#### MURRINA

By

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## MURRINA, DEFINED

When a severe disease of any nature affects the larger domestic animals in Panama, the natives call it "Murrina". This may lead to confusion as the term, "Murrina" also is applied specifically to equine trypanosomiasis. This latter interpretation is implied in the following discussion. When murrina results in posterior weakness, incoordination or paraplegia, the condition is known locally as "Derrengadera".

### HISTORICAL DISCUSSION

Representatives of some of the older families in Panama believe that murrina, of the same clinical picture as exists today, was indigenous many years prior to Darling's discovery of the disease in 1909; at times sweeping as a veritable scourge through the horse population especially in the eastern part of the Isthmus.

Until the last few years this disease was reported as being confined principally to the area east of the Canal region. Since 1930, however, it has extended west as far as Santa Maria and to the vicinity of Santiago. How far this disease may travel north with the opening of the proposed International Highway, is of course problematic.

Following are some of the popular beliefs concerning murrina:

(a) That it was first encountered during the Canal construction period.

- (b) That it was brought to the Isthmus in a race horse imported from South America.
- (c) That it occurs in five years cycles.
- (d) That it develops especially during the dry season when streams dry up and animals of necessity must drink from stagnant pools of water.
- (e) That murrina outbreaks usually start near the seacoast and spread inland.
- (f) That murrina existed locally prior to the first importation of Indian cattle.

# ETIOLOGY

The pathogenic organism of murrina is described as being Trypanosoma hippicum, Darling. Morphologically this parasite simulates T. evansi. One authority believes that murrina is a modified form of surra, and to be in close relationship to other equine trypanosomiases of South America.

In fact, some of the symptoms of positive cases of murrina studied by the author in Panama, seemed to vary so greatly in character in different localities, as to raise the question as to whether we are dealing with more than one trypanosomal infestation in the general entity, "Murrina",—or if appreciable strain variations of *T. hippicum* occur.

It is interesting to note that Chagas disease of man, due to Schizotrypanum cruzi, also is present in Panama.

# ANIMALS AFFECTED

Many species of quadrupeds, both domestic and wild, are susceptible to the disease.\(^1\) Murrina is quite fatal to equines, although mules and donkeys occasionally recover spontaneously. Cattle may act as carriers of the disease for several months without becoming ill.\(^9\) During this period of time they are reservoirs of infection, available to vectors, especially vampire bats. Reservoirs of lesser importance include swine, sheep, deer and cats.\(^2\)

In Panama, murrina affects principally the range horses. Frequently these animals are grouped by the owners into herds

#### as follows:

- (a) Broodmares, young colts, fillies and sometimes a few stallions.
- (b) Surplus saddle animals and young males.
- (c) Adjacent to the ranch house usually there are kept a few special mounts, and one or more stallions.

At times just one of these classes or herds as obtaining to a particular area, will have murrina.

## TRANSMISSION

In 1931, Dunn of the Gorgas Memorial Laboratory began a study of the susceptibility of various bats to *T. hippicum* in infection.<sup>3</sup> In 1932 Dunn definitely proved the vampire bat to be a vector of murrina.<sup>4</sup>

At present this blood ingesting bat which is common to Tropical America, is considered to be a most important biological vector of T. hippicum. In the vicinity of its home in caves, rock crevices and hollow trees, it feeds nightly on the blood of accessible animals, and when necessary may fly several miles to secure its blood meal.

Bat bites on horses and cattle are noticed especially in areas containing brush and trees. However, bat bites on an animal are not in themselves an indication of the presence of murrina.

Vampire bats which have acquired *T. hippicum* infection usually die from the disease, in from ten days to four weeks. During its period of infection this ambulatory vector is a dangerous disseminator of murrina to susceptible animals on whose blood it feeds.

Murrina can be transmitted mechanically also by flies and other agencies, wherein fresh blood from a positive case is carried or applied immediately to an active abrasion or wound in the skin of a susceptible animal, or when kept in prolonged contact with, or massaged into, a mucous membrane. Breeding of a positive case is regarded as a source of danger in the dissemination of the disease.

#### DIAGNOSIS

Blood Surveys.—Upon the discovery of murrina following a routine periodic regional blood survey, or when reported by a livestock owner, immediately thereafter an inclusive blood survey should be made of all equines on the farm or in the section involved, using the thick blood film method,<sup>5</sup> 6 and this supplemented by a physical examination for the detection of clinical cases.

