Hemorrhagic septicemia

Disease Monograph Series – 17

Bacteria | *Pasteurella multocida* | Cattle | Buffalo

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Table of Contents

Acronyms

AlOH	Aluminium hydroxide
AU	African Union
AU-IBAR	African Union Inter-African Bureau for Animal Resources
AU-PANVAC	African Union – Pan African Vaccine Centre
APV	Alum-precipitated vaccine
BPM	Bovine pneumonic pasteurellosis
CI	Confidence Interval
CVO	Chief Veterinary Officer
DG	Director General
DoI	Duration of immunity
DVS	Director Veterinary Services
ELISA	Enzyme-linked immunosorbent assay
FAO	Food and Agriculture Organization of the United Nations
HS	Hemorrhagic septicemia
IM	Intramuscular
IN	Intranasal
Lkt	Leukotoxin
LPS	Lipopolysaccharide

ME	Multiple emulsion
NGO	Non-governmental organization
OAV	Oil-adjuvanted vaccine
OIE	World Animal Health Organization
OMP	Outer membrane proteins
PCR	Polymerase chain reaction
SC	Subcutaneous
SHF	Small holder farmer
TPP	Target Product Profile

Executive Summary

Although the main focus of the present monograph is Hemorrhagic septicemia, diseases caused by bacteria of the genus *Pasteurella*, namely *Mannheimia haemolytica* and *Pasteurella multocida* in cattle, sheep and goat are of great importance especially in Africa, where large number of vaccine doses are sold annually. Unfortunately, most of the time these vaccines do not match the circulating strains, due to limited or lack of diagnostic and typing capacity, or do not include key components like leukotoxins. These other Pasteurellosis will also be covered in this monograph when possible.

Disease, etiology, epidemiology and impact

Hemorrhagic septicemia (HS) is an acute, often fatal and septicemic disease of mainly cattle and buffaloes, caused by *P. multocida*. It is important in tropical regions of the world, especially in Asian and African countries. HS epidemics are devastating, and jeopardize not only the economic return of animal production, but more importantly, animal traction, and the harvest of vital crops such as rice. Cattle and buffalo are the major reservoir hosts. Outbreaks have been reported occasionally among pigs in some Asian countries, and cases are seen infrequently in sheep and goats. HS is caused by two serotypes of *P. multocida* designated B:2 (Asian serotype), and E:2 (African serotype; although in some African countries, B types have also been isolated).

Cattle and buffaloes, reared mainly as draught animals but also for milk production, are mainly affected. Buffaloes have a shorter course of disease and are believed to be more susceptible. Natural infections are generally by inhalation and/or ingestion of contaminated materials. HS is endemic in many countries. In endemic areas, most deaths are in older calves and younger adults. In non-endemic areas, massive epizootics occur.

A detailed study conducted recently in India, revealed that the morbidity losses account for 23% of the total losses and 77 % are due to mortality. The total economic loss per infected animal due to HS was estimated at Rs 6816 (USD 102) in case of cattle and Rs 10,901 (USD 163) in buffalo. These losses when scaled-up at the national level have indicated a loss of Rs 5,255 crore (Rs 52,550 million – USD 785 million). A different but also recent study, estimated mortality losses per animal at Rs 27,467 (USD 410) and Rs 31,901 (USD 476) for indigenous and local buffalo respectively.

Other pasteurellosis

The nomenclature of the genus *Pasteurellaceae* has been changing, and that has created and still creates some confusion. Annex 2 shows the old and new names. Besides HS caused by *P. multocida*, the other key diseases in ruminants caused by bacteria of the genus Pasteurellaceae are: 1- Bovine Pneumonic Pasteurellosis (BPM)

caused mainly by *Mannheimia haemolytica* A1 and less frequently by *P. multocida* A, 2- Pneumonic pasteurellosis of sheep and goats, caused mainly by *M. haemolyitica* (usually A2) and 3- Septicemic pasteurellosis in sheep, caused by *Bibersteinia trehalosi* (Table 1).

Incidence / Prevalence

HS has a wide distribution with the highest occurrence in South East Asia. The disease is important in Africa, the Middle East and some countries in southern Europe. Incidence and distribution of HS vary greatly from a few cases to high numbers depending on the type of husbandry practices, geographical area and agro-climatic conditions prevailing in a particular region. Except for Bangladesh and India, there seems to be little information on HS available at country level (for the countries of interest) but more information might be locally available (in local languages).

DIagnostics

The diagnosis of HS depends on the isolation of the causative organism, *P. multocida*, generally from the blood or bone marrow of a dead animal by cultural and biological methods, and the identification on the organism by biochemical, serological and molecular methods. Conventionally, the identification of the serotype is carried out using one or more serological methods. Molecular techniques can be used for capsular typing confirmation (currently not available for somatic typing). Most African countries have difficulties in typing, and therefore matching the field strains to the vaccines, but this diagnostic capability has not been explored by country as it was beyond the remit of this monograph, despite that it has a huge impact on the effectiveness of vaccines.

Control

Treatment is of little use once signs of HS have appeared, but could be effective in the early stages. However, treatment is constrained by a variety of practical considerations. Generally, clinical cases are treated with oxytetracycline, co-trimoxazole, a combination of penicillin and streptomycin or sulphaquinoxaline. In recent times, a gradual development of resistance has been observed.

Sanitary control measures include early detection and isolation of new cases and their immediate treatment with antibiotics, deep burial of carcasses or incineration, and the prevention of movements of animals to disease free areas. Vaccination of susceptible animals in endemic areas is the only practical approach to prevent HS.

Measures to be adopted in endemic countries on a prophylactic basis, in the event of an outbreak, and measures necessary for prevention of spread across regional or national borders are discussed in Section 5. No country has ever attempted to eradicate HS. This reflects the belief that the existence of carriers makes eradication too hard. Of the countries of interest, only Mali has said HS is a notifiable disease.

Current vaccines for HS

Vaccination is the most efficient and cost-effective method to control HS. Although the available vaccines are effective in providing protection, disease outbreaks still occur due to low vaccination coverage, particularly of animals kept extensively.

Simple bacterins for *P. multocida* protect relatively well, as they are based on the capsule of *P. multocida.* The OIE recommends to use a local isolate of *P. multocida* representing the prevalent serotype. Seed cultures for vaccine production should contain capsulated organisms.

The commonly used HS vaccines are bacterins: alum-precipitated vaccines (APV), aluminum hydroxide (AlOH) gel vaccines, and oil-adjuvanted vaccines (OAV). To provide sufficient immunity with bacterins, repeated vaccination is required. Administration of dense bacterins can give rise to shock reactions, which are less frequent with the APV and almost nonexistent with the OAV. The APV provides immunity for 6 months, while the OAV provide immunity for up to 1 year. However, the OAV have not been popular because of their high viscosity making it difficult to use.

A live attenuated vaccine administered intranasally has been used extensively in Myanmar. It was recommended by the Food and Agriculture Organization of the United Nations (FAO) as a safe and potent vaccine for use in Asian countries. However, there are no reports of it being used elsewhere.

Vaccine potency: The OIE recommends back passaging of the master seed in calves before producing the vaccine (only subculture once or twice). It has been demonstrated that back passaged vaccine seed provides better immune efficiency than the original stock culture, and therefore it is recommended the back passaging of the seed vaccine at least once in 6 to 8 months in calves, which is laborious and expensive.

Cross protection: The protective immunity for *P. multocida* resides in the capsule. There is very little, if any cross protection between capsule types. More recent studies also question role of LPS polymorphism in vaccine failure in poultry (not studied for HS). It is interesting to note, that some of the commercial vaccines produced in Africa for HS, seem to have the B (Asian) serotype, instead of the E (African) serotype, and some other do not specify the serotype used. This might be a potential reason for vaccine failure.

Commercial vaccines for other pasteurellosis

Bovine pneumonic pasteurellosis: Vaccines based on virulence factors like leukotoxins (Lkt) seem to work better than vaccines based on serotypes, as the Lkt from cattle is relatively homogenous (the Lkt protects against *M. haemolytica*, not against *P. multocida*). Other vaccines have been developed based on iron regulated proteins, and outer membrane proteins (OMP). The iron-regulated proteins are related to the serotype. A better protection is shown if the iron-regulated outer proteins are derived from a number of different strains, and there is some cross-protection between unrelated strains. Modern vaccines use culture supernatants containing leukotoxin (Lkt) and other soluble antigens, or bacterial extracts, alone or combined with bacterins. Some

vaccine manufacturers claim that Lkt based vaccines protect cattle against nearly all *M. haemolytica* infections. But others say that these modern vaccines have 50–70% efficacy in prevention of *M. haemolytica* pneumonia. It is interesting to note that the majority of African vaccine manufacturers do not mention the inclusion of Lkt or other proteins, while the international pharmaceutical companies include them in all their vaccines. From the information collected, only OBP (South Africa) produces a Lkt based vaccine in Africa. If they are not included in the current African vaccines, it might be valuable to consider their incorporation to improve efficacy.

It has also been noted that the use of the name pasteurellosis creates confusion. For example, one country replied in the questionnaire that they are importing *a P. multocida* based vaccine for control of BPM, which would not be effective, as it is not based on *M. haemolytica*.

Pneumonic pasteurellosis of small ruminants and Septicemic pasteurellosis of sheep: Because sheep strains show much more variability between different strains of any single serotype, leukotoxin type vaccines do not work as well as they do in cattle. The existing leukotoxin vaccines are based on the cattle *M. haemolytica* A1 strain of the Lkt, which may be the same or may differ from the Lkt of the strain which is infecting the sheep. Serotype based vaccines, seem to work better in sheep. There are only 4 commercial vaccines for small ruminants produced in Africa. Two of them are based on *P. multocida* only so their efficacy is questionable. Only one contains *B. trehalosi*, the cause of septicemic pasteurellosis.

A big multinational pharmaceutical company manufactures a vaccine for sheep based on iron-regulated proteins, which contains 9 serotypes and is available in South Africa.

In the databases researched, there was no mention of BPM or small ruminant pasteurellosis vaccines produced in Asia.

Potential new vaccines and the way forward

For the control of HS, vaccines with longer duration of immunity, easy to use and with no side effects would be desirable. Characteristics of an ideal HS vaccine, can be seen under the Target Product Profile in Section 9.

Live attenuated vaccines look promising, as they would overcome some of the issues. Would be good to understand why the strain used in Myanmar hasn't been used elsewhere. There have been developments on live attenuated strains in Iran, in Malaysia and UK. Unfortunately, none of the researchers provided an update on their developments. As for DIVA vaccines, the candidate from Dr Saxena looks interesting, but detailed evaluation of the technical information is needed (not published yet). The candidate from Prof Singh, has not yet been evaluated in the target species and also needs detailed analysis of the technical data.

There are groups working on DNA and sub-unit vaccines, but they seem far from commercialization. There is also a group working on improved delivery system with nanoparticles.

Besides all the new technologies, there is a great need for improvement of the currently used vaccines (ensuring use of the correct serotype, back passaging in calves, etc.), especially on the African continent. Most of the African vaccine manufacturers do not have Pasteurella typing capacity (capsular and somatic typing capacity). If properly empowered and capacitated, AU-PAVAC could provide the required serotyping service to all vaccine manufacturers. AU-PANVAC has also expressed the need in capacity building of the African vaccine manufacturers in bacteriology aspects which goes beyond Pasteurella (for example there is a need to train the labs in terms of isolation and characterization of the local strains of clostridum for Black leg vaccine). This would seem less of a challenge in Asia but needs to be confirmed.

Clinical disease overview

Etiology & Epidemiology

Historically, the term pasteurellosis has been used to indicate disease caused by bacteria of the genus *Pasteurella*. The genus *Pasteurella* is a member of the *Pasteurellaceae* family, which includes a large and diverse group of Gram-negative *Gammaproteobacteria*, whose members are not only human or animal commensals and/or opportunistic pathogens but also outright pathogens. Ancestral relationships among bacterial taxa within the *Pasteurellaceae* family can be inferred by comparing their 16S rRNA genes (See Annex 1). Comparative genomic and phylogenetic analyses of the *Pasteurellaceae* have revealed that many members of this highly diverse family were poorly classified. Indeed, a number of the *Pasteurellaceae* have already been renamed including *Mannheimia* (formerly *Pasteurella*) *haemolytica* in 1999 and *Bibersteinia* (formerly *Pasteurella*) *trehalosi* which was renamed in 2007. However, as can been seen from the 16S rRNA phylogenetic tree shown in Annex 1, further reclassification or renaming may be warranted [1]. Current and old names are shown in Annex 2.

Although the reclassification makes continued use of the term pasteurellosis awkward, new terms have not emerged yet. Table 1 shows the different diseases in cattle and small ruminants with the causative agents.

P. multocida isolates are classified based on a combination of capsular polysaccharide serotyping, which distinguishes isolates into one of the five capsular serogroups A, B, D, E and F, using indirect hemagglutination. Isolates are also subtyped based on their lipopolysaccharide (LPS), which separates isolates further into 16 serovars using the agar gel immunodifusion test. Isolate designations usually consist of a capsular serogroup letter followed by a somatic serovar number (e.g., A:1, A:2, A:3, B:2, etc.).

Table 1: Feasibility of OIE recommended sanitary control measures in smallholder poultry settings.

Host species	Disease	Organisms involved (most commonly)
Cattle & buffalo	1- Hemorrhagic septicemia	• Africa: Mainly *Pasteurella multocida* E:2 • Asia: *Pasteurella multocida* B:2

Cattle	2- Pneumonic pasteurellosis or mannheimiosis (Shipping fever, bovine enzootic pneumonia)	• *Mannheimia haemolytica* A:1 (main cause) • *Pasteurella multocida* A (less frequently involved)
Sheep & goats	3- Pneumonic pasteurellosis	• *Mannheimia haemolytica* (mainly A:2)
	4- Septicemic pasteurellosis	• *Bibersteinia trehalosi* (used to be called *Pasteurella trehalosi* and before *P. haemolytica* T)

Pathogenomics of P. multocida

The genomes of six different *P. multocida* have been sequenced, three from poultry, two from pigs and one from cattle. For cattle, the strain *P. multocida* 36950 was obtained from the lung of a bovine with respiratory infection. A number of genes or gene clusters, have been implicated as important for virulence of *P. multocida*. Some of these genes encoding putative virulence factors are universally present in all six *P. multocida*. These include genes encoding outer membrane proteins (*ompA*, *oomph* and *ompW*), iron acquisition proteins (*exbB-exbD-tonB*, *hgbA*, and *fur*), thiamine metabolism proteins (*tbpA*, *thiP*, and *thiQ*), and the adhesion/Flp pilus assembly cluster (*tadZABCDEFG*). Homologs of the *tad* gene locus are also present in many other *Pasteurellaceae* and Gram-negative bacteria, where they play key roles in biofilm formation, colonization, and pathogenesis.

Unique genes correlated with virulence are present in almost each of the sequenced *P. multocida* genomes. For instance, *P. multocida* strain 36950, contains the large integrative conjugative element (ICE) ICE*Pmu1* of 82 kbp that carries 88 genes, including 12 antimicrobial resistance genes. This ICE is not found in any of the other five sequenced genomes; however, a similar ICE was found in *Histophilus somni* 2336 and *Mannheimia haemolytica* PHL213, both of which are bovine respiratory pathogens and thus share the same host niche as *P. multocida* strain 36950.

Clinical Signs

Pasteurella species cause numerous endemic and epizootic diseases of economic importance in a wide range of domestic and wild animals and birds. *P. multocida* is a common commensal or opportunistic pathogen found in the upper respiratory tract of most livestock, domestic and wild animals. Under stress conditions, the commensal *P. multocida* becomes a pathogen by overwhelming the immune system of the host, proliferating in the nasopharynx and spreading to the lungs. The clinical signs vary from asymptomatic or mild chronic upper

respiratory inflammation to acute, often fatal, pneumonic and/or disseminated disease. The main diseases caused by the genus *Pasteurella* in cattle and small ruminants are:

- 1-Hemorrhagic septicemia (HS): Is a major disease of cattle and buffaloes characterized by an acute, highly fatal septicemia with high morbidity and mortality (both vary between 50 and 100%). These two species are also the major reservoir hosts. Outbreaks have been reported occasionally among pigs in some Asian countries, and cases are seen infrequently in sheep and goats. Goats have been infected experimentally. HS has also been reported in bison, African buffalo, camels, elephants, horses, donkeys and yaks. Experimental infections are readily established in laboratory rabbits and mice. HS is caused primarily by serotypes B:2 and E:2 (Carter and Heddleston system), corresponding to 6:b and 6:E (Namioka-carter system). In Africa it is associated to *P. multocida* type E:2 but some African countries have also reported B:2 (Namibia in 1997, Cameroon in 1993 and Zimbabwe in 1990). In Asia, the only reported serotype is B:2. HS may be asymptomatic until the onset of the acute stage, which is characterized by a rapid onset and progression. Symptoms include fever, lethargy, edema, copious salvation, lacrimation, and nasal discharge, followed by respiratory distress, septic shock with widespread hemorrhaging, and death within 1-3 days. Buffaloes have a shorter course of disease, and are generally believed to be more susceptible than cattle. Animals between the ages of 6 and 24 months are the most severely affected during outbreaks. There are direct losses associated to mortality, but also indirect losses associated to loss of productivity, milk, meat, draught power, cost of alternate sources of draught power and impairment of the reproductive potential. HS is endemic in the tropical and subtropical regions the world including Asia, India, the Middle East and Africa. In endemic areas, most deaths are confined to older calves and younger adults. In non-endemic areas, massive epizootics occur. Case fatality approaches 100%, if treatment is not carried at an earlier stage.

- 2- Bovine pneumonic pasteurellosis or mannheimiosis (BPM): *M. haemolytica* is the main agent responsible for BPM. It is an acute fulminating, fibrino-purulent pleuropneumonia with hemorrhage or coagulation necrosis, due to intense LPS-induced inflammation and production of a ruminant-specific leukotoxin. It is an economically significant disease of cattle, accounting for about 30% of the total cattle death in the world. *M. haemolytica* type A exists in 12 different serotypes (1-17 except 3, 4, 10, 11 and 15). A1 and A6 are the most frequent isolates from pneumonic lungs [2]. Several serotypes have been isolated in Africa: Kenya reported predominance of serotypes A1 and A2 (1978), and South Africa (1995) 30% untypable and 40% A1. Also serotypes 14, 6, 2 and 15 were identified.

- 3- Pneumonic pasteurellosis in small ruminants: Small ruminants are particularly susceptible to respiratory infections complicated by *M. haemolytica* and it is the most common form of respiratory disease in sheep and goats. The disease occurs worldwide in temperate, subtropical and tropical climates, although the prevalence of serotypes may vary by region and flock. Serotype A2 is a common cause of pneumonic pasteurellosis in sheep, but there is also an increased prevalence of serotypes A5, A6 and A7 [2]. In South Africa, in a study done in 1995, serotype 6 was the most prevalent, followed closely by types 9 and 2 [3].

- 4- Septicemic pasteurellosis in sheep: *B. trehalosi* causes systemic pasteurellosis only in sheep and the main feature is sudden death. It exists in 4 serotypes, T3, T4, T10 and T 15.

Transmission:

Is through direct contact with nasal secretions, where a chronic infection ensues in the nasal cavity, paranasal sinuses, middle ears, lacrimal and thoracic ducts of the lymph system and lungs. The organism is capable of surviving in moist soil and water for up to 2-3 weeks. Preexisting or coinfection with other respiratory pathogens, particularly *Bordetella bronchiseptica* or *Mannheimia haemolytica*, significantly enhances colonization by *P. multocida*, leading to more severe disease.

Primary infection with respiratory viruses or with *Mycoplasma* species also predisposes animals to secondary infection with *P. multocida* and/or *M. haemolytica*. Environmental conditions, stress, and the overall health of the animal also appear to play important roles in disease severity and likelihood of transmission

In endemic areas, large numbers of animals that survive after an outbreak become latent carriers. They intermittently shed the organisms but, since the herd immunity is also high, there are no new clinical cases. A new outbreak occurs when a shedder comes into contact with a susceptible animal, which may be one born after the previous outbreak or one introduced into the herd. The chance of a new outbreak increases with time with an increase in size of the susceptible population.

Once the first clinical case occurs, more bacteria are shed and disseminated. Their survival in the environment and transmission to other animals depend on factors such as closeness of contact, hygiene and climate (wet conditions prolong the survival of the causative bacteria outside the animal, making an outbreak more likely). Occasional sporadic outbreaks allow the build-up of nonimmune animals and a major outbreak may occur. Regular seasonal outbreaks result in much higher herd immunity (through frequent exposure) and outbreaks tend to be less significant.

Diagnosis

Until very recently, conventional methods for detection and diagnosis of infection with *Pasteurella* relied on the isolation of the bacteria from the blood or bone marrow of a dead animal, and observation by microscopy using staining and/or isolation by in vitro culturing on selective media, followed by phenotypic and/or serological characterization.

Phenotypic characterization of *P. multocida*, based on morphology, carbohydrate fermentation patterns, and serology, is challenging. Identification of *P. multocida* using biochemical strips (such as API 20E/20NE, Minitek, or Oxi/Ferm strips) remains a rapid method commonly used in diagnostic laboratories, but it has limited accuracy and can lead to confusion of *P. multocida* with *Mannheimia* (*Pasteurella*) *haemolytica*, *H. influenzae*, or other *Pasteurellaceae* species.

Several serotyping tests are used for the identification of the HS-causing serotypes of *P. multocida*. Capsular typing can be done by rapid slide agglutination, indirect haemagglutination, agar gel immunodiffusion tests (AGID) and counter immunoelectrophoresis. Somatic typing can be done by AGID and other tests. Hyperimmune antisera for most of these tests, are prepared against specific reference strains in rabbits.

PCR- plus sequence-based ribotyping analysis using universal primers for 16S rRNA genes, genomics, and other DNA sequence based molecular techniques have now superseded phenotypic methods for identification, characterization, and differentiation of *P. multocida* and other *Pasteurellaceae*. Conventional ribotyping based on PCR amplification alone is still generally considered a reliable and discriminative method for characterizing clinical isolates of *P. multocida*. However, PCR amplification of 16S rRNA genes, followed by sequencing and sequence comparison against known ribosomal databases, such as the NCBI or the RDP (**http://rdp.cme.msu.edu**) database, is now the predominant and most reliable method of taxonomically identifying isolates at the genus and species levels. The OIE mentions multiplex capsular PCR typing systems, and type-B and type A specific PCRs. Molecular techniques for somatic typing don't seem to be available.

A loop-mediated isothermal amplification assay (LAMP) has been developed for the identification of *P. multocida* of pigs [4]. Bhimani and colleagues in 2015, used newly designed primers to identify *P. multocida* in cases of HS, and they demonstrated to be a more sensitive test than PCR [5]. LAMP is a rather newer molecular technique, which can be used for rapid detection of the infectious agent at field level and which does not require sophisticated instruments, i.e. thermal cycler. LAMP requires lesser time than a conventional PCR to perform (the reaction time took only 30 minutes), and results can be read visually. Data on LAMP has also been reported by Moustafa and Bennet in 2014 (**http://pasteuraceae-2014.p.asnevents.com.au/days/2014-05-15/abstract/12799**).

OIE listed tests:

Identification of the agent:

- Cultural and biochemical methods

Serotyping:

- Rapid slide agglutination test (capsular typing)
- Indirect haemagglutination (capsular typing)
- Agar gel immunodiffusion tests (capsular and somatic typing)
- Counter immunoelectrophoresis – a rapid method for the identification of capsular types B and E cultures.
- Agglutination tests (somatic antigen)

Nucleic acid recognition methods

- *P. multocida* specific PCR assay
- *P. multocida* multiplex capsular PCR typing system
- HS-causing type-B-specific PCR assay
- *P. multocida* type A specific PCR

Serological tests (for the detection of antibodies): are not normally used for diagnosis.

Serotyping in Africa: Dr Marijke Henton from IDEXX South Africa (personal communication), said that she used to type both Mannheimia and Pasteurella whilst she was at Onderstepoort, and prepare the antisera as well. There appears to be no commercial sources for typing sera, and since she left Onderstepoort in 2002, the typing of both has fallen into disuse? Disrepair? Anyhow, no fresh sera have been prepared since that time. Many African laboratories including AU-PANVAC do capsular typing by PCR, but have no capability for somatic serotyping (Nick Nwankpa, AU-PANVAC personal communication).

Serotyping is critical for sheep vaccines, as all the available sheep vaccines are still serotype based. Serotyping of isolates is currently the only way to match up the problem on the farm with an available vaccine.

Differential diagnosis

The differential diagnosis of HS includes other causes of sudden death such as lightning strikes, blackleg (*Clostridium chauveoi*) and anthrax. Acute salmonellosis and pneumonic pasteurellosis should also be considered.

Zoonotic disease

There are no reports of human infections with *P. multocida* serotypes B:2 and E:2; however, other serotypes do infect humans, and precautions should be taken to avoid exposure. Humans acquire *Pasteurella* infection primarily through contact with animals, most usually through animal bites, scratches, licks on skin abrasions, or contact with mucous secretions derived from pets. A survey of the literature over the past 30 years suggests that 20 to 30 human deaths due to pasteurellosis occur annually worldwide [1]. Among the *Pasteurella* species, *P. multocida* is the predominant human pathogen encountered, especially in severe disease cases although *P. canis* may be more prevalent with dog bites.

Incidence and Prevalence in Selected Countries

Global

Hemorrhagic septicemia has a wide distribution with the highest occurrence in South East Asia. The disease is important in Africa, the Middle East and some countries in southern Europe. It has also been recognized in Japan and North America. The African form of HS occurs sporadically, it is limited in extent and appears to be associated with stress conditions. The B:2 serotype has been reported in southern Europe, the Middle East, South East Asia and North America. The E:2 serotype occurs primarily in Africa but Namibia, Cameroon and Zimbabwe have also reported serotype B. Both B and E serotypes have been reported in the Sudan and in Egypt. Neither serotype has been found in South America, Central America, Australia or New Zealand.

It is important to note that Table 2 includes South Africa as endemic, but the disease is very rare. In Southern Africa the disease is very sporadic, and mainly restricted to the wetter subtropical regions of Zambia, Zimbabwe and the northern parts of Namibia.

Table 2: Summary of the global distribution of Hemorrhagic septicemia. Source: De Alwis, 1999. http://aciar.gov.au/files/node/2144/MN057%20part%201.pdf

Disease endemic	Low sporadic or exceptional occurrence	Suspected but not confirmed	Probably existed Now free[a]	Never occurred
ASIA Bhutan, China, India, Indonesia, Malaysia, Mongolia, Myanmar, Philippines, Sri Lanka	Malaysia (Sabah)	Kuwait, Qatar	Hong Kong (1984), Israel (1948), Singapore (1930)	Cyprus, Japan, Jordan
AFRICA Central African Republic, Chad, Ivory Coast, Ghana, Guinea, Kenya, Mozambique, Niger, Nigeria, South Africa, Zimbabwe	Angola	Botswana	Algeria, Republic of Congo, Egypt (1970), Eritrea (1993), Mauritius (1989), Morocco, Namibia (1988), Seychelles, Sudan, Tunisia	Cape Verde Libya
AMERICA Argentina, Brazil (part), Ecuador, Falkland Islands, Honduras, Nicaragua, Venezuela	Canada, Jamaica, United States	Antigua and Baruda, El Salvador (1990), Paraguay (1985), St Kitts and Nevis	Barbados, Belize Bolivia, Chile, Cuba, Haiti, Mexico	
EUROPE	Estonia, Latvia, Portugal, Russian Federation, Spain	Albania, Austria, Croatia, Greece	Finland (1993), Germany (1986), Iceland, Italy, Luxembourg, Macedonia, Malta, Poland (1985), Romania (1993), Sweden, Switzerland, Isle of Man, Yugoslavia	Czech Republic, Denmark, France, Iceland Lithuania, Maldova, Slovania, United Kingdom
OCEANIA				Australia, New Caledonia, New Zealand, Vanuatu

Incidence data by country

There are two main sources, OIE and AU-IBAR (which includes only Africa), but data are not always similar.

1- Source: OIE.

Data of outbreaks reported to the World Animal Health Organization (OIE) are shown in Tables 3 and 4. Data are not always reliable, as many countries dosn't seem to report, or report inconsistently over time.

http://www.oie.int/wahis_2/public/wahid.php/Diseaseinformation/statusdetail

Similar information presented in a different manner can be seen in Annex 3.

Number of cases reported to the OIE by disease and by country:

- No information, + Present but quantitative data not known, ? Disease suspected

Table 3: ASIA – Hemorrhagic septicemia outbreaks notified to OIE from the Asian countries of interest.

Country	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015
Bangladesh	-	-	+	+	+	+	+	+	+	+	-
India	775	770	470	359	296	380	315	248	135	131	
Indonesia	+	+	+	+	+	+	+	+	+	-	-
Myanmar	+	>2	27	9	10	3	7	2	5	>1	-
Nepal	374	>215	11	>68	126	113	165	82	52	58	21
Vietnam	553	1,309	1,673	2193	536	239	163	192	116	130	70

Table 4: AFRICA - Hemorrhagic septicemia outbreaks notified to OIE from the Asian countries of interest.

Country	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015
Burkina Faso	33	24	28	33	41	64	39	23	12	18	>9
Ethiopia	53	998	404	349	400	566	370	491	498	154	-
Ivory Coast	-	-	-	-	-	6	4	3	0	0	0

Kenya	0	0	0	0	0	0	0	0	0	0	0
Madagascar	0	0	0	0	0	0	0	0	0	0	-
Malawi	-	-	-	-	0	0	0	0	-	-	-
Mali	0	0	-	-	-	0	0	0	0	1	1
Mozambique	0	0	0	0	0	0	0	0	0	0	-
Rwanda	-	-	-	?	-	-	-	-	-	-	-
Senegal	34	11	7	9	7	11	8	?	7	6?	3
South Africa	0	0	0	?	0	2	1	0	0	0	-
Tanzania	+	>9	13	>1	+	+	+	+	+	2	2
Uganda	-	-	-	-	-	-	-	-	-	-	-
Zambia	-	16	13	30	48	43	+	14	21	26	-

2- Source: AU-IBAR.

Number of outbreaks per year as reported to AU-IBAR and published in the Pan African Animal Resources YearBook. (**http://www.au-ibar.org/pan-african-animal-resources-yearbook?showall=&limitstart**=)

Table 5 shows the number of pasteurellosis outbreaks reported to AU-IBAR, but interpretation should be done with caution. Reporting of Pasteurellosis from AU-IBAR has varied over time. Some years they reported only HS (2007), for some years there are no details (2008, 2009), and in the later years there seem to be a distinction between HS and other pasteurellas.

Table 5: Number of Pasteurellosis outbreaks per year as reported to AU-IBAR and published in the Pan African Animal Resources YearBook. Please see notes below the table for each year. NS= Not specified

Country	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015
Burkina Faso	16	48				63	40	23	18	18	
Ethiopia						769	570	718	1,063	292	
Ivory Coast						7		5			

Kenya											
Madagascar		1				NS	7				
Malawi											
Mali	3					2			1	1	
Mozambique						2					
Rwanda											
Senegal	1	23	15			30	14	7	4	5	
South Africa						4		1		2	
Tanzania								1		1	
Uganda											
Zambia						21	11	10	22	21	

Reported as Pasteurellosis, no details given.

2005: A total of 1,230 pasteurellosis outbreaks were reported in 6 countries, and were restricted to bovine, caprine and ovine.

2006: Seven African countries reported 106 outbreaks of pasteurellosis. Bovines were most affected, followed by small ruminants.

2007: It is noted that the report is on HS. 4 member countries reported a total of 27 outbreaks.

2008: 2,410 pasteurellosis outbreaks from 15 countries were reported to AU-IBAR but no details are given. It says 4 countries reported HS, but no details are given either.

2009: 416 outbreaks of pasteurellosis were reported, and 132 outbreaks of HS.

2010: 994 outbreaks were reported from 23 countries.

2011: 13 countries recorded a total of 1,016 outbreaks. Additionally, 3 countries reported HS (no details).

2012: 18 countries reported HS and other pasteurellosis, with a total of 1257 outbreaks.

2013: 10 countries reported HS and other pasteurellosis, with a total of 1,211 outbreaks. It is clear that the 10 outbreaks relate to HS.

2014: 14 countries reported HS and other pasteurellosis, with a total of 391 outbreaks. The report makes clear that Mali, Senegal and South Africa are not HS, but other pasteurellosis. HS was reported in 9 countries, and other pasteurellosis in 5 countries.

Regional

Outbreak data by country

Incidence and distribution of HS vary greatly from a few cases to high numbers depending on the type of husbandry practices, geographical area and agro-climatic conditions prevailing in a particular region [6]. Outbreaks of disease occur throughout the year irrespective of the season. This is in contrast to the perception that the disease is mostly precipitated during the monsoon or post-monsoon period.

Bangladesh

Number of outbreaks reported based on passive surveillance 2010-2012 [6]:

- 2012: 4,769
- 2011: 5,885
- 2010: 2,782

India

In India, the incidence of disease outbreaks varies considerably in different states and from year to year in each state [6].

2014: There were 93 outbreaks of HS in bovines (304 dead animals), 32 outbreaks in buffaloes (201 animals dead), and 6 in ovine/caprine (6 animals dead). Source: Annual report 2014-2015. Department of Animal Husbandry, Dairying & Fisheries, Ministry of Agriculture, Government of India. **http://dahd.nic.in/dahd/WriteReadData/Animal%20Husbandry%20English%202014-15%20(1).pdf**

1991 – 2010: A epidemiological study was conducted by the Project Directorate on Animal Disease Monitoring and Surveillance (Bengaluru) in 2012. It showed a constant variation in the number of states reporting HS. Also a progressive trend was seen in occurrence of HS since 1992. Maximum outbreaks were reported in 1998. There has been a gradual decline in HS outbreaks since 1998 and many states reported fewer outbreaks during 2006-

2010. The decline in the occurrence can be attributed to the availability of cost-effective programs for prevention and control. Many districts of Madhya Pradesh, Odisha and Rajasthan have reported the disease for the first time during the last 5-year study period. (Figure 1).

http://nadres.res.in:8080/Nadres_Uploads/UploadedFiles//VetEpiReports/ThirdSlot/Tech%20HS%20final.pdf

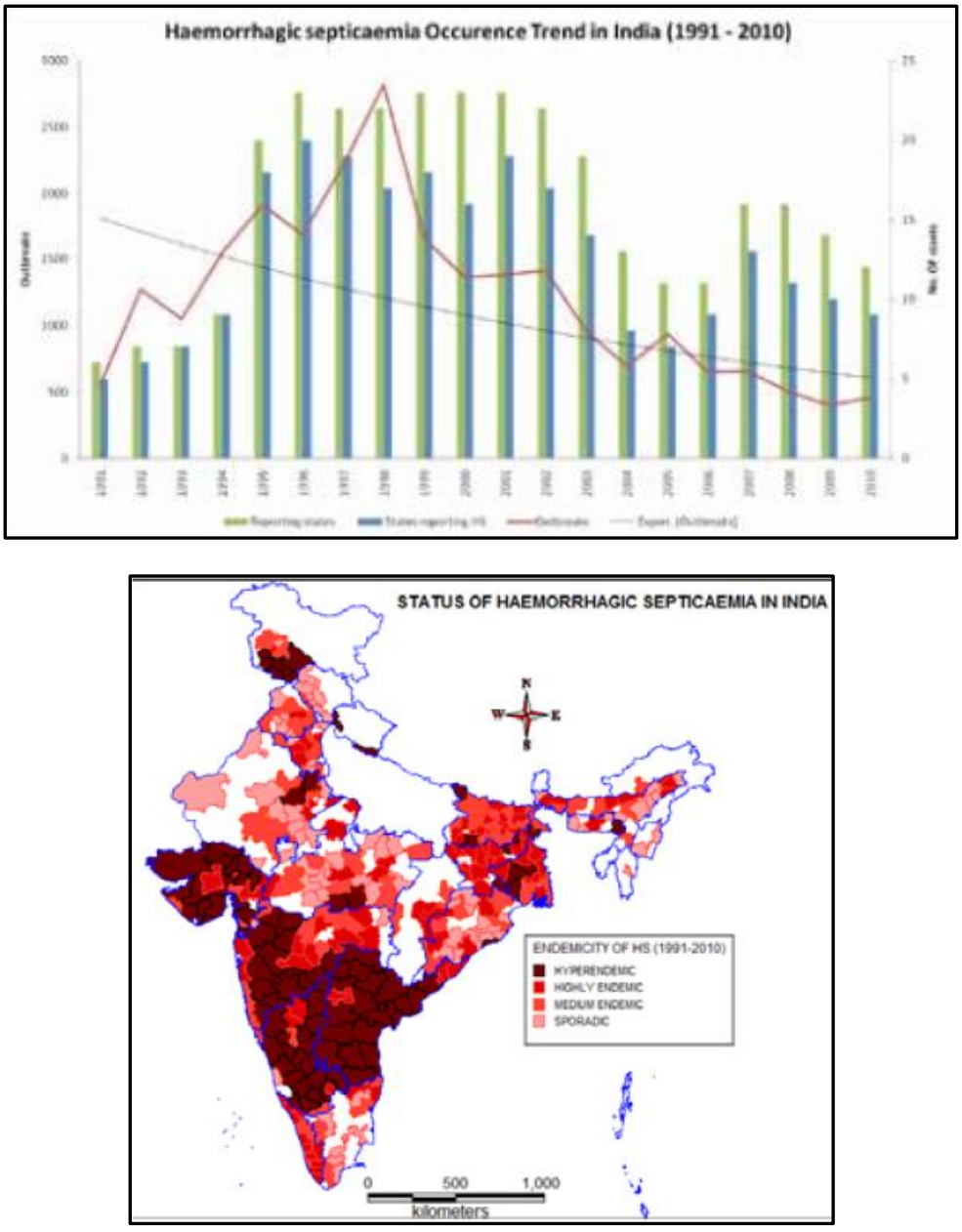

Figure 1: Status of HS in India. (Source: PD-ADMAS/ TECHNICAL BULLETIN/ 11 / 2012)

Indonesia

The disease has been reported in Java Rhinoceros. **http://www.livingfossil.org/science/javan-rhino.php**

Gayo Leus District: 690 deaths have been reported 2013 – early 2015, in 11 sub-districts. The majority of the dead animals were water buffaloes. **http://civas.net/2015/08/29/hemorrhagic-septicemia-in-livestock-in-aceh/?lang=en**

Myanmar (Burma)

No country specific information was found on PubMed or the internet.

Nepal

No country specific information was found on PubMed or the internet.

Vietnam

No country specific information was found on PubMed or the internet.

No data has been found for the African countries of interest.

Prevalence data by country

Bangladesh

Data on the prevalence of HS in Bangladesh, can be seen in Table 6 below.

Table 6: Estimated number of diagnosed cases, prevalence, death cases and vaccination coverage of Hemorrhagic septicemia in livestock in Bangladesh, 2010–2012. Source: Mondal et al, 2014 [7]

Disease	2010	2011	2012	Total
Haemorrhagic septicaemia				
Diagnosed cases	2,782	5,885	4,769	13,436
Prevalence rate, % (95% CI*)	0.18 (0.17–0.18)	0.31 (0.30–0.32)	0.38 (0.37–0.39)	0.28 (0.28–0.29)
Death cases	64	221	101	386
Case fatality rate, % (95% CI)	2.30 (1.74–2.86)	3.76 (3.27–4.24)	2.12 (1.71–2.53)	2.87 (2.59–3.16)
Vaccination	272,395	367,862	433,101	1,073,358
Vaccination rate, % (95% CI)	0.64 (0.63–0.64)	0.86 (0.86–0.87)	1.02 (1.01–1.02)	0.84 (0.83–0.84)

India

The majority of the data found was on number of outbreaks. Only one article was found on prevalence, in the province of Uttar Pradesh.
http://www.indianjournals.com/ijor.aspx?target=ijor:abr&volume=13&issue=2&article=009

The presence of antibodies against *P. multocida* B:2 in 10 (22.2%) serum samples by CIE showed the prevalence of *P. multocida* B:2 infection in the area.

No specific data has been found for the remaining Asian countries, or for any of the African countries of interest.

Economic and Social Impacts at Global and Regional Levels, and in Selected Countries

HS is a disease of utmost economic importance particularly in Asia and to a lesser extent in Africa. Few countries have attempted to quantify the losses due to HS, and there is no uniformity in the methods adopted. Thus, these studies are not strictly comparable, but reflect the trends. Most of the available information is derived from passive reporting systems.

Most estimates of losses take into account only direct losses, i.e. value of animals that die of HS. A true estimate of losses should take into account a variety of factors which constitute indirect losses, including:

a) Loss of productivity – milk, meat, draught power, and cost of alternate sources of draught power.

b) Impairment of the reproductive potential of the animals.

c) A reliable differential diagnosis as there is tendency during the Monsoon to attribute any mortality to HS. Thus a possible over-estimation of these losses should be taken into account. However, it must also be kept in mind that reported losses constitute only a fraction of the actual losses. This is bound to be so since HS is a disease that occurs in situations where husbandry practices are poor and therefore disease reporting system will also be poorly developed.

Asia

A good review has been done in 2002 by Benkirane and De Alwis [8]. In Asia the susceptible animal population consisted of 432 million cattle and 146 million buffaloes at the time of the publication, which constituted 30% and 95% of the world's cattle and water buffalo population, respectively.

Milk production

Cattle and buffaloes are reared mainly for use as draught animals in the rice fields, but in India and, to a lesser extent, Pakistan, milk production is also important (FAO 1994, 1995). In Asia as a whole, buffaloes contribute 37% of milk production but in India, where the production of milk is the highest in Asia, nearly 50% of the milk is produced from buffaloes.

Draught power

Most of the cattle and buffaloes are used as draught animals in the rice fields, and rice is the staple diet in many countries. Where animals are used for draught power, which is a seasonal activity, they are managed in an extensive, free-range system for most of the year. Such conditions are an ideal environment for the spread of diseases such as HS because herds of animals belonging to different owners roam together in common grasslands, drink in common village tanks and are often even paddocked together at night. Such animals are often less well managed, with lower vaccination coverage, than more intensively farmed animals.

Thus, the high population of buffalo in Asia, the high susceptibility of buffalo to HS and the high case fatality, all point to the significance of the economic losses due to the disease (FAO, 1994, 1995).

Individual country data

In **India,** during the past four decades it has been found that HS accounted for 46–55% of all bovine deaths. During the twelve-year period since 1974 to 1986 it accounted for 58.7% of the aggregate of deaths due to five endemic diseases, with foot-and-mouth disease, rinderpest, blackquarter, anthrax and HS.

- More recent data from the NIVEDI Annual report 2014-2015, mentions that in a study done by Govindaraj et al in three districts of Gujarat state (Ahmedabad, Mahisagar and Patan) showed an average case fatality rate among the local buffalo of 78%. The estimated mortality loss per animal was Rs 27,467 (USD 410) and Rs 31,901 (USD 476) for indigenous and local buffalo respectively.
- A detailed study conducted by Singh et al, and published in 2014 [9], revealed that the morbidity losses account for 23% of the total losses and the rest (77 %) are due to mortality of the animals. Of the total morbidity losses, about half have were due to reduction in growth and one-fifth due to loss in milk. The total economic loss per infected animal due to HS was estimated at Rs 6816 (USD 102) in case of cattle and Rs 10,901 (USD 163) in buffalo. These losses when scaled-up at the national level have indicated a loss of Rs 5,255 crore (Rs 52,550 million – USD 785 million). The direct losses contribute 80.3 % and indirect losses contributed 19.7 % to the total economic loss. The study has found that calves contribute 74.8 % and adults contribute 25.2 % to the total economic loss due to HS. **http://ageconsearch.umn.edu/bitstream/196991/2/12-B-Singh.pdf**

In an active surveillance study in **Sri Lanka**, it was shown that in the 1970‘s, around 15% buffaloes and 8% cattle in the HS endemic areas died of HS annually. During the same period, the passive reporting systems recorded only 1 200 to 1 500 deaths a year in a cattle and buffalo population of approximately 2.5 million.

Pakistan reported that 34.4% of all deaths in susceptible stock is due to HS. In 2002, with a cattle and buffalo population of 17.7 and 18.8 million respectively, the annual economic losses were estimated at 1.89 billion rupees (350 million USD). In 2007, the HS prevalence was estimated at 49%. In 2010, there were about 29.9 million heads of buffaloes in Pakistan.

In the South-East Asian region, countries such as Indonesia, Malaysia, Thailand, Myanmar, Laos, Cambodia and the Philippines, rank HS high among the economically important diseases of cattle and buffaloes.

In **Myanmar** it is thought that 50% of the government's effort in animal disease control is directed towards HS.

Malaysia, with a relatively small population of 735,000 cattle and 186,000 buffaloes, estimates the animal losses due to HS to be 2.25 million Malaysian Ringit (0.85 million USD) (FAO, 1979).

In a study in **Bangladesh** in 1996, an attempt was made to compute the economic losses resulting from three important endemic diseases, anthrax, blackquarter and HS. It was found that the direct losses which took into account the market value of the animals that died and the cost of treatment was 2.3 million US dollars. Their computation of indirect losses took into account the value of the rice that would have been produced from the land left uncultivated as a result of loss of draught power. These losses amounted to 148 million USD annually due to these three diseases. It may therefore be concluded that no accurate estimates are available on the actual deaths due to HS, and the available information on direct and indirect economic losses is incomplete.

Verma and Jaiswal, mentioned in their paper published in 1998, recent reports of annual losses due to HS of approximately USD 1.4 million in **Laos**, and approximately USD 1 million in **Malaysia** [10].

Africa

HS is of less economic importance in the African region than in Asia. This is because Africa has less of the world's cattle and buffalo populations. Also, many other animal diseases cause more severe economic losses. These include the African endemic diseases, such as trypanosomiasis (nagana), theilleriosis (East Coast *fever)* and contagious bovine pleuropneumonia. Published reports on economic losses in the African region are scarce.

Analysis by the World Bank:

The World Livestock Disease Atlas – a quantitative analysis of global animal health data [11], published by the World Bank (with cooperation of OIE and FAO) in 2011 is an attempt to understand which livestock diseases cause the heaviest losses, which countries suffers the worst disease-related losses and which livestock species are most affected.

http://www-wds.worldbank.org/external/default/WDSContentServer/WDSP/IB/2012/02/17/000356161_20120217030841/Rendered/PDF/668590WP00PUBL00Livestock0Atlas0web.pdf

The World Livestock Disease Atlas bases its analysis on the Livestock Units (LSU). Each species has a LSU value, and the losses of LSU have been given a value. See Figure 2. For more information on the methodology description, please refer to the World Bank Atlas itself (pages 6 & 7). HS is one of the top 10 diseases causing losses for cattle and buffalos, as shown in Figure 3. However, looking at the data in detail, there are few data from sub-Saharan Africa and Asia.

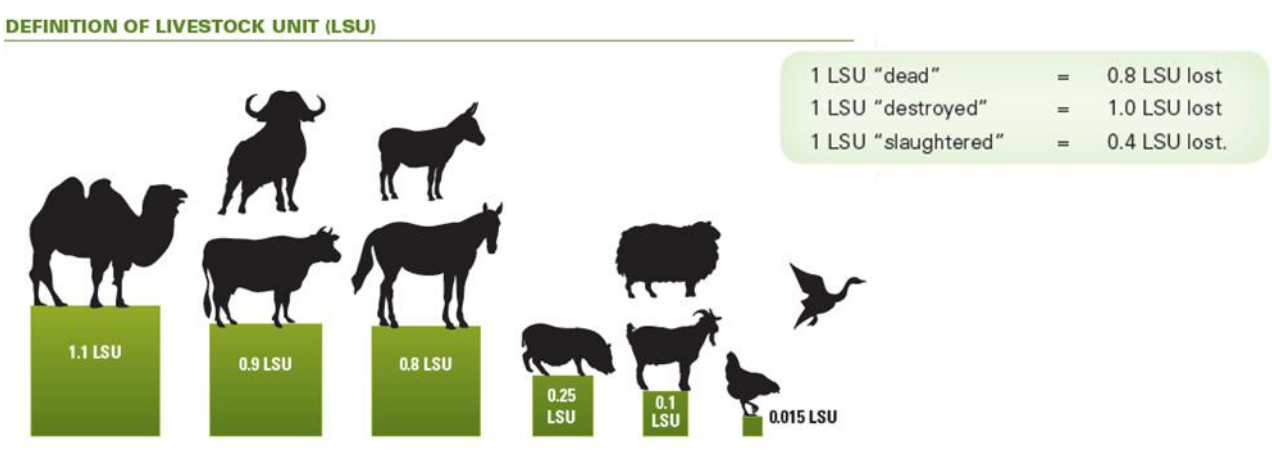

Figure 2: Livestock Units. Source: World Livestock Disease Atlas – The World Bank, 2011 [11].

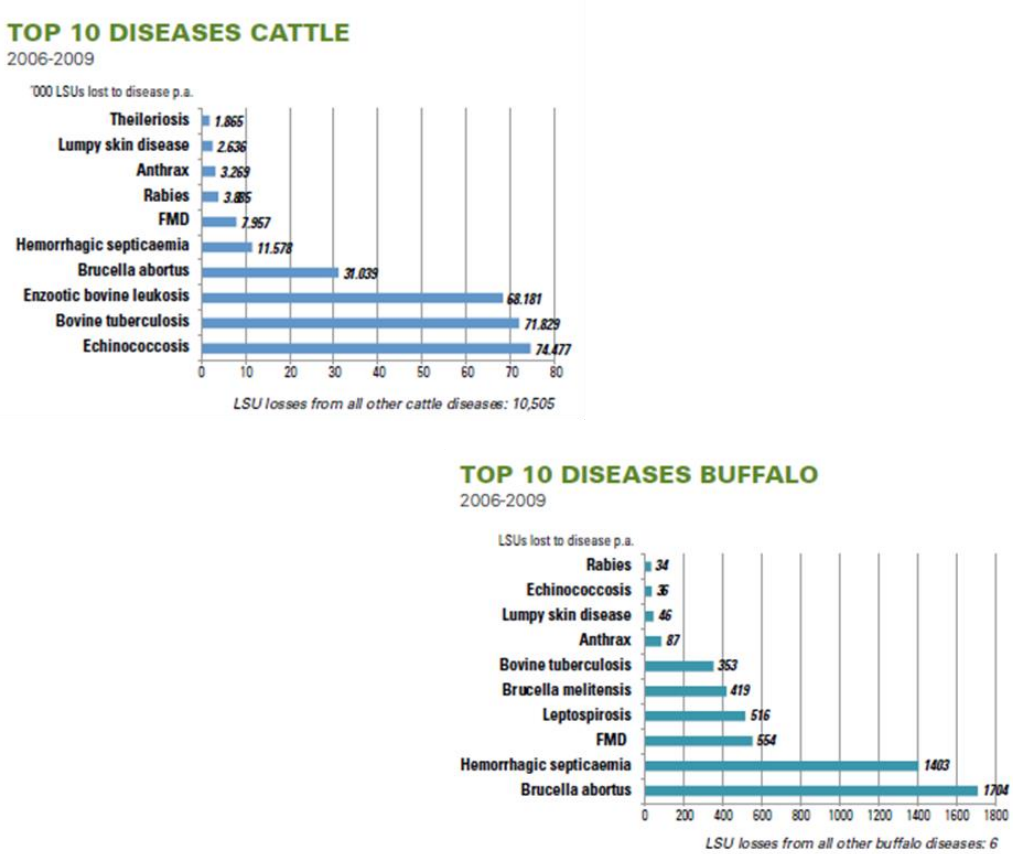

Figure 3: Top 10 diseases in terms of LSU losses for cattle, buffalo, and sheep & goats. Source: World Livestock Disease Atlas – The World Bank, 2011 [11].

Disease Prevention and Control Methods

Treatment (Control)

Treatment is of little use once signs of HS have appeared, but could be effective in the early stages (OIE, 2009). As HS is a primary bacterial disease with no other biological agents involved, treatment may appear simple using the wide range of antibiotics currently available. In reality, however, treatment is constrained by a host of practical considerations. It has been found in practice that animals can only be cured if they are treated in the very early stages of the disease. As the disease occurs mainly in situations with very basic husbandry practices most field cases escape detection in the early stages, thus rendering treatment ineffective. In organized farms, however, a practical method of achieving early detection and successfully treating animals is to check the rectal temperatures of all in-contact animals regularly once an outbreak has been detected. Any animals showing an increased temperature can be separated and treated with a course of an appropriate antibiotic.

Generally, clinical cases of HS are extensively treated with oxytetracycline, co-trimoxazole, a combination of penicillin and streptomycin or sulphaquinoxaline. However, in the recent times, shifts in antibiotic sensitivity spectrum of *P. multocida* are evident and there have been increases in incidences of high morbidity and mortality. Compromised therapy may be due to emergence of multi-drug-resistant strains. A rise in multiple drug-resistant strains of *P. multocida* have been identified, especially for sulphadiazine, the drug of choice in the field for control of HS, as well as many other commonly used antibiotics such as amikacin, carbencillin, erythromycin and penicillin, where there is gradual development of resistance. Strains of *P. multocida* vary in susceptibility to chemotherapeutic agents. Most *P. multocida* isolates were resistant to sulpha drugs with varied sensitivity to chloramphenicol, gentamicin, tetracycline, kanamycin, penicillin-G, streptomycin, sulphonamides and trimethoprim as assessed by agar dilution and determination of the susceptibility of transconjugants and transformants by disc diffusion assay [6].

The injudicious use of antibiotics in developing countries is probably leading to the severe problem of antibiotic resistance, the genes of which may be on R-plasmids. Conjugative R-plasmids are commonly responsible for inter-species and intergenic spread of multiple drug resistance and transfer of such plasmids among pathogenic strains may give rise to epidemic spread of infection. Co-existence and the spread of small plasmids are used by *P. multocida* to become multi-resistant [6].

Prophylaxis (Prevention)

Sanitary measures include early detection and isolation of new cases and their immediate treatment with antibiotics, deep burial of carcasses or incineration, and the prevention of movements of animals to disease free areas. Vaccination of susceptible animals in endemic areas is the only practical approach to prevent HS. The different types of vaccines, and their advantages and disadvantages, are discussed in Section 6. During an outbreak, the whole herd should be vaccinated, irrespective of previous vaccination history.

Options and strategies for control programs at national, sub-national or regional level

There are three categories of measures for prevention and control of HS (12) : 1- measures to be adopted in endemic countries on a prophylactic basis; 2- measures to be taken in the event of an outbreak; and 3- measures necessary for prevention of spread across regional or national borders. Eradication is also discussed.

Measures to be adopted in endemic countries on a prophylactic basis:

- Vaccinate on a routine prophylactic basis. Vaccination is best done two to three months before the high-risk season (in areas where seasonality occurs) so as to ensure peak immunity during the period of maximum risk.
- Establish a good reporting system. This will enable information on suspected outbreaks to reach animal health authorities as quickly as possible.
- Create awareness of the disease among farmers.
- Prevent mixing of animals from endemic and non-endemic areas. In endemic areas, a significant proportion of animals are latent carriers and are potential sources of infection. In non-endemic areas, animals are not regularly exposed to infection, lack naturally acquired immunity, are not usually vaccinated, and are highly susceptible. An outbreak originating from an activated carrier in such an instance can be explosive. If contact between such animals is unavoidable, it is of utmost importance that susceptible animals from non-endemic areas are vaccinated at least two weeks before contact withanimals from an endemic area.

Measures to be taken in the event of an outbreak:

- Continue vaccination programs. Vaccination is recommended even in the face of an outbreak. In such situations broth bacterins or the alum precipitated (or aluminium hydroxide gel) vaccine is preferred. Broth bacterin and the oil adjuvant vaccine may be administered at different sites, simultaneously.

- Isolate and treat animals showing clinical signs with a parenteral broad-spectrum antibiotic (relatively easy in organized farms or herds that are paddocked at night or for part of the day).
- Check the rectal temperature of all immediate in-contact animals in the herd. This should be done at least once every morning; those animals showing increased temperatures should be treated.
- Search daily for sick animals or carcasses of dead animals (free-roaming, nomadic herds).
- Confine herds as much as possible, and prevent movement of animals in and out of diseased premises or villages.
- Take immediate action to carry out postmortem examinations (local veterinarians) and dispatch samples to the nearest diagnostic laboratory.
- Dispose of carcasses of dead animals properly. Deep burial or effective incineration is recommended. Often, after animals die, carcasses are disowned by farmers. Stray dogs and other scavenging animals can disseminate infection by carrying away portions of infected carcasses, and carcasses dumped into streams and waterways are important sources of infection.
- Properly dispose of unconsumed fodder, bedding etc. from infected premises. Deep burial or drying and burning should be carried out within the premises. Effluent from cattle sheds, dung etc. should be prevented from being washed away from the premises. Drains carrying such material should be led into a deep protected pit within the premises, or subjected to disinfectant treatment.
- Closely monitor or stop rain-associated activities. Activities such as ploughing and preparation of fields for rice cultivation cause considerable movement of draught cattle and buffaloes, and they should be minimized. Might be very difficult for the farmers to stop these activities completely. HS often breaks out during wet seasons.

Measures necessary for prevention of spread across regional or national borders:

(note that this come from recommendations from De Alwis, 1999, and are not OIE guidelines)

- Ensure that the animals originate from a region where no outbreaks of HS have occurred for a minimum of one year. How extensive this disease-free 'region' should be will depend on the system of management practiced in the region.
- Bleed a random sample of animals. This should include the herd or farm of origin and/or other in-contact animals. Test for the presence of antibody by the indirect haemagglutination (IHA) test. High antibody titres is an indication of recent exposure to the disease.
- Hold animals under observation for two to three weeks before transport. During this time repeated attempts should be made to check nasopharyngeal swabs for pasteurellae by mouse inoculation and culture. Blood collected at the beginning and end of such a period should be checked for antibody by the
- IHA test. Animals harbouring type B pasteurellae and/or showing IHA titres should be eliminated. The detection of any carriers or animals with antibody titres would justify an extended period of pre-transport observation for other in-contact animals in the group.

- Quarantine animals after transport to the new location. The animals can be held for a similar period of time to that used before transport. During this time, the same procedures should be carried out.
- Vaccinate animals from disease-free locations in endemic countries. A dose of oil adjuvant vaccine may be given at the end of the quarantine period, followed by a booster three months later. It is equally important to vaccinate all animals in the country of import that are likely to come into contact with animals introduced from endemic countries or regions. Vaccination is the most important control procedure adopted in all countries where the disease is endemic.

Eradication

No country has ever attempted to eradicate HS. This reflects the belief that the existence of carriers makes eradication too hard. This has been strengthened by recent findings indicating that a larger proportion of animals are carriers than was originally thought and that for most of the time the disease remains latent. The existence of carriers among feral ruminants may be a further factor that will make eradication difficult. The only known attempt at eradication was made by the Government of Indonesia on a pilot scale on the island of Lombok with a cattle population of 300,000. The program started in 1978 with intensive vaccination campaigns that were targeted to achieve the highest possible coverage in all susceptible species. The coverage actually achieved over a three-year period was 89% in cattle, 94% in buffaloes, 82% in goats, 93 % in sheep and 80% in pigs. The program was evaluated in 1981, after three annual vaccinations using the oil adjuvant vaccine. During this period, 53 cases of HS were reported. Culture of pharyngeal mucous membrane samples from 220 animals slaughtered in an abattoir yielded positive isolations from five animals (2.2%). Mass vaccination was continued, and a second evaluation was made in 1985. A total of 450 abattoir samples from cattle, buffaloes, goats and pigs were negative. Also, 103 specimens from animals suspected of HS were negative. This island was declared free of the disease in 1985. However, subsequent evaluations based on culture, serology and field reports indicate that HS is still present in Lombok. These observations provide useful indicators of the complexities involved in attempting a total eradication program for HS even on a small island; the implications are much more complex in countries with land borders and a large wildlife population.

Disease situation and government policies by country:

Tables 7 to 12 below have been completed with the information received from the questionnaires sent to the Director Generals and Department of Veterinary Services of the countries of interest.

Tables 7 and 8 show the information received for HS, tables 9 and 10 the information received for Pneumonic pasteurellosis of cattle, and tables 11 and 12 for small ruminant pasteurellosis.

Table 7, 9 and 11 cover the disease situation (if it is notifiable or not), the presence of official surveillance and/or control programs, and the treatment situation. Tables 8, 10 and 12 refer to the vaccination situation.

The definitions that were given to the respondents are:

[1]Surveillance: is the systematic ongoing collection, collation and analysis of data and the timely dissemination of information to those who need to know so that action can be taken.

[2]Control: a program which is approved, and managed or supervised by the Veterinary Authority of a country for the purpose of controlling a vector, pathogen or disease by specific measures applied throughout that country, or within a zone or compartment of that country.

Table 7: Official status, official programs and treatment for Hemorrhagic septicemia in the countries of interest. Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Notifiable (yes/no)	Official surveillance[1] program (yes/no) (if yes, active or passive)	Official control[2] program (yes/no)	Treatment (Chemotherapy)	
				Treatment authorised (yes/no)	Frequently practiced (yes/no)
ASIA					
Bangladesh	Yes	Yes, passive	Yes (targeted)	Yes	Yes
India					
Indonesia					
Myanmar (Burma)	Yes	Yes, passive	Yes	No	Yes
Nepal	No	Yes, passive	No	Yes	Yes
Vietnam	Yes	Yes, passive	No	Yes	Yes
AFRICA					
Burkina Faso					

Côte d'Ivoire (Ivory Coast)	Yes	Yes, passive. But active in case of outbreaks	Yes	Yes	When animals are sick
Ethiopia					
Kenya	No	Yes, passive	No	Yes	No
Madagascar					
Malawi	Yes	No	No	Yes	No
Mali	Yes	Yes, passive	Yes	Yes	Yes
Mozambique					
Rwanda	-	-	-	-	-
Senegal					
South Africa					
Tanzania	Yes	Yes, passive	No	Yes	No
Uganda	No	No	No	N/A	N/A
Zambia	Yes	Yes, passive	Yes	Yes	Yes

Table 8: Vaccination for Hemorrhagic septicemia in the countries of interest.
Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Vaccination			
	Compulsory vaccination (yes/no)	Who pays for the vaccine (Government, farmers, combination, others-specify)	Who delivers the vaccine (official, private vaccinators or both)	Species vaccinated (cattle, sheep, goats, pigs, poultry)
ASIA				

Bangladesh	No	Combination (Government subsidy, owner pays service charge)	Official	Cattle and buffalo
India				
Indonesia				
Myanmar (Burma)	Yes	Government	Official	Cattle
Nepal	No	N/A	N/A	N/A
Vietnam	Yes	Farmers	Both	Cattle, buffaloes, pigs
AFRICA				
Burkina Faso				
Côte d'Ivoire (Ivory Coast)	No	Farmer	-	-
Ethiopia				
Kenya	No	N/A	N/A	N/A
Madagascar				
Malawi	No	N/A	N/A	N/A
Mali	No	Combination	Official	Cattle
Mozambique				
Rwanda	-	-	-	-
Senegal				
South Africa				
Tanzania	No	Not done	Not done	Not done
Uganda	No	N/A	N/A	N/A

Zambia	No	Farmers	Both	Cattle

- Questionnaire received, but left blank by respondent.

Table 9: Official status, official programs and treatment for Pneumonic pasteurellosis in the countries of interest. Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Notifiable (yes/no)	Official surveillance[1] program (yes/no) (if yes, active or passive)	Official control[2] program (yes/no)	Treatment (Chemotherapy)	
				Treatment authorised (yes/no)	Frequently practiced (yes/no)
ASIA					
Bangladesh	No	-	-	-	-
India					
Indonesia					
Myanmar (Burma)	No	No	No	No	Yes
Nepal	No	No	No	No	No
Vietnam	No	No	No	Yes	Yes
AFRICA					
Burkina Faso					
Côte d'Ivoire (Ivory Coast)	Yes	Yes, passive. But active in case of outbreaks	Yes	Yes	When animals are sick
Ethiopia					
Kenya	No	Yes, passive	No	Yes	No
Madagascar					

Malawi	No	No	No	Yes	No
Mali	Yes	Yes, passive	Yes	Yes	Yes
Mozambique					
Rwanda	-	-	-	-	-
Senegal					
South Africa					
Tanzania	Not reported. Confused with CBPP	Yes, active	No	No	No
Uganda	No	No	No	N/A	N/A
Zambia	Yes	Yes, passive	Yes	Yes	Yes

- Questionnaire received, but left blank by respondent.

Table 10: Vaccination for Pneumonic pasteurellosis the countries of interest. Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Vaccination			
	Compulsory vaccination (yes/no)	Who pays for the vaccine (Government, farmers, combination, others-specify)	Who delivers the vaccine (official, private vaccinators or both)	Species vaccinated (cattle, sheep, goats, pigs, poultry)
ASIA				
Bangladesh	-	-	-	-
India				
Indonesia				

Myanmar (Burma)	No	-	-	-
Nepal	No	N/A	N/A	N/A
Vietnam	No	Farmers	Both	Pigs
AFRICA				
Burkina Faso				
Côte d'Ivoire (Ivory Coast)	No	Farmer	-	-
Ethiopia				
Kenya	No	N/A	N/A	N/A
Madagascar				
Malawi	No	N/A	N/A	N/A
Mali	Yes	Combination	Official	Cattle, sheep, goats, pigs
Mozambique				
Rwanda	-	-	-	-
Senegal				
South Africa				
Tanzania	No	Not done	Not done	Not done
Uganda	No	N/A	N/A	N/A
Zambia	No	Famers	Both	Cattle

- Questionnaire received, but left blank by respondent.

Table 11: Official status, official programs and treatment for small ruminants pasteurellosis in the countries of interest. Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Notifiable (yes/no)	Official surveillance[1] program (yes/no) (if yes, active or passive)	Official control[2] program (yes/no)	Treatment (Chemotherapy)	
				Treatment authorised (yes/no)	Frequently practiced (yes/no)
ASIA					
Bangladesh	No	-	-	-	-
India					
Indonesia					
Myanmar (Burma)	No	No	No	No	Yes
Nepal	No	No	No	No	No
Vietnam	No	No	No	Yes	Yes
AFRICA					
Burkina Faso					
Côte d'Ivoire (Ivory Coast)	Yes	Yes, passive. But active in case of outbreaks	-	-	-
Ethiopia					
Kenya	No	Yes, passive	No	Yes	No
Madagascar					
Malawi	No	No	No	Yes	No
Mali	Yes	Yes, passive	Yes	Yes	Yes
Mozambique					

Rwanda	-	-	-	-	-
Senegal					
South Africa					
Tanzania	Not reported. Confused with CCPP	Yes, active	No	No	No
Uganda	No	No	No	N/A	N/A
Zambia	N/A	N/A	N/A	N/A	N/A

- Questionnaire received, but left blank by respondent.

Table 12: Vaccination for small ruminants pasteurellosis the countries of interest. Information provided by the questionnaire sent to the DG/DVS as part of this monograph.

Country	Vaccination			
	Compulsory vaccination (yes/no)	Who pays for the vaccine (Government, farmers, combination, others-specify)	Who delivers the vaccine (official, private vaccinators or both)	Species vaccinated (cattle, sheep, goats, pigs, poultry)
ASIA				
Bangladesh	-	-	-	-
India				
Indonesia				
Myanmar (Burma)	No	-	-	-
Nepal	No	N/A	N/A	N/A

Vietnam	No	Farmers	Both	Sheep, goats
AFRