states of India and appears to be on the increase in recent times, perhaps due to increased trade and rapid movement of livestock. Current management practices and herd structure also allow for endemic brucellosis. The preponderance of natural bull service in rural India, especially in buffalo, is perhaps may be an important factor in the maintenance and spread of infection. However, a National Animal Disease Control Programme for brucellosis control is being implemented in the country with the aim to eradicate the disease through vaccinations. Yet another

Prevalence in India

Earlier reports of serological evidence has also by enlarge suggested the disease to be highly endemic in most parts of India (Chauhan *et al*., 2000). Among the states, Punjab shows the highest disease prevalence probably owing to the constant screening programme running in the state and the number of bovine population. On the other hand, the seroprevalence rate ranged from 6.6% (123/1860) in central state of Madhya Pradesh (Mehra *et al*., 2000) to 60% in a northeastern state of Assam (Chakraborty *et al*., 2000).

In India, brucellosis was first recognized in 1942 and is now endemic throughout the country. Rapid and easy travel and trade further has the potential to increase the endemicity. The disease is reported in cattle, buffalo, sheep, goats, pigs, dogs and humans. The long-term serological studies have indicated that 5% of cattle and 3% of buffaloes are infected with brucellosis (Renukaradhya, 2002).

Progress reports of monitoring programs from 2012–2013 by the Indian Council of Agricultural Research also estimates that the current national seroprevalence of brucellosis in cattle is roughly 13.5% and at a stable, endemic equilibrium (Rahman, 2013). The true epidemiological status of the disease in the country remains a concern owing to the absence of proper laboratory facilities, lack of awareness, under-reporting along with improper recording of the history of the disease. Buffalo keepers were totally unaware of the disease and the vaccine available for the disease (Kant *et al.,* 2018). Most of them drink raw milk, sleep in cattle sheds, do not isolate sick cattle, do not test buffaloes blood for any disease before purchasing them, apply intrauterine medication with bare hands to buffalo after abortion of foetus, never clean their cattle sheds with a disinfectant and believe that they can only acquire skin infections from cattle.

Human brucellosis

Human brucellosis is reported from most parts of the country and is closely related to animal husbandry activities (Hemashettar and Patil, 1991). Several reports indicate it to be a quiet common disease in India. Numerous researches report the serological evidence of human brucellosis ranging between 0.9 and 18.1% in the country (Kumar *et al.,* 1997).

Humans in India live in close proximity with the animals thereby stand at a greater risk to zoonotic infections. As brucellosis in animals is prevalent throughout the country, cases of human brucellosis are witnessed regularly with B. *melitensis* and *B. abortus*, of which the *B. melitensis* exhibits higher virulence and with much severe and extended illness with harsh consequences. Mathur (1985) isolated 53 strains of *Brucella* confirmed as B. *melitensis* biotype-1. He also concluded that brucellosis occurred more frequently in villages than in cities. It was also inferred that most human infections occurred with *B. melitensis* in the geographic regions where *B. abortus* was primarily responsible for bovine brucellosis, indicating the role of sheep and goats as the source of infection. In addition isolation of other *B. melitensis* biotypes-2 and 3 along with biotype-1 was reported from Delhi and Haryana (Sen and Sharma, 1975). Moreover, Hemashettar *et al*., (1987) recorded the presence of *B. melitensis* biotype-1 infection in a patient who did not show any agglutinating antibodies.

Several risk groups have been screened and have been found to be significantly associated with a risk of picking the infection. In India, abattoir workers, laboratory personals, dairy farmers and veterinary clinicians have been studied extensively for the presence of the disease. A much higher prevalence has been initiated in abattoir workers (Barbudhe *et al*, 2016). Studies on veterinarians, para-veterinarians and farm attenders revealed 25% infected in New Delhi (Kumar *et al*., 1997b); 21% in Goa (Barbuddhe and Yadava, 1997); 6.8% in Assam (Hussain *et al.*, 2000); 9.7% Maharashtra (Mohanty *et al*. 2000) and 6.8% in Orissa (Kumar et al,1997b). The study by Thakur and Thapliyal (2002), revealed a prevalence rate of 4.97% in samples obtained from persons exposed to animals. An overall prevalence recorded was 7.04% in personnel engaged in veterinary health care in Karnataka, India (Shome *et al*., 2017). The study also indicated high brucellosis prevalence of 16% in para-veterinarians and animal handlers compared to 5-6% in veterinarians and artificial insemination workers. The association of Para-veterinarians, animal handlers and veterinarians (*p*-value < 0.05) was reported to be significant in comparison

artificial insemination workers and veterinary students. Another study in Punjab during 2012-13 revealed maximum in vet para-clinical staff (25.28 %) followed by dairy workers (16.10%) and veterinarians (11.01 %). Proch *et al.,*2018 observed that in India, the risk is higher in para-veterinary staff than veterinarians and in those who have been practicing for a longer period of time The seroprevalence rates have been recorded to be as high as 17-34% in high-risk groups like abattoir workers, veterinarians and animal attendants (Appannanavar *et al*., 2012). High prevalence among butchers and attoabir workers was reported in Delhi. Around 5.31% of animal handlers were positive for Brucella agglutinins (Pandit and Pandit, 2013).

Human brucellosis is characterized by various symptoms especially pyrexia of unknown origin (PUO). A prevalence of 0.8% - 6.8% from different areas has been observed in persons complaining of PUO (Sen *et al*., 2002). Shome *et al.* (2017) recorded intermittent fever to be the most predominant symptom (71.62%) followed by spondyloarthropathy (52.70%), epididymo-orchitis (12.16%) and problem of infertility (8.10%). A 10 year study conducted in Chandigarh on persons with PUO reported 9.94% prevalence on serological basis. However, Barbuddhe *et al* (2000) reported maximum number of positives in patients with spondylitis followed by acute polyarthritis. Fever and upper back pain were also assessed as significant predictors for both acute and chronic forms of brucellosis, respectively (Patra *et al*., 2018). Noteworthy association [$P < 0.0001$] was also established between fever, joint pain, low backache, and fatigue and significant tube test titers, whereas no association was found between weight loss, headache, and sleep disturbance((Mangalgi *et al*., 2015). About 4.2% women with abortion were reported by Randhava *et al.* (1974) to possess *Brucella* agglutinins.

Extensive studies related to age group have been performed in Karnataka. In a study on children in Bijapur, Mantur *et al.,* (2004) observed a prevalence of 1.6% with a Standard Tube agglutination titre of $\geq$ 1:160 while a prevalence of 1.8% was observed in adults in the same region (Mantur *et al.* (2006).Since then, several reports of human brucellosis from the same region have been reported (Tikare *et al.,* 2008). Children and young adults were most commonly affected in Karnataka rural area as compared to the persons beyond 60 years ((Mangalgi *et al*., 2015). High brucellosis seroprevalence between 6.75 -8.90% was observed by Shome *et al.,* 2017 in persons between 21-40 years of age. Regarding sex association, higher percentage of infection in female children (14.3%) was observed compared to male children (10.9%) (Dutta *et al*., 2017). This was in accordance with Patil *et al.* (2016) who observed that the median age of the patients with brucellosis was 31 years in his study and males outnumbered females unlike Dutta *et al.* (2017).

The disease is prevalent in almost all the states/cities of the country with wide variation. Of all, the Punjab reports the highest (26.6%) cases of human brucellosis.A prevalence of 0.8% in Kashmir, 0.9% in Delhi, 6.8% in Varanasi, 8.5% in Gujarat and Belgaum, 11.51% in Andhra Pradesh, 19.83% in Maharashtra. Patil *et al*. (2016) reported disease from Gadag (21.1%), Haveri (17.4%) and Koppal (18.5%) districts of Karnataka. Thus systemic review suggests that the states like Punjab, Orissa, Andhra Pradesh, Rajasthan, Maharashtra, Gujarat, Uttar Pradesh, Uttarakhand and Goa have endemicity of the disease.

In cattle

The two *Brucella* species of main concern in India are *B. melitensis* and *B. abortus*. *B. melitensis* is concern with goats and sheep and related animals and most virulent for man. *B. abortus* is the dominant species in cattle and *B. suis* is mainly confined to pigs. In India, different B. abortus biotypes (types-1, 2, 4, 6 and 9) have been isolated from cattle. *B. abortus* was also isolated from buffalo and from goat and sheep. *B. melitensis* biotypes 1 and 3 have been isolated from goats and sheep and cattle. *B. suis* may also be present in cattle, buffalo and goats. Though *B. melitensis* is more infectious to man than *B. abortus* and in general is the dominant causative agent of brucellosis, disease caused by infection with *B. abortus* is indistinguishable from that by *B. melitensis* and may be equally severe (Smits and Kadri, 2005)

Brucella biotypes have been observed to have certain dominancy over a region (Sen and Sharma, 1975) such as, *B. abortus* biotype-1 appears to be the predominant biotype (21 out of 39) in most parts of the country, followed by *B. abortus* biotype-3 (8 out of 39) in northern states of Uttar Pradesh and Haryana and the eastern state of Orissa; *B. abortus* biotype 9 in Orissa and *B. abortus* biotypes-4, -6 and -9 and *B. melitensis* biotype-2 in the southern state of Tamil Nadu. Further, *B. melitensis* biotype-1 was encountered in cattle and buffalo from Haryana and in the southern state of Andhra Pradesh and in another southern state of Karnataka (Hemashettar *et al.*, 1987). Later, multiplicity of infection with *B. abortus* biotypes-1, 3, 6 and 9 was recorded in Orissa (Mohanty and Panda, 1988). In the northern state of Punjab, the association of *B. suis* in cattle and buffalo abortions has been reported (Mathur, 1985).

Bovine population in India is spread through the country and is seen in majority as compared to other species. Bovine brucellosis is widespread all over the Indian subcontinent. Much number of cases of bovine brucellosis makes the plausible transmission to other species as well. Isloor *et al* (1998) reported overall seroprevalence of 1.9 % in cattle and 1.8 % in buffaloes studied from 19 of 23 states. A seroprevalence study from Uttar Pradesh by Upadhyay *et al* (2007) recorded 7.25 % prevalence in bovine (12.77 % in cattle and 3.55% in buffaloes). Various reports from Punjab recorded as worst affected bovine population with constant presence with 11.23% overall prevalence (Dhand *et al.* (2005) varied from 0% to 24.3% in different villages. Earlier studies had estimated the disease in the state from as low as 7.54% to as high as 18.07% (Sharma *et al.* 2007). Aulakh *et al.*, 2008 estimated a 17.68% prevalence of bovine brucellosis in Punjab and Senthil *et al.* (2013) ranging from 3.3% – 11.4% with various diagnostic tests in Chennai. Milk ring test and milk-ELISA conducted on the samples of the same state revealed a prevalence of 4.35% and 5.38% respectively (Kumar, 2017). As high as 29.61% cattle and buffalo were reported to be affected in Uttarakhand (Maansi and Upadhyay, 2015).

Organized sector (41.30% on serological basis and 27.02% through milk tests) bears a greater burden as compared to non-organized or small herds (4.34% on serological basis and 3.06% through milk tests). Mehra *et al.* (2000) reported 6.5% (111/1629) prevalence in cattle from organized farms, compared to 5.1% (12/231) from unorganized sector. Similar observation was made by Isloor *et al.* (1998) in a detailed study of 47 organized farms in Karnataka, wherein 207 of 4995 (4.1%) serum samples from cattle showed titers for brucellosis. This high prevalence of animal brucellosis is responsible for human infections due to close contact with animals.

In sheep and goats

Polding (1942) first reported the isolation of *B. melitensis* in goats. Thereafter, *B. abortus* was isolated from cases of abortion in Haryana (Mathur, 1967). *B. melitensis* biotype-1 was isolated in the states of Karnataka, Andhra Pradesh, Maharashtra and Gujarat, and *B. melitensis* biotypes-1 and 3 in Haryana (Sen and Sharma, 1975, Hemashettar *et al.*, 1987). After investigations of 50 isolates from goats and 38 from sheep, Mathur (1985) opined that *B. melitenses* and *B. abortus* infections were common in sheep and goats. The sheep isolates included 32 isolates of *B. melitensis* and 6 of *B. abortus* as compared to 39 isolates of *B. melitensis* and 11 *B. abortus* from goats. He concluded that *B. abortus* infections of these animals were much higher than compared to other countries. *B.abortus* biotype 4 has been observed as a predominant biotype in small ruminants of Tamil nadu (Darshana *et al.*, 2016)

B. melitensis is the major cause of abortion in sheep and goats in many countries including India. The infection is wide spread in India (Ghosh and Verma, 1985). Serological surveys of small ruminant brucellosis have indicated varying levels of infection in different states. A number of 4.9% of sheep and 7.6% of goats in Karnataka (Desai *et al.*, 1995); 11% of sheep and 18% goats in northern state of Delhi; 50% sheep and 16% goats in Punjab and 33% sheep and 30% goats in the western state of Rajasthan (Kumar et al., 1997b); 55% of goats in Andhra Pradesh (Mrunalini *et al.*, 2000) and 24% of goats and 4.7% of sheep in Uttar Pradesh (Singh *et al.*, 2000) have been recorded. It was observed that flocks with history of abortions had high incidence of brucellosis (Mrunalini *et al.*, 2000). In a national survey of sheep and goat brucellosis, Isloor *et al.* (1998) examined serum samples originating from 10 states, which included 6305 from sheep, and 3849 from goats with cumulative incidence in sheep as 7.9% compared to 2.2% in goats. The serological evidence of *B. ovis* infection in 6 out of 102 rams has been reported in the northern state of Himachal Pradesh (Katoch *et al.*, 1996). Mangalgi *et al.*(2015) recorded a prevalence of 8.23% in sheep and 4.43% in goats. None of the sheep while 5.81% goats were found to be affected in Uttarakhand region (Maansi and Upadhyay, 2015). The organized sector samples showed higher seroprevalence in goat (7.79 %, 35/449) than sheep (4.06 %, 35/861) by RBPT. Similarly, in iELISA, goat samples showed a higher seroprevalence (9.35%, 42/449) compared to sheep (7.50%, 65/861) (Kanani *et al.,* 2018).

In pigs

Pig farming is restricted to certain parts of the country and lack of emphasis account for only a few reports on porcine brucellosis. Mathur (1985) isolated *B. suis* biotype-2 from Yorkshire pigs in Tamil Nadu. Two organized piggeries having animals with clinical history of abortion in sows and orchitis in boars revealed the presence of *B. suis* biotype 1 in the farms of Southern India (Shome *et al.*, 2018).

Records show the seroprevalence levels of 3.2% in Madhya Pradesh (Soni and Pathak, 1969), 11.3% in Tamil Nadu (Kumar and Rao, 1980) and 6.3% in Karnataka (Krishnappa *et al.*, 1981) states. However, Thoppil

(2000) observed 9.5% seroprevalence in 756 pigs slaughtered in Karnataka. Shome *et al.*, (2018) established an association in clinical symptoms as abortion in sows and orchitis in boars with brucellosis seropositivity.

In dogs

Pillai *et al*. (1991) first reported about presence of *B. canis* infection in in Tamil Nadu using *B. canis* antigen in mercaptoethanol test (MET) on 640 dogs with 2.18% (14) presence. These initial findings were reconfirmed in a similar serological survey of 460 dogs, which showed 2% infection (Srinivasan *et al*., 1992). There was no evidence of breed or sex predisposition in canines. However, Maansi and Upadhyay (2015) on 26 dog samples recorded a prevalence of 7.69% in male dogs through RBPT and ELISA and none of the female dogs was positive by serological test. A study by Sharma (2014) on canines exhibiting symptoms of abortion, orchitis, anorexia, persistent temperature, itching etc. revealed a prevalence of 32.6%.

Conclusion

Brucellosis is an endemic disease in India. It is widely prevalent in all the domesticated species of animals and in humans as well. Despite having the knowledge about the disease and its easy mode of transmission, the disease has faced negligence as far as its control is concerned. India needs to have effective plan to control the disease either by vaccination or by easy implementable policy for the removal of the infected animals of a herd. The challenge persists as the country has its own religious beliefs. With a much higher prevalence observed in humans, the effective strategies for controlling the disease require immediate and stern action.

Acknowledgement

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Cinico-therepeutic management of Toxocara vitulorum infection in buffalo calf: A case report

[*]Kapil Kumar Gupta, Neha Gupta, Brahmanand and P.I. Ganesan
[*]Assistant Professor, department of Veterinary Clinical Complex
Apollo College of Veterinary Medicine, Jaipur

Corresponding author: **Kapil Kumar Gupta** (dr.kapil09@gmail.com)

Abstract

A buffalo calf aged about one and half month was presented to Veterinary Clinical Complex, Apollo College of Veterinary Medicine Jaipur with the history of diarrhoea, inappatance, oliguria, poor growth, recumbency and fever since last few hours. On distant examination animal was looking weak, lethargic and debilitated with slight serous secretion from nostril and frothy salivation. Close physical examination revealed high rectal temperature (108.8°F), dyspnea with snoring sound, slightly pale mucus membrane, hyperpnoea and twitching of jaw muscles. Fecal examination reveled presence of eggs of *Toxocara vitulorum* in direct fecal smear. On the basis of history, clinical signs and fecal examination the case was diagnosed as Toxocariasis and treated with Intravenous administration of normal saline, meloxicam, Vitalgin, oxytetracycline, multivitamins, deriphyline, intramuscular injection of ivermectin and single oral administration of suspension consisting of 1.5% fenbendazole and 0.5% praziquental. Treatment was continued for five days. Fecal examination was done on definite interval (0, 3 and 7 day) and found negative for eggs of *Toxocara vitulorum* on 7th day of treatment. Animal was started improving from 3rd day of treatment and fully recovered within a week of start of treatment.

Key words: Toxocara, Fenbendazole, Praziquental

Introduction

Toxocara vitulorum is a worldwide ascarid parasite residing in the intestine of bovine **(Rast et. al., 2013)** and considered as major constraint in organized dairy farms, due to high morbidity and mortality **(Srivastava and Sharma 1981)**, production loss and ineffective implementation of breeding programmes **(Chowdhury 2002).** The parasite causes damage to the intestinal mucosa by its mechanical activity resulting into hemorrhagic enteritis. Besides, larvae of parasite also migrate to different organs like liver and lungs via hepatic portal vein and hepatic vein respectively. Extensive tissue larval migration results into development of different clinical signs including pneumonia. Major clinical signs of Toxocariasis are anorexia weight loss, anemia, diarrhoea (foul smell), poor hair coat, eczema, and bloat in young buffalo calves **(Wickramasinghe et al. 2009 and Roberts 1993).** Diagnosis of toxocarosis can be done on the basis of clinical signs, faecal examination for eggs, serological tests and necropsy findings. Wide variety of anthelmintic drugs such as Pyrantel, Levamisole, fenbendazole, Piperazine, Ivermectin etc. are used to treat the immature as well as adult parasites.

Materials and methods

About one and half month old buffalo calf was brought to Veterinary Clinical Complex, Apollo College of Veterinary Medicine Jaipur with the history of diarrhoea, inappatance, oliguria, poor growth, recumbency and fever since last few hours. On distant examination animal was found weak, lethargic and debilitated with slight serous secretion from nostril and frothy salivation. Close physical examination revealed high rectal temperature (108.8°F), dyspnea with snoring sound, slightly pale mucus membrane, hyperpnoea and twitching of jaw muscles. About 5 ml of blood was collected from the jugular vein of the animals in screw capped sterilized glass vials containing ethylene diamine tetra acetic acid (EDTA) as an anticoagulant and 2 ml of blood was collected in sterilized centrifuge tubes for haematobiochemical analysis. Small amount of fecal sample was collected directly from rectum of calf in sterile plastic container and examined under microscope in direct smear which revealed presence of eggs of *Toxocara vitulorum* in direct fecal smear. On the basis of history, clinical signs and fecal examination the case was diagnosed as Toxocariasis.

Table 1: Physical parameters before and after treatment

Parameters	Before treatment	After treatment		
		Day 0	Day 3	Day 7
Rectal Temperature(RT) $^{/0}$F	108.6	105.4	97.8	100.6
Heart Rate (HR)/Min	144	132	112	118
Respiration Rate (RR)/ min.	52	72	48	22

Table 2: Hemato-biochemical parameters in affected calf

S. No.	Parameters	Value	Reference range
1	Haemoglobin (g/dl)	6.8	8-15
2	Packed Cell Volume (%)	28	25-45
3	Total Erythrocyte Count ($\times 10^6$ /cumm)	8.4	5-10
4	Total Leucocyte Count ($\times 10^3$ /cumm)	14.2	4-12
7	Total protein (g/dl)	4.2	5.7-8.1
8	Urea (mg/dl)	16	8-27
9	AST (IU/L)	92	78-132
10	ALT (IU/L)	28	11-40
11	Creatinine (mg/dl)	0.8	1-2

Reference values from: Veterinary Medicine. A textbook of the diseases of cattle, horses, sheep, pigs and goats, 10th edition

Treatment and discussion

In the present case reports of T. vitulorum infestation in young buffalo calf treatment was started with Intravenous administration of normal saline (NSS @30ml/kg), meloxicam

(Melonex[R]@0.2mg/kg), Vitalgin (@1mg/kg), oxytetracycline (Oxynex[R]@20mg/kg), multivitamins (Tribivet[R]@1ml/20 kg), intramuscular injection of ivermectin (Neomec[R]@200µg/kg) and single oral administration of 30 ml suspension consisting of 1.5% fenbendazole and 0.5% praziquental. Treatment therapy was continued for five days (except oral suspension). Fecal examination was done on definite interval (0, 3 and 7 day) and found negative for eggs of Toxocara vitulorum on 7th day of treatment. Animal was started improving from 3rd day of treatment and fully recovered within a week of start of treatment.

Toxocara vitulorum is very common intestinal parasite in young cattle of tropical and subtropical region with either clinical or subclinical infection **(Roberts, 1993).** In present study the severe clinical signs of *Toxocara vitulorum* infection were seen and symptomatic treatment was started accordingly. Clinical signs of present study are almost similar to the findings of **Ahmed et al. (2015)** except increase in physiological parameters (rectal temperature, respiratory rate and heart rate) which were in normal range in his study. Decrease in the value of Hb in present study is in accordance with the findings of **Sarma et al. (2012).** Lower level of Hb in the infected calf might be due to inappatance and less availability of ingested nutrients (iron, copper and vitamin B12) to host because of competition for nutrients by adult parasite in the intestine **(Coles, 1974).** PCV value in present report was found to be normal but on lower side of reference range. This is in contrast with the findings of **Kumar and Verma (2006)** who reported significant increase in PCV in calf naturally infected with *Toxocara vitulorum.* Change in TLC value in present study is similar to the findings of **Devi et al. (2000)** who reported significant increase in the TLC during *Toxocara vitulorum* infection in calf which might due be to increase in the number of phagocytic cells because of stimulation of the host defence mechanism. Total serum protein was found lower than the normal which is in accordance with the findings of **Chaudhry et al. (1999).** It might due to lack and improper absorption of sufficient nutrients from the intestinal tract due to gastrointestinal disturbances caused by T. vitulorum. Urea, creatinine, AST and ALT were found within normal range which suggested either no or mild changes in liver and kidney.
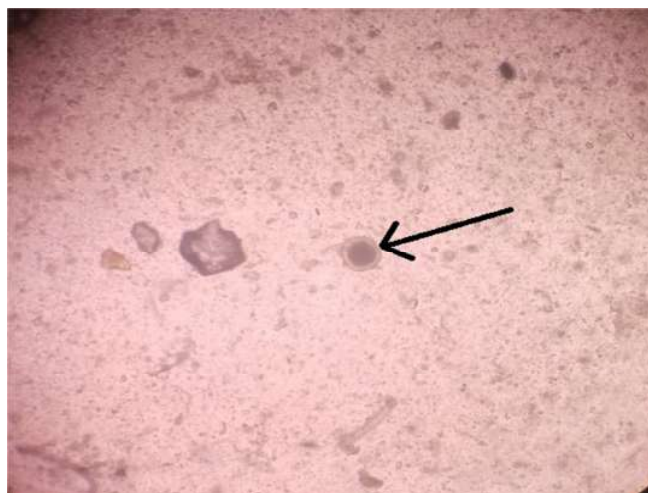

Fig: Egg of Toxocara vitulorum in direct fecal smear

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Evaluation of the acaricidal therapies for sarcoptic mange in cattle in and around Jabalpur

Anjali Singh, Amita Tiwari*, D.K.Gupta, Shivangi Sharma and Brejesh Singh

Department of Veterinary Medicine, College of Veterinary Science and Animal Husbandry, Nanaji Deshmukh Veterinary Science University, Jabalpur (M.P.)

*Corresponding author- amitasandhu@gmail.com

Abstract

The present study was undertaken to evaluate acaricidal therapies for sarcoptic mange in cattle. Out of 684 cattle screened from various organized and unorganized sectors in and around Jabalpur, 112 cattle were showing signs of dermatological disorders and were selected for further study. The observed clinical signs in sarcoptic mange affected cattle were found to be pruritus (100%), alopecia (100%), erythema (58.33%), exocoriation (75%) and thickening and wrinkling of skin (75%).Confirmation of sarcoptic mange was done by microscopic detection of *Sarcoptes scabiei* mite in the skin scraping. After confirmation of the presence of mite *Sarcoptes scabiei,* 12 cattle were selected for therapeutic study and 6 apparently healthy cattle were selected for healthy control group. Cattle of groups T_1, T_2, were treated with 1% Injection Ivermectin and 3.15% Injection Ivermectin, respectively. Skin scraping examination was done on day 0 and on days 14, 28, 42, respectively. On the basis of clinical recovery response on day 42 post treatment and skin scraping examination results therapeutic evaluation of various acaricidal drugs were done. On the basis of clinical observation and presence of mite injection ivermectin 1% @ 200 mcg/kg b.wt. was found to be most efficacious.

Introduction

In the Indian subcontinent, dairy cattle are usually infested with several parasites, among which *Sarcoptes scabiei* infestation is the common and serious problem, generally affecting sparsely haired parts of the body. Infestations generally located at the base of the tail, the inner thigh, under the neck and the brisket. It is recognised as one of most serious and contagious parasitic skin disease of dairy animals in the Indian subcontinent. Animals in poor condition appear to be most susceptible for the disease. Factors like stress, overcrowding, poor nutrition, cold weather and immune-supression make the animal succeptible for the disease. The disease also has zoonotic importance as the infection can be transferred to

human beings during milking. Diagnosis of sarcoptic mange is based on the clinical manifestations and the demonstration of mites or their developmental stages in host skin scrapings.

Control of mange on cattle can be achieved by application of topical chemical acaricides such as organophosphates and pyrethroids. However, burrowing habit of mites into the epidermis makes them difficult to control hence sometimes two or more applications are necessary at 7–10 days intervals for effective cure.

Sarcoptes mites burrow deep into the epidermis creating tunnels of up to 1 cm in length in which they feed and reproduce. Their burrowing and feeding activities cause inflammation and severe pruritus, loss of hair, excoriation, marked thickening and proliferation of the epidermal layer of the skin. Besides other pathological changes, decrease in feed digestibility, nutrient absorption and alteration in hepatic structure and function occurs (Dimri and Sharma, 2004). Economic losses associated with the disease are of a very high magnitude due to hide damage, decreased milk and meat production, morbidity and mortality. The morbidity rate varies from 1.5 to 82%, and reaching up to 100% in severely affected herds, with heavy losses of young animals (Gill *et al*., 1989)

The avermectins have revolutionised the approach towards control of mange as well as internal parasites. Ivermectin is a derivative of the avermectins, macrocyclic lactones produced from *Streptomyces avermitilis*. Ivermectin has been reported to have a wide spectrum of activity and excellent potency against many immature/mature nematodes and arthropod parasites of cattle. Their popularity is related to high potency, persistent activity and low toxicity. Macrocyclic lactones are safer for environment , easy to administer and so far no reports have been published for their resistance. The macrocyclic lactones are well absorbed when administered *per os* or parenterally. However, the route of administration and formulation may affect the drug pharmacokinetics. (Lavigne and Smith ,1983)

Despite its importance, sarcoptic mange in cattle has not been given due attention and a very scarce literature is available with respect therapeutic aspects of sarcoptic mange in cattle. So, keeping in view the above facts, this study was aimed with the objective to evaluate the acaricidal therapies for sarcoptic mange in cattle.

Materials and methods

The work was conducted in the Department of Veterinary Medicine, College of Veterinary Science and Animal Husbandry, N.D.V.S.U., Jabalpur (M.P).The study was conducted for a

period of 10 months i.e**.** between July 2017 to April 2018. Total 684 cattle screened from various organized and unorganized sectors in and around Jabalpur, 112 cattle were showing signs of dermatological disorders and were selected for further study. Confirmation of sarcoptic mange was done by microscopic detection of *Sarcoptes scabiei* mite in the skin scraping. After confirmation of the presence of mite *Sarcoptes scabiei,* 12 cattle were selected for therapeutic study and 6 apparently healthy cattle were selected for healthy control group. The infested cattle were randomly divided into two groups i.e. $T_1 - T_2$ (each group comprising of 6 cattle) and six apparently healthy cattle were selected to serve as healthy control Group (T_3). Cattle of different groups were given treatment as per the following therapeutic regimen (Table 01).

Table 01: **Therapeutic regimen for sarcoptic mange in cattle**

Group	Number of animals	Drugs and Dosage
T_1	06	Injection Ivermectin 1% @ 0.2 mg/kg b.wt., s/c, weekly for 6 weeks
T_2	06	Injection Ivermectin 3.15% (LA) @ 630 mcg/kg b.wt., s/c, once
T_3	06	Healthy Control group

In addition to the above therapy, injection enrofloxacin @ 5mg/kg b.wt, i/m and injection pheniramine maleate @ 0.5 – 1 mg/ kg b.wt., i/m were used as per the clinical condition of animals.

Skin scraping examination was done on day 0 and on days 14, 28, 42, respectively. On the basis of clinical recovery response on day 42 post treatment and skin scraping examination

Evaluation of therapeutic Response

The response of therapeutic study was evaluated on the basis of

1. Improvement in clinical status of the animals.

2. Skin scraping test results for *Sarcoptes scabiei*

Results

Clinical symptoms such as alopecia, pruritus, erythema, excoriation, thickening and wrinkling and skin scraping examination of all the cattle under therapeutic trial were recorded on day 0 and on days 14, 28 and 42 post treatment and compared . On the basis of per cent recovery of various clinical observation on day 42 post treatment therapeutic evaluation of acaricidal drugs used in group T_1 and T_2 was done.

The result revealed that in T1 group on day 0, alopecia and pruritus were observed in all six cases, erythema in three cases, excoriation, thickening and wrinkling in four cases. On day 42 recovery from pruritus, excoriation, thickening and wrinkling was seen in 100 per cent cattle whereas alopecia and erythema recovered in 83.3 per cent cattle. Mites were absent in all the six cases on day 42 (post treatment) (Table 02) while in T2 group on day 0 alopecia and pruritus were seen in all the six cases whereas excoriation, erythema and thickening and wrinkling of skin in five, four and five cases, respectively. On day 42 post treatment recovery from pruritus, erythema was seen in 100 per cent cattle whereas alopecia, excoriation and thickening and wrinkling were recovered in 83.33 per cent, 60 per cent and 60 per cent cases, respectively. Absence of mites were seen in five cases only on day 42 post treatment (Table 02).

On the basis of clinical observation and presence of mite injection ivermectin 1% @ 0.2 mg/kg b.wt. was found to be most efficacious.

Table : 02 Evaluation of comparative therapeutic response of different treatment groups

Clinical observation	Percent recovery	
	T1	T2
Alopecia	83.33	83.33
Pruritus	100.00	100.00
Erythema	83.33	100.00
Excoriation	100.00	60.00
Thickening and wrinkling	100.00	60.00
Absence of mite	100.00	83.33

Discussion

Varying degrees of frequency of occurrence of symptoms of sarcoptic mange were recorded in the present study. Topping the order was the signs of alopecia and pruritus being presented in 100 % of the cases. This was followed by excoriation (75%), thickening and wrinkling (75%) and erythema (58%) .

The pattern of signs observed in this study was in accordance with, Dimri *et al.* (2008) and Kazmi *et al.* (2009).

Proliferation of mast cells and resultant increase in chymase and tryptase activity is supposed to play an important role in development of skin lesions (Dimri and Sharma, 2004). The sarcoptic mange mites penetrate deeper into the stratum corneum. During their penetration, they might cause lysis of the tissues and later feed on the lysed tissues resulting in deep burrow formation. During the burrowing process, the mite may deposit the

body secretions, saliva and faecal material, therein stimulating the hosts immune system and causing characteristic lesions, inflammation, pruritus and scratches and rubbing on hard objects or against one another resulting in alopecia. Cutaneous hypersensitisation noticed in mange infestation might be due to the keratinization and proliferation of connective tissue (Thakar, 2004).

Therapeutic response evaluation of various treatment groups

Group T_1 (Injection ivermectin 1%)

Cattle of group T_1 were treated with injection ivermectin 1% at weekly interval for 6 weeks and the results revealed that on day 42 post treatment, recovery of pruritus, excoriation, thickening and wrinkling was 100 per cent. The recovery of erythema and alopecia was 83.33 per cent. All the six cattle showed no mites on day 42 post treatment.

These findings are in corroboration with the findings of Kazmi *et al.* (2009) who reported 86.66% efficacy of ivermectin in mange affected buffaloes. The results of present therapeutic study were in accordance with the previous studies and the outcome of therapy was best as compared to other groups. Ivermectin is a derivative of the avermectin a macrocyclic lactone produced from *Streptomyces avermitilis*. It has been reported to have a wide spectrum of activity and excellent potency against many immature and mature nematode and arthropod parasites of cattle. Ivermectin is believed to act by potentiating the release and binding of gamma-aminobutyric acid (GABA) in certain nerve synapses, and thus blocking GABA-mediated transmission of nerve signals (Wang & Pong, 1982).

Group T_2 (Injection ivermectin 3.15%)

Cattle of group T_2 was treated with Injection ivermectin 3.15 per cent s/c once and the results revealed that on day 42 post treatment recovery of pruritus and erythema was 100 per cent. The recovery of alopecia, excoriation, thickening and wrinkling was 83.33 per cent, 60 per cent, 60 per cent, respectively. Out of 6 cattle, only 5 showed absence of mites on day 42 post treatment.

The efficacy of treatment in cattle of this group was less as compared to cattle of group T_1. The result of present study was partially in accordance with Hamel, (2014) who reported that resolution of skin lesions in 3.15% ivermectin treated cattle in the sarcoptes infestation was observed from day 21 onwards and animal had a healthy skin from day 49 onwards. On the contrary, Lifschitz *et al.* (2007) reported that compared to the conventional

1% injectable preparation, a single 3.15% ivermectin administration leads to a longer-lasting presence of ivermectin in the systemic circulation and thus provides a more prolonged persisting biological activity of ivermectin.

In the results of present study, although 3.15% ivermectin resolved the lesions of sarcoptic mange but the efficacy of treatment was less as compared to 1% ivermectin. Long-acting endectocide formulations are currently used for the advantage of persistent anthelmintic efficacy in strategic programs for controlling nematodes and ectoparasites. Long acting (3.15%) ivermectin have been developed mainly in order to prolong the period of residual protection in ruminants from re-infection with gastrointestinal and pulmonary nematodes during grazing period (Vercruysse and Rew, 2002). Variations in the results of present study with previous work might be due to the difference in the severity of infestation of sarcopitic mites and other managemental conditions of the cattle.

Conclusion

On the basis of therapeutic response evaluation Injection Ivermectin (1%) @0.2 mg/kg/week s/c was found to be most effective followed by 3.15 per cent Injection Ivermectin .

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Efficacy of silymarin in therapeutic management of dogs affected with ascites

Tara, S., B. Roopali., S. Roy and M. Roy

Department of Veterinary Medicine

College of Veterinary Science & AH, Durg

Chhattisgarh

Abstract

Ascites is defined as pathological accumulation of serous or serosanguinous fluid in the peritoneal cavity and is usually reserved for a transudate that is associated with liver or right side heart failure. The present study was conducted to assess the therapeutic efficacy of silymarin in ascites in dogs. For therapeutic studies, ascites affected dogs were divided into three groups: Group I, Group II and Group III each consisting of 7 animals. Group I animals were treated with silymarin @ 20 mg/kg, Group II dogs were administered silymarin @ 30 mg/kg and Group III dogs were given silymarin @ 50 mg/kg once a day PO for 15 days. The clinical recovery in dogs affected with ascites was assessed on the basis of improvement in clinical signs and restoration of the altered haemato-biochemical parameters following treatment. The clinical recovery was noticed in all groups but earlier and best response to therapy was recorded in the animals of group III followed by group II and I respectively. On the basis of clinical signs of improvement along with restoration of hematobiochemical parameters, it was observed that Silymarin @50 mg/kg b.wt along with supportive therapy proved better in therapeutic management of ascites of hepatic origin in dogs.

Introduction

Liver plays a pivotal role in the regulation of body metabolism, secretion and detoxification process of many substances. It is the largest parenchymal organ in the body

(Zakim, 1985) having the biological property of tremendous storage capacity, functional reserve and regenerative capabilities. Ascites is defined as pathological accumulation of serous or serosanguinous fluid in the peritoneal cavity and is usually reserved for a transudate that is associated with liver or right side heart failure (Reynolds, 2000 and Moore *et al.,* 2003). Generalized description of ascites includes distension of abdomen with other fluid i.e. chyle, blood and inflammatory exudates. Ascites is always an indication of disease, therefore thorough investigation should be aimed at identifying the underlying primary condition. The pathogenesis of ascites is related to renal, hepatic and cardiovascular insufficiencies. The clinical signs directly associated with ascites includes abdominal distension, respiratory distress, diarrhoea and weight loss in PLE, polyuria / polydipsia in chronic renal disease, thrombo-embolic disease in PLE and PLN, jaundice in liver failure or bile peritonitis, excessive intolerance and collapse in heart failure and cardia tamponade. Anuria and uraemia in uro-abdomen and collapse if hemoperitoneum (Hall, 2005 and James *et al.,* 2008). Coagulopathies are usually seen in animals with liver disease. Probable mechanism include decreased clotting factor synthesis, decreased vitamin K absorption in cholestatic disease, and disseminated intravascular coagulation (Leveille-Webster CR, 2000). In India, ascites has been reported from various parts of the country by various researchers. The present study was conducted to assess the therapeutic efficacy of silymarin in ascites in dogs.

Materials and methods

Therapeutic management of canine ascites in and around Durg were studied by prospective data. For prospective study, cases presented to Teaching Veterinary Clinical Complex (T.V.C.C) of Veterinary College, Durg and Government Veterinary Hospitals in and around Durg district of Chhattisgarh were analysed. Naturally infected dogs with ascites were clinically examined. Clinical examination of distended abdomen, anorexia, vomiting, mucous membrane, depression and fluid thrill during tactile percussion were recorded.   For

therapeutic studies, ascites affected dogs were divided into three groups: Group I, Group II and Group III each consisting of 7 animals. Group I animals were treated with silymarin @ 20 mg/kg, Group II dogs were administered silymarin @ 30 mg/kg and Group III dogs were given silymarin @ 50 mg/kg once a day PO for 15 days. All the affected animals were treated with furosemide in combination of spiranolactone, B- complex vitamins for 5-7 days as supportive therapy.

Results and Discussion

Therapeutic efficacy of silymarin was evaluated in dogs (n=21) affected with ascites of hepatic origin. Animals were divided into three groups with each group having seven animals which received silymarin at the dose rate of 20, 30 and 50 mg/kg body weight respectively orally once daily for 15 days. Dogs of all groups also received supportive therapy of furosemide and spironolactone combination at the rate of 2mg/kg body weight for 5 days orally. The clinical recovery in dogs affected with ascites was assessed on the basis of improvement in clinical signs and restoration of the altered haemato-biochemical parameters following treatment. The clinical recovery was noticed in all groups but earlier and best response to therapy was recorded in the animals of group III followed by group II and I respectively. On the basis of clinical signs of improvement along with restoration of hematobiochemical parameters, it was observed that Silymarin @50 mg/kg b.wt along with supportive therapy proved better in therapeutic management of ascites of hepatic origin in dogs. The hepatoprotective action of silymarin has also been reported by various workers (Tiwari *et al.,* 2005; Pandey and Sahni, 2011; and Ramdas, 2013). Silymarin possesses antioxidant, anti-inflammatory and detoxifying actions as well as it promotes cellular repair and liver regeneration. Saller *et al.,* (2001) have also reported that silymarin may play a role in the therapy of (alcoholic) liver cirrhosis. The administration of furosemide @ 2mg/kg b.wt

and spiranolactone @ 2mg/kg b.wt combination has also been reported by various workers in dogs with ascites (Center, 2006 and Twedt, 2003).

Silymarin is obtained from *Silybum marianum* (Milk thistle) that has been used medicinally for centuries as a herbal medicine for the treatment of liver related disorders. It is widely prescribed by herbalists and has almost no known adverse effects. The plant is native to the Mediterranean and grows throughout Europe and North America. It also grows in India, China, South America, Africa, and Australia (Dixit *et al.,* 2007). The active ingredient of milk thistle is 'Silymarin' that is found in the seeds. Silymarin is a complex of polyphenolic compounds, consisting of mainly seven closely related flavonolignans, like silibin-A, silibin-B, isosilibin-A, isosilibin-B, flavonoid taxifolin, silidianin, silichristin and isosilichristin. Silymarin has a strong anti-inflammatory and antioxidant actions along with detoxifying actions and promotes cellular repair and hepatic regeneration (Pandey and Sahni, 2011). Silymarin controls the cell membrane permeability, inhibits inflammatory biochemical pathways, have free radical scavenging properties, increase protein production by liver cells, may stabilize mast cells and higher doses of silymarin increases the flow of bile. Milk thistle is consumed as a supplementary food having hepatoprotective effect. Silymarin is known to have antioxidant, immunomodulatory, anti-fibrotic, anti-proliferative, and antiviral properties. In addition to, silymarin maintains the integrity of hepatocyte membrane and impedes the entrance of toxic substances or xenobiotics. Due to its phenolic nature, it is capable of donating electrons to stabilize endoplasmic reticulum and reactive oxygen species (ROS). Silymarin also affects intracellular glutathione, which prevents lipoperoxidation of membranes.

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Molecular Characterization of ESBL positive *Escherichia coli* detected
from Bovine Milk in West Bengal

Abhiroop Banerjee[1], Kunal Batabyal[1*], Samir Dey[1], Devi Prasad Isore,[1] and Abhishek Dharm Singh[2]

[1]Department of Veterinary Microbiology, Faculty of Veterinary and Animal Sciences
West Bengal University of Animal and Fishery Sciences, Kolkata – 37, West Bengal, India;
[2]Department of Veterinary Public Health, F/VAS, WBUAFS, Kolkata – 37, West Bengal, India

*Corresponding author: Dr. Kunal Batabyal, e-mail: drkb.micro@gmail.com

Abstract

Bovine Milk is regarded as a complete food and is included in the human diet especially for the children all India. Milk can get contaminated by *Escherichia coli* from various sources causing huge infections, particularly in infants. ESBL-positive *E. coli* may be found which makes the treatment tougher due to increased resistance to commonly used antimicrobials. A total of 102 bovine milk samples collected from different dairy farms of West Bengal, were examined to detect 14(13.7%) *E. coli* strains, molecularly confirmed by detection of the 16S *rRNA* gene. Nine (64.2%) *E. coli* strains were found to be positive for extended-spectrum beta-lactamase (ESBL) production both phenotypically and genotypically with the presence of the *bla_{CTX-M}* gene. *In-vitro* antibiotic sensitivity assay of these 09 ESBL positive isolates revealed, colistin (100%), levofloxacin (88.9%) and imipenem (77.8%) to be effective against this pathogen but other drugs *viz.* cefotaxime (100%), ceftazidime & ampicillin (both 88.9%), amikacin & gentamicin (both 77.8%), azithromycin & tetracycline (both 66.7%) and others to be very much resistant.

Keywords – bovine milk, *bla*_{CTX-M}, *Escherichia coli,* 16S *rRNA*, antibiogram

Introduction

Indian dairy industry plays an important role in improving the rural sector and economy. Milk production in India is among quite high in the World [DAFD, 2017]. The milk industry in India generates huge self-employment through the production and marketing of milk in the rural sector. Milk mainly bovine milk is generally considered to be a great source of proteins to infants and is almost matching with mothers' milk. Milk is having great nutritional value so it can get easily spoilt due to faulty handling and storage due to the rapid multiplication of bacteria [Oliver *et al.*, 2005]. *Escherichia coli* is one common food contaminant which can cause food intoxication through contaminated milk leading to huge human health hazard [Hickey *et al.*, 2015]. Extended-spectrum beta-lactamases (ESBLs) are the bacterial enzymes produced by *E. coli* which can nullify the effect of many commonly used antimicrobials. Nowadays these ESBL-producing *E. coli* are commonly found all around the globe including India. These pathogens are now a major challenge for the treatment of general infections and causing a huge health hazard [Reich *et al.*, 2013]. ESBL positivity in *E. coli* is governed by few resistance genes which can easily get transferred to other pathogens leading to the

spread of drug resistance in those too [Hu *et al.*, 2016]. Presence of ESBL producing *E. coli* in the food chain possibly lead to cause a health hazard, in this background, the present study was aimed at the detection and characterization of ESBL positive *E. coli* from raw bovine milk samples collected from different dairy farms followed by detection of their antibiotic resistance patterns *in vitro*.

Materials and Methods

Sample collection:

Fresh bovine milk samples (n=102) were collected from different unorganized or private dairy farms/ farmers of West Bengal from May to August 2019. Approx. 10ml milk from each cow was directly collected in sterile plastic containers followed by transfer to the laboratory under ice cover for further study. All the collected samples were processed on the same day of receiving the lab.

Isolation and characterization of *Escherichia coli*:

All collected milk samples were streaked on to MacConkey's agar (Hi-Media, India) plates after overnight enrichment at 37°C. After 10-12 hrs incubation, tentative pinkish colonies were selected for isolation of *E. coli* by streaking on to Eosin Methylene Blue (EMB) agar (Hi-Media, India) plates. The characteristic 'metallic sheen' producing colonies were picked up for further characterization by Gram's staining and tests like the indole test [Carter and Wise, 2004, Quinn *et al.*, 2011].

Confirmation of *E. coli* isolates:

Confirmation of tentative *Escherichia coli* isolates was done by PCR detection of the 16S *rRNA* gene (585bp) specific for this bacterium, as per the protocol of Wang *et al.* (1996). Bacterial genomic DNA was extracted from the over-night broth culture of *E. coli* by the conventional phenol-chloroform method. The total reaction volume was 25µl which contains 5µl of bacterial DNA, 2mM $MgCl_2$, 0.2mM each dNTP, 0.25µl of each primer (F: 5'-GACCTCGGTTTAGTTCACAGA-3', R: 5'-CACACGCTGAC GCTGACCA-3'), and 1U of *Taq* DNA polymerase. The amplification conditions were: initial denaturation at 94°C for 5mins followed by 35 cycles of denaturation at 94°C for 30s, annealing at 58°C for 1min, and elongation at 72°C for 1min and a final extension at 72°C for 10mins. The PCR product was visualized by gel documentation system (UVP, UK) after electrophoresis in 1.5% (w/v) agarose (SRL, India) gel containing ethidium bromide (0.5µg/ml) (SRL, India).

Detection of ESBL production in *E. coli* isolates *in-vitro*:

Phenotypic detection of the presence of ESBL in *E. coli* isolates was done *in vitro* by disc diffusion method (Bauer *et al.*, 1966) using both Cefotaxime (30µg) and ceftazidime disks (30µg) with and without clavulanate (10µg) as per CLSI methods by Patel *et al.* (2015). A difference of >5mm between the zone diameters of each disk and their respective clavulanate disk is measured to phenotypically confirm the ESBL production by the *E. coli* isolates under study (Patel *et al.*, 2015).

PCR detection of ESBL production in *E. coli* isolates:

Escherichia coli strains positive for ESBL production *in-vitro*, were selected for PCR detection of the bla_{CTX-M} gene (540bp) responsible for ESBL production. The protocol described by Weill *et al.* (2004)

with some modifications is followed for the detection of the bla_{CTX-M} gene in selected *E. coli* isolates. Five µl DNA templates, 50pmol of each primer (forward $_{CTX-M}$-F 5'-CAATGTGCAGCACCAGTAA-3' and reverse $_{CTX-M}$-R 5'-CGCGATATCATTGGTGGTG-3'), 200mM deoxynucleoside triphosphate, 1U *Taq* DNA polymerase (Promega, USA), 2mM MgC1$_2$, and 10% dimethyl sulfoxide was added in a 25µl reaction mixture and subjected to PCR amplification. The cycling conditions included 10mins of initial denaturation at 94°C followed by 30s of denaturation at 94°C, 30s of annealing at 53°C and 1min of extension at 72°C for 35 cycles and 10mins of final extension at 72°C. The amplified product was visualized by gel documentation system (UVP, UK) after electrophoresis in 1.5% (w/v) agarose (SRL, India) gel containing ethidium bromide (0.5µg/ml) (SRL, India). A strain of *E. coli* (O2) maintained in the laboratory and one *Pseudomonas aeruginosa* (ATCC 27853) strain were used as positive and negative controls respectively.

In-vitro Antibiotic Sensitivity Test (AST) of ESBL positive *E. coli* isolates:

Antibiogram of the ESBL positive *E. coli* isolates was performed using 12 antimicrobials *i.e.* amikacin, ampicillin, azithromycin, colistin, cefoxitin, cefotaxime, ceftazidime, gentamicin, imipenem, levofloxacin, piperacillin-tazobactam and tetracycline by disc diffusion method (Bauer *et al.*, 1966). Young broth cultures of all the ESBL positive isolates were poured on to Mueller Hinton agar (Hi-Media, India) plates with uniform spreading followed by overnight incubation at 37ºC. The results were interpreted by measuring the inhibition zone diameter and comparing those with the standard chart (Patel *et al.*, 2015).

Results and Discussion

A total of 14 (13.7%) samples were found to be positive for *E. coli*, out of 102 bovine milk samples tested. Approx. 12% *E. coli* positivity was also reported by Kamaruzzaman (2015) and Badri *et al.* (2017) from milk samples from different countries like Malaysia, Sudan, etc. Geser *et al.* (2012) and Ali *et al.* (2016) also reported approx. 12.2-13.7% *E. coli* positivity in the bovine milk samples which almost matches the current findings. All the positive isolates produced typical 'metallic sheen' on EMB agar plates and matched morphological (Gram –ve bacilli) and biochemical characters (positive to indole) and were detected to possess 16s *rRNA* gene-specific for *E. coli* **(Figure 1)** (Carter and Wise, 2004; Samanta, 2013 and Quinn *et al.,* 2011).

Phenotypical ESBL detection by double disc method indicated, 09 (64.2%) *E. coli* isolates to be positive which was specifically confirmed by the presence of the bla_{CTX-M} gene (540bp) **(Figure 2)** in all these isolates by PCR study (Geser *et al.*, 2012; Osman *et al.*, 2012; Ibrahim *et al.* 2016). Higher positivity of ESBL *E. coli* from milk samples (66.7% and 23.53%) was also reported by Kamaruzzaman (2015) and Ali *et al.* (2016) respectively. Sharma *et al.* (2016) and Badri *et al.* (2017) also reported such ESBL positivity in *E. coli* strains from bovine milk samples and these may be of great concern as these pathogens may be carried out to the human consumers as well as calves leading to the spread of the antibiotic-resistant pathogens over a large human and animal population.

Antibiogram of all the ESBL-positive *E.coli* isolates (09) indicated higher resistances to most of the antimicrobials used *viz.* cefotaxime, ceftazidime, ampicillin, tetracycline, gentamicin, etc. (66-100%) but colistin (100%), levofloxacin (88.9%), and imipenem (77.8%), piperacillin-tazobactam (77.8%) were found to be sensitive against these pathogens **(Table 1)**. These findings almost match with the report of Faruk *et al.* (2016) who detected high-level resistance to ampicillin, cefotaxime, ceftazidime, and cefuroxime (all 100%) and tetracycline (93.5%) but sensitivity to imipenem (100%) of the ESBL *E. coli* strains isolated from cattle. Findings of Kamaruzzaman (2015), Ibrahim *et al.* (2016) and Hinthong *et al.* (2017) also match with this present report and thus stand confirmed. Ali *et al.* (2016) also detected ESBL positive *E. coli* pathogens to be resistant to ampicillin (86.11%), amoxicillin-clavulanic acid (63.89%), cefotaxime (100%), ceftazidime (66.67%), tetracycline (72.22%) and gentamicin (61.11%) which almost matches with the current findings.

Summary

Approx. 14% of the bovine milk samples studied were contaminated with *E. coli* among which 64% isolates were having the drug-resistant bla_{CTX-M} gene *i.e.* positive to ESBL production, which is of high significance and may be of great health concern for human beings. This transferrable drug resistance can easily get spread among closely related pathogens *in vivo* leading to fatal human health hazards. So, proper care should be taken to combat these dreadful pathogens. Very few drugs *viz.* colistin, levofloxacin, and imipenem were effective against these pathogens.

Acknowledgment

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TABLE:

Table 1: Antibiogram of 09 ESBL-producing *E. coli* strains isolated from bovine milk samples in West Bengal

| Sl. No.	Antimicrobials (Conc. in µg)	Sensitive		Intermediately sensitive		Resistant	
		No.	%	No.	%	No.	%
1.	Amikacin (AK - 30)	1	11.1	1	11.1	7	77.8
2.	Ampicillin (AMP - 10)	0	0	1	11.1	8	88.9
3.	Colistin (CL - 10)	9	100	0	0	0	0
4.	Cofoxitin (CX - 30)	3	33.3	1	11.1	5	55.6
5.	Cefotaxime (CTX - 30)	0	0	0	0	9	100
6.	Ceftazidime (CAZ - 30)	0	0	1	11.1	8	88.9
7.	Imipenem (IPM - 10)	7	77.8	2	22.2	0	0
8.	Gentamicin (GEN - 10)	0	0	2	22.2	7	77.8
9.	Levofloxacin (LE - 5)	8	88.9	1	11.1	0	0
10.	Pipercillin-Tazobactam (PIT - 100/10)	2	22.2	7	77.8	0	0
11.	Azithromycin (AZM - 30)	1	11.1	2	22.2	6	66.7
12.	Tetracycline (TE - 30)	2	22.2	1	11.1	6	66.7

FIGURE CAPTIONS:

Figure 1: Confirmation of *E. coli* isolates by detection of the 16S *rRNA* gene (585bp) by PCR

Figure 2: PCR detection of the *bla*$_{CTX-M}$ gene (540bp) in ESBL positive *E. coli* strains isolated from bovine milk samples

FIGURES:

Figure 1:
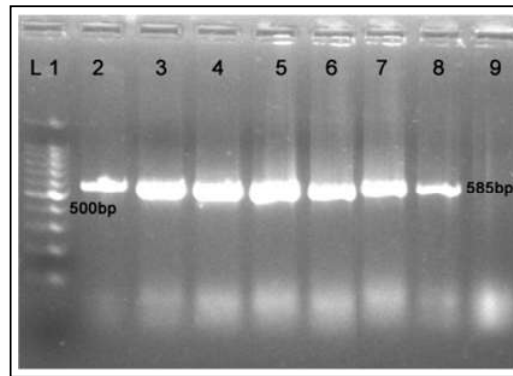
(L1: 100bp ladder, L2 – L7: Test samples, L8: Positive control, L9: Negative control)

Figure 2:
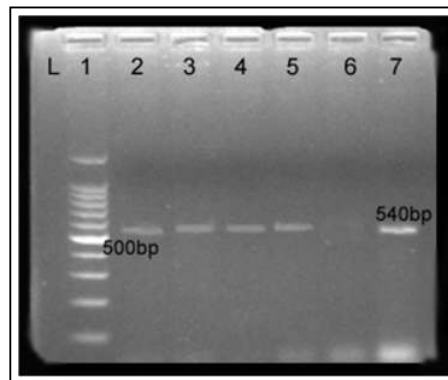
(L1: 100 bp ladder, L2-L5: Test samples, L6: Negative control, L7: Positive control)

Canine masticatory myositis in a Cross-breed dog- A case Report

Magar, S. A, Bhojne, G. R & V.M.Dhoot

Nagpur Veterinary College, Nagpur

Nagpur-440010

Abstract

A one year old male cross breed dog was brought with history of inability to open the mouth and unable to eat and drink from last 10 days. There was no history of dog bite nor of trauma. Clinical sign revealed anorexia, wasting of masticatory muscles, locked jaw (trismus). Neurological and orthopedic examinations revealed no abnormalities. Hematology revealed increase in eosinophil values. Muscle biopsy was not done as owner did not give consent for it. Based on clinical findings the case was tentatively diagnosed as masticatory muscle myositis. Dog was treated with prednisolone 1 mg/kg body weight PO BID for 15 days. Prednisolone therapy was tapered slowly for subsequent two weeks. Dog showed uneventful clinical recovery after a month.

Key words: Masticatory muscle myositis, Dog.

Introduction

Masticatory Muscle myositis (MMM) is an autoimmune disease in dogs which leads to focal inflammatory myopathy with clinical constraint of the chewing muscle group mainly the temporalis, masseter, pyterigoideus and rostral digastricus muscles, which are innervated by the mandibular arm of the trigeminal nerve (Melmed *et al*., 2004). Masticatory muscle myositis is a well-documented disease in dogs (Shelton, 2007). In most of the dogs,age of onset for masticatory muscle myositis is 3 years, while patients have been reported as young as 4 months of age (Gilmour *et al*.,1992). The disease can occur in any breed. No gender predilection has been found. Canine masticatory myositis leads to progressive demolition of type 2M muscle fibers by making

focal myositis (Bolfa *et al*., 2011). The disease etiology is unknown, but it is examining that myositis may arise from antibodies which are produced in response to an infectious agent that cross-reacts with endogenous antigens. Another researcher suggested that early myofibril damage in dogs with masticator myositis is commence by cytotoxic CD8 + T cells and later leads to antibody production against muscle fiber proteins (Reiter *et al*., 2007). The very common clinical symptoms of masticator myositis include difficulty in opening the jaw, pain in the jaw and masticatory muscle (Evans *et al*., 2004).

Case presentation

One-year old, male cross breed dog was brought with a complaint of inability to open the mouth and unable to eat and drink from last 10 days. As per the owner's information NSAID (Meloxicam) and antibiotic (Ceftriaxone) were prescribed in a private clinic. The presented case showed chronic phase of masticatory muscle myositis as there was bilateral atrophy of temporalis and pterygoid muscle (Fig.1 & 2). Clinical Examination revealed atrophy of the temporalis muscles, pain in the jaw and difficulty in opening the jaw. Generally, the disease represents in two phases i.e. acute and chronic. The acute phase is clinically characterized by pain in jaw, pyrexia, unable to eat, drooling of saliva, however in some cases edema in the masticatory muscle with restricted jaw movement (trismus) is also seen. Temporalis and pterygoid muscle swelling may cause exophthalmos resulting in an inability to blink properly. While the chronic phase is characterized by marked muscle atrophy due to decreased size of the muscle fibers that lead to fibrosis.

Diagnosis and finding

The dog with masticatory muscle myositis is presented with clinical sign such as inability to open the jaw (trismus), pain in jaw, and swelling or atrophy of the muscles of mastication. Dogs suffering from masticatory muscle myositis have a rigid jaw tone, whereas dogs with trigeminal neuritis usually have a flaccid jaw tone. In the initial examination, a blood was collected and laboratory examination was performed. Hematological parameters were within the normal range except for the elevated eosinophil values on hemogram. Because of the financial constraint owner declined for

the muscle biopsy and hence not performed. The patient was diagnosed with masticatory myositis based on clinical sign and initiated on corticosteroid treatment because of the eosinophil increase observed in the patient's blood examination.

Fig.1: Restricted jaw movement	Fig.2:Bilateral atrophy of temporalis and pterygoid muscle
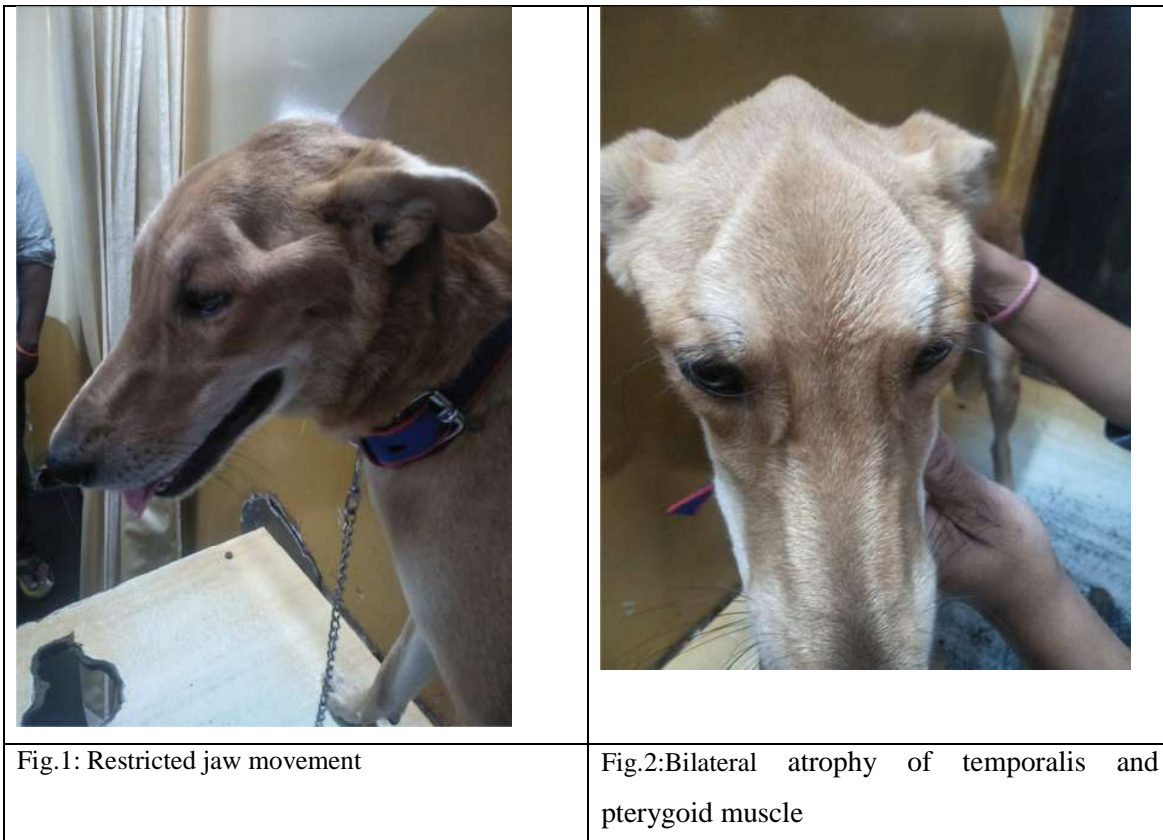
Treatment

For favorable result in masticatory muscle myositis requires early exact diagnosis and relevant therapy. Treatment for masticatory muscle myositis is amined on aggressive immunosuppression, which is attained by administration of oral Tab.Wysolone (Prednisone) at 1 mg/kg PO bid. This dose was continued until maximum jaw function has been retrieve. Following treatment, the clinical sign slowly reduced and the dog was

able to open the mouth and able to chew the food. The animal showed fully recovery after a month of treatment with prednisolone. After that the dose of prednisone was slowly tapered.

Discussion

Inflammatory myopathies are common in dogs and are most commonly seen as masticatory muscle myositis (Nanai *et al*., 2009). Muscle myositis is an inflammatory myopathy and selectively affects the muscles (temporalis, masseter, pyterigoideus) which help for chewing. This muscle group contains 2M myofibrils, which are specific to this muscle group and are not found in the extremities (Paciello *et al*., 2007). In dogs with acute phase of the disease, typically show bilateral swelling, painful masticatory muscles, jaw pain. Even under general anesthesia, the jaws often cannot be opened. In chronic cases, the muscles are atrophic and the trismus may continue (Czerwinski *et al*., 2015). In this case the dog had pain in the jaw, difficulty in opening the jaw, pain in the masseter muscles and atrophy of the temporalis muscles. This disease was formerly called eosinophilic myositis or atrophic myositis (Melmed *et al.,* 2004). Although some dogs have eosinophilia, this finding was seen in this case also. In many literature, predominant cell-type infiltrating in the masticatory muscles is eosinophils (Reiter *et al*., 2007). In the present case the laboratory examinations revealed elevated levels of eosinophils in the blood.

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Seasonal prevalence of different helminthic infection in goats in Bilaspur district of Chhattisgarh

Namrata Ottalwar, D.K. Sharma, P.K. Sanyal, Tanmay Ottalwar

Deptt. of Veterinary Parasitology, College of Vety. Sci. & A.H., Anjora, Durg (C.G.)

Abstract

The current study was carried out in and around Bilaspur district of Chhattisgarh state. During the study 410 goats were examined out of which 238 were positive for gastrointestinal helminthes. The overall prevalence percent of gastrointestinal helminthes was 58.04% with its maximum intensity in the monsoon season respectively. Low prevalence in winter season was due to reduced grazing hours of the animals, which helps in reducing the chances of contact between host and parasites.

INTRODUCTION

The Department of Animal Husbandry and Dairying (DAHD) released a census report of livestock population for the year 2019 on October 16. The data revealed the Goat Population in the country is 148.88 million during 2019. Total goat has increased by 10.14% over previous livestock census (2012). About 27.8 % of the total livestock is contributed by the goats. Goat rearing is practiced mainly by resources poor families and is a supplementary source of income for farmers during lean seasons for marginal, women and landless farmers. Small ruminants have high survival rates under drought conditions compared to large ruminants. Moreover, because of their higher reproductive rates and smaller reproductive cycle flock number can especially be restored more rapidly. In goats water economy is also an important biological feature. Due to their short reproductive cycle (short kidding interval) and high incidence of multiple births, there is a potential for a higher annual off take of goats, than seen in cattle and buffalo. This allows farmers\producers a quick interval of selling part of their flock and generating cash income. Goats can also efficiently survive on available shrubs and trees in adverse harsh environment in low fertility land where no other crop or animal can survive. In view of high susceptibility of grazing animals to helminthes infection, the control of these gastrointestinal parasites becomes a challenge in the sanitary management of the herd/flock. The economic losses associated with the treatment and control of gastrointestinal parasites and the constant deterioration of the animal's

health due to hematophagous behaviour of some worm species are regarded as low productivity. The gut helminths induce many functional disturbances in the host body including metabolic changes, retardation of growth, weight loss, haemato-biochemical changes and increased

susceptibility to a variety of diseases (Khan *et al.,* 2015; Fleming *et al.,* 2006). However use of Anthelmintic compound does not always lead to the desired effect, because of the emergence of resistance by the parasites as a result of the long term uses of the drug, under dosage and an inadequate control strategy (Sanyal, 2005)

MATERIAL AND METHOD

In the present study prevalence of different helminth infestation was done by examination of faecal samples collected from different farms and flocks subjected to faecal egg count (FEC) over a period through Mc-Master's technique. The FEC data analysed on the basis of following criteria:

Age- 0-3 month, 3-6-month, 6-12 month

Sex- Male and Female

Time- month wise and quarter wise

The positive sample was subjected for calculation of prevalence by using following formula:

$$\text{Prevalence (\%)} = \frac{\text{Number of positive cases}}{\text{Total number of Population examined}} \times 100$$

Collection of clinical samples

Faecal sample of about 5 gm from each animal was collected in a zip lock cover from individual goats per-rectally. Care was taken to avoid intermixing and gross contamination of faecal sample with urine or bedding materials. Fortnight visits were made for collection and for studying the prevalence of nematode helminths. The processing and examining were done macroscopically as well as microscopically by the method as described by Soulsby (1982) and Ruprah *et al.,* (1986). Gross and microscopic examination of collected faecal samples was done for the presence of nematodes. Microscopic examination of faecal samples was carried out as follows.

Direct Smear Technique

A small quantity of faecal material was taken on a glass slide with the help of glass rod and mixed with 4-5 drops of water and covered with coverslip and examined under 10x power of microscope to detect parasitic eggs/ larvae. The samples found negative were subjected to further examination.

Sedimentation technique

About 3-5 gm of faeces was emulsified with 20-30 ml of water in beaker. The content was strained into a sedimentation flask. The flask was filled upto its brim with water or saline and allowed to stand for 20 minutes. The supernatant was thrown off. This process was repeated until the clear supernatant is obtained· After last sedimentation the supernatant was discarded and drop of sediment was taken on clean glass slide, covered with coverslip and examined thrice under low power of microscope to detect the presence of trematode/ nematode eggs/ larvae, as directed by Ruprah *et al.,* (1986).

Quantitative examination

The faecal samples of heavily infected animals were taken up for quantitative examination to estimate Egg Per Gram (EPG) of faeces. The Stoll's technique as described by Soulsby (1982) was used.

Results and Discussion

The current study was conducted in the various regions of Bilaspur. During the study 410 goats were examined of 238 out of which goats 58.04% were found positive from in and around Bilaspur region respectively. The month wise percentage of total positive goats out of total examined goats in region respectively.

TABLE 1: Month wise prevalence of Helminthes infections in goats of Bilaspur district of Chhattisgarh

Month	Total Examined cases	Total positive cases	Prevalence (%) of infection
December	70	55	78.57
January	111	78	70.27

February	30	15	50
March	32	14	43.75
April	30	15	50
May	25	9	36
June	35	17	48.57
July	37	11	29.8
August	40	24	60
Total	410	238	58.04

The overall of prevalence of gastrointestinal helminthes recorded respectively and showed prevalence of different helminthic infection in goats in Bilaspur district of Chhattisgarh.

TABLE 2: Prevalence of GI helminths in Goats in and Bilaspur District of Chhattisgarh in 2019-2020

Month	Total Examined cases	Total positive cases	*Paramphistome*	*Ostertagia*	*Strongyle*	*Trichuris*	*Strongyloides*	*Moniezia*
December	70	55	5 (10.42)	8(27.90)	5(10.34)	-	-	10(32.01)
January	111	78	7 (24.53)	5(12.01)	4(12.40)	7(23.90)	2(5.51)	7(24.81)
February	30	15	6 (14.65)	2(4.98)	3(10.67)	8(26.60)	3 (10.01)	9(27.98)
March	32	14	8 (27.98)	4(12.49)	2(5.40)	6(14.51)	5(11.18)	7(24.62)
April	30	15	6 (15.01)	3(10.98)	3(10.78)	4(14.28)	3(10.71)	6(15.86)
May	25	9	9 (28.42)	2(5.84)	1(3.02)	6(24.01)	3(11.01)	3(10.45)
June	35	17	9 (28.39)	1(2.98)	3(12.01)	8(28.01)	4(12.56)	2(5.08)
July	37	11	8 (26.40)	1(2.49)	5(11.42)	8(28.01)	6(17.81)	4(15.01)
August	40	24	6 (14.54)	3(10.01)	6(14.42)	10(30.01)	8(27.84)	6(14.43)
Total	410	238	64 (190.34)	29(89.68)	32(90.46)	57(189.33)	34(106.63)	54(170.25)

The variability in the overall prevalence of gastrointestinal helminth's infections might be due to variation in agro- climatic conditions of the region which could affect the survivability and development of infective larval stage of nematode parasites. Further the use of various anthelmintic and grazing practices adopted might have contributed for the difference in the rate of incidence (Getachew *et al.,* 2017).

In conclusions the current study was carried out in and around Bilaspur district of Chhattisgarh state. The overall prevalence percent of gastrointestinal helminth was 58.04% with its maximum intensity in the monsoon season respectively. Low prevalence in winter season was due to reduced grazing hours of the animals, which helps in reducing the chances of contact between host and parasites.

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