Microscopic examination of fresh wet blood films in the field also may be employed to expedite the inauguration of treatment and other control measures.

The causative parasite is not always present in thick blood films from positive cases of murrina. During the incubation period of the disease, from a few days to more than a week may elapse after infection of the animal takes place, before *T. hip-picum* may be demonstrated in the blood films. At times in the early stages of murrina, and sometimes in advanced cases, the parasites may be absent in thick blood films.

During the disease, the number of trypanosomes in the blood appears to increase and then to subside, in cycles of from four to eleven days. However, these cycles vary considerably and sometimes in the latter stages of the disease are not present. These alternate periods of increase and decrease in the number of *T. hippicum* in the blood stream, are believed to be due to the partial destruction of the parasites by Lysins, or products associated with acidosis.

From a practical standpoint, successive examinations of thick blood films secured at seven-day intervals from animals under observation usually are sufficient to demonstrate trypanosomes, and to establish a definite diagnosis for positive cases of murrina, in from several days to two weeks prior to the appearance of clinical symptoms.

The method used at the Gorgas Memorial Laboratory for their own convenience in field surveys to record the number of protozoa found in thick blood films, each estimated to contain approximately one-thousand fields using a one-twelfth objective, follows: Four plus (++++)—above 400 parasites per microscopic field (pf="per field.").

Three plus (+++)—from 100 to 400 pf.

Two plus (++)-from 25 to 100 pf.

One plus (+)—from 1 to 25 pf.

Few (f)—about one parasite to every alternate field, or about 400 parasites in the thick blood film.

Very few (vf)-from 80 to 300 per film.

Below "v f," the total number of parasites in the film are noted.

Negative (N)-no parasites in the film.

Trypanosomes-T.

Microfilariae-F.

Spirochetes—Sp.

Piroplasma-P.

e. g. T ++ would be "Trypanosomas two plus."

T 26 indicates twenty-six trypanosomes in the film.

Symptomatology.—Physical examination of a case of murrina does not always reveal the disease as such; an animal may have the infection two or more weeks before symptoms appear. Also it is difficult to differentiate murrina clinically inasmuch as most of the usual symptoms—depression, intermittent fevers, unthrifty appearance, progressive loss of flesh, anemia, anasarca, partial depilation, posterior incoordination or decubitus,—may be seen when certain other diseases or conditions are present.

The clinical picture may vary. In two separate outbreaks acute keratitis and coryza were present in several cases and seemed to be associated with murrina. One case showed amaurosis which disappeared upon treatment for trypanosomiasis, while another case acquired a distinct cough.

Sometimes an affected animal will become emaciated and die without any posterior weakness or paraplegia. Occasionally such a case will walk continuously in a circle for one or two days before death.

During a period of three years' observation, no cases of derrengadera were seen in any of several outbreaks of murrina confined to the area east of Panama City.

One horse with posterior incoordination was negative to

blood examination for murrina. The cause of this condition was undetermined, and the animal made an eneventful recovery without treatment.

One owner attempted to distinguish murrina cases, based on their state of nutrition. He selected several of the most unthrifty horses of a lot under primary survey and destroyed them. The blood of all of these selected animals, however, was negative upon microscopic examination. At the same time, other horses in the same corral, but in a good condition of flesh, were found to be positive cases.

Other Diagnostic Methods.—In addition to microscopic bloodfilm and physical examinations for the detection of murrina, or to establish that an animal is negative to the disease, recourse may be made to guinea pig or other animal inoculations, and the complement fixation test.<sup>8</sup>

No definite lesions especially indicative of murrina were found on necropsies of positive cases.

#### CONTROL

It is important to establish an early diagnosis of murrina in order that immediate control measures may be put into effect. Initial blood surveys and physical examinations should be made as outlined under Diagnosis above, and when necessary to expedite the treatment or destruction of horse reservoirs of the disease, fresh blood film examination or complement fixation tests may be conducted.

As soon as practicable after the receipt of the laboratory report covering a blood survey, and with the owner's consent, intravenous treatment should be given to all positive murrina cases with a distinct possibility of recovery, and which would be economical to treat, using the naganol-tartar emetic combination described under Remedial Treatment, as given below.

If for any reason suitable treatment is not given a positive equine case, its immediate destruction should be recommended to eliminate it as a reservoir of infection.

Following the initial survey, and treatment of murrina cases in an infected lot of horses and mules, successive thick blood film surveys and further treatments of positive cases in such lot or area, should be made at regular weekly intervals until the disease is eradicated.

When several positive cases are found in a small horse herd, and many bat bites are present on the animals, a series of "blanket" treatments may be given the entire herd, and thus reduce the number of trips and time required to control the outbreak.

Animals free from murrina should be kept away from infected areas. No horse from an infected herd or territory should be sold or moved to a new area, unless first determined to be negative. In this respect other reservoirs of *T. hippicum* present a problem.

The Department of Agriculture of Panama has expressed the following policy:

"Attention of the Department of Agriculture has been called to the fact that a livestock owner wishes to transfer some horses from a murrina infected district where many animals are dying of the disease to a section of Panama where this disease has never existed.

"It is believed that the transportation of these animals from an infected zone to a clean area is dangerous.

"Horses must not be removed from an infected area until they have been confined for 21 days in stables adequately screened against horse flies and bats, during which time thick blood films will be taken every three days for microscopical examination. Also blood will be taken from each horse under observation and inoculated into two guinea pigs.

"Both the quarantine and the above mentioned examinations will be made under the direct supervision of a graduate veterinarian appointed by the Department of Agriculture, and the transportation and other expenses incident to the quarantine and examinations will be at the owner's expense."

When many cattle are present in an infected territory, it may be found advisable, subsequent to the initial treatment of positive equine cases and where conditions permit, to remove all of the horses and mules in the infected herd several miles away to an open plains country, and there segregate them in quarantine, pending continued surveys and treatment until the disease is eradicated.

When no cattle are present in the infected area, to remove the infected horse herd undergoing treatment, may disperse the infection through diseased vampire bats flying into adjacent districts in search of their blood meal.

Usually many adverse conditions are encountered in the control of murrina outbreaks:

Frequently an outbreak may become widespread before discovery. Owners may refuse to cooperate with control measures, not only refusing to permit treatment of positive equine cases, but maintain such active reservoirs until their removal by death. Sometimes an owner is too poor to afford treatment.

Usually owners neglect to gather all of the horses and mules in an infected area for weekly surveys and treatment. Subsequent to the initial survey, usually only a portion of the animal population involved is presented for examination. Frequently owners fail to return sick animals for the second and third treatments.

In one outbreak of murrina where most, but not all, of the horses and mules were collected weekly for surveys and treatment, five months were required to eradicate the disease. In the same area more than a year later, in a second outbreak where more thoroughness was used in collecting the animals weekly, only six weeks were necessary to effect a complete control of the disease.

The resistance of positive cases frequently is greatly lowered from heavy tick infestation, internal parasites, scarcity of food and water in the dry season, the working of a case under treatment or during convalescence, or through sick animals being run several miles to the collecting pen. Acute laminitis cases unable to walk to water or food may die.

Many times an owner fails to carry out treatment as outlined for inflamed anal structures, the result of naganol-tartar emetic treatment, so that frequently screw worm infestation of these parts follows with resultant toxemia and even death of the patient.

Prevention.—At night valuable horses may be placed in box stalls adequately screened against bats. Illumination in stables prevents molestation of horses by bats to some extent. However, seldom are native animals stabled. It is said that occasionally the pinuela, a cactus-like plant, is hung in an inverted position over the backs of stabled horses, and that a type of "saw grass" is utilized in the construction of hog houses, to act as bat deterrents. It is doubtful if the objective is attained in either instance.

As yet no definite steps have been taken locally to eradicate vampire bats through cyanide fumigation of their habitats, as hollow trees and caves.

Army Animals.—It has been found desirable during field exercises to make night marches of public animals across areas of murrina infection to prevent bat bites, and to avoid camping near known murrina districts.

Ten days after the return of Army animals from maneuvers, usually, thick blood films are secured from all animals for laboratory examination. Also, close watch of animals should be maintained at all times and the diagnosis of all fever cases cleared up through laboratory tests prior to administration of any systemic treatment so as not to mask early detection of murrina, if present.

In the event Army animals contract murrina, positive cases may be placed in tightly screened stalls, there treated, and kept until proved negative by daily blood examination and guinea pig inoculation. All other animals of the command should be examined every three days, through the medium of thick blood films, until found negative for a period of three weeks.

It would seem that the minimum requirements for shipping private mounts from Panama to the United States would be evidence of freedom from murrina before shipment, through guinea pig inoculations plus thick blood film examination every three days for three weeks, keeping the animal in a well screened box stall during such tests and until embarkation. The complement fixation test also may be applied.

## HOSPITALIZATION

Where practicable, murrina cases should be hospitalized for treatment in stalls screened against flies, vampire bats and other vectors. Here proper supervision can be given the care, alkalization and other treatment as is indicated. Screening also excludes screw worm flies. Treated cases having sensitized, depigmented skin areas will be protected from insolation. Animals with laminitis, piroplasmosis, and other concurrent diseases and conditions may be observed and corrective treatment given more readily. Ticks can be removed and anthelminitics may be administered when required. Here also, special efforts can be made to maintain a high state of resistance in sick animals through an abundant water supply, plenty of forage of good quality, salt, and cleanliness and other proper care of the patients.

## REMEDIAL TREATMENT

The naganol-tartar emetic treatment as outlined below was used in the field control of murrina during the past three years under the direction of the Gorgas Memorial Laboratory and in cooperation with the Department of Agriculture of Panama.

It is very important that all infected horses and mules should be treated or destroyed. Sometimes medicine is furnished gratis by the Government agencies for treatment of murrina when the owners cannot afford the cost of the medicine used. The naganol-tartar emetic method of treatment has proved valuable both in eliminating reservoirs of infection, and in saving the lives of a large percentage of positive cases treated.

Apparatus and materials used in a field set-up include:

- 1 Wassermann injection apparatus in a box, consisting of of two graduated glass cylinders of 250 c.c. capacity each; five feet of flexible rubber tubing of one centimeter diameter, and one slip attachment for hypodermic needles, inserted and tied in one end of the tube.
- 12 Hypodermic needles, 16 gauge, 3 inches long.
  - 1 Eight ounce cylindrical, wide-mouthed bottle with ground glass stopper two-thirds full of glycerine, for storage of hypodermic needles.
  - 1 Pair of slender forceps.
- 12 Pieces of gauze six inches square.
- 12 Rubber bands, 1/4 inch wide and 31/2 inches long.
- 24 Strong cylindrical bottles of 1 pint size.

- 30 Corks of suitable size for bottles.
  - 8 Ounces of liquor cresolis compositus.
  - 1 Pint of grain alcohol, 95% strength.
  - 1 White enameled bucket in good repair.
  - 1 Pound of absorbent cotton.

A quantity of naganol in 2 gram vials.

- 100 Powders of tartar emetic, 1 gram each put up aseptically.
  - 1 Notebook and pencil.
  - 3 Hand towels.

Rope and containers for equipment suitable for pack transportation.

The bottles of uniform size are washed, drained, filled completely with distilled water; placed in an autoclave, together with the corks in a separate container, and sterilized under 18 pounds pressure for 30 minutes. As soon as the autoclave is opened after sterilization, the bottles are stoppered tightly under aseptic precautions, and permitted to cool slowly. Each bottle now contains about 400 c.c. of sterile distilled water.

Immediately before going to the field for use, each bottle is inspected to exclude those with sediment, mould, defective corks and cracked glass. Bottles passing this inspection are wrapped in newspaper and packed in containers in an upright manner.

New Hypodermic needles, clean, sharp and free from rust, with stylets removed, are immersed in grain alcohol for 30 minutes; dried, and placed into the bottle containing glycerine. Glycerine is more easily removed from needles in the field than is oil or grease.

Field Preparation of Medicine.—It has been found desirable to compound the naganol-tartar emetic solution in the field immediately prior to its administration. For this purpose a windward corner of the corral containing the animals is selected.

A three per cent cresol solution is prepared in the bucket, using well water. This solution is used for cleansing and disinfecting the hands, assembled Wassermann injection apparatus, site of injection on the neck of each patient, and for dipping the necks and corks of stoppered bottles when necessary.

After making an estimate of the amount of the naganoltartar emetic solution which will be required, based on the number of positive cases to be treated and their weights,—one gram of tartar emetic and four vials of naganol (2 grams each) are introduced aseptically into each bottle of water to be used. The bottles are then agitated until complete solution of the ingredients is effected. Just prior to opening these bottles at any time, the necks and exposed portion of the corks are first disinfected either with 95% grain alcohol or with 3% cresol solution.

The Wassermann gravity injection apparatus is assembled and a double thickness of protecting gauze is secured over the open or top end, by means of a rubber band. The apparatus and hypodermic needles are washed out with 3% cresol solution, drained, and again disinfected with 95% alcohol. The needles are again placed in the cresol solution until used. The injection apparatus is then filled aseptically to the 250 c.c., graduation with the tartar emetic solution, and all air bubbles are removed from the rubber tube and connections, to prevent danger from air emboli formation during the intravenous injection. The apparatus from this point is handled by a trained assistant.

Description of Animals.—On blood surveys and during treatment of murrina cases, it is important to identify all animals by recording a complete, detailed description of each animal. It has been found desirable to note color of animals, sex, white and other contrast markings, scars, deformities, brands-hair, paint or permanent, and the names of the animals and owner as well as location (see A.R. 40-2245).

Restraint of Animals to be Treated.—To prevent forcible removal of the hypodermic needle from the vein with resultant infiltration of the tissues of the neck with the naganol-tartar emetic solution (see Complications and Sequelae) due to the struggling of an animal undergoing treatment, great care is given proper restraint of the patient and quiet is maintained during the operation.

All nervous and fractious animals are cast, all four feet tied together, the right side of the animal is placed uppermost, while the head is held flat on the ground.

A quiet animal is blindfolded, a twitch is applied to the upper lip, and an attendant twists the right ear of the patient with his left hand and places his right hand above the horse's nose immobilizing the head, without any lateral bending of the neck. Conversation and loud noises are prohibited.

Injection.—When the patient is restrained in a standing position, the left jugular vein is selected for the injection; and when cast, the right jugular vein. The skin over the jugular furrow is cleansed and disinfected using a piece of absorbent cotton saturated with 3% cresol solution.

Using his left thumb the operator presses over the jugular vein in the lower neck region thus promoting stasis. The hypodermic needle with open lumen is grasped with the right thumb and forefinger about 1½ inches from the sharp end with the bevel face outward, and thrust upward through the skin and into the axis of the embossed jugular vein in the middle neck region, securing a free flow of blood. Care is taken that the needle does not penetrate through the vein.

Pressure is released from the vein, and the air is expelled from the end of the injection tube, which then is affixed to the needle in the vein. The assistant elevates the cylinder of medicine, and the naganol-tartar emetic solution flows by gravity into the vein.

Dosage.—The average dosage used by the author for each injection of native animals, follows:

Weight of Animal	Number c.c. of Solution	
(pounds)	Injected	
500		
400		
300	120	
200		
100	50	

In one outbreak colts one month old were affected.

During the time of injection great care is given that the needle remains in the vein. Meanwhile the assistant checks the flow of the medicinal solution and indicates the graduation attained by the solution in the cylinder.

At the completion of the treatment, the rubber tube is compressed to stop the flow of the solution, detached from the needle and handed to the assistant. A small amount of blood is permitted to flow through the needle, which is now quickly removed from the animal, and washed.

Massaging the site of injection is contra-indicated. However, any blood contamination of the neck region is washed off with cresol solution, after which the animal is released from restraint.

Where animals with murrina under range conditions have received three injections or naganol and tartar emetic at week intervals, recoveries have varied from 60 to 100 per cent, with an average of about 80%, with but few recrudescences.

Recovery from an attack of murrina does not prevent subsequent attacks from new infection. Relapses sometimes occur after treatment possibly due to an insufficient amount of concentration of the naganol-tartar emetic solution to reach some remote recess or space of the central nervous system or other tissue containing trypanosomes.

# COMPLICATIONS AND SEQUELAE

Injection of any of the naganol-tartar emetic solution into the neck tissues outside of the jugular vein may result in acute peri-phlebitis, cellulitis, phlegmon or abscess formation, and sometimes leading to rupture of the jugular vein and death of the animal.

As a result of the treatment usually the anal tissues become inflamed, excoriated, sometimes fissured, and occasionally there is a partial rectal eversion. When the owner neglects to treat these parts as directed, frequently screw worm infestation follows, with destruction of tissues, difficult defecation, toxemia—and in some cases,—death of the animal.

Laminitis which sometimes occurs as a complication, also depilation, may be aggravated by the treatment.

Depigmented skin areas subjected to solar rays following treatment, frequently become inflamed, edematous; ulcerate, and bleed.

Wounds and abrasions attending decubitus usually are aggravated from the treatment, and frequently necrosis of the tissues involved results.

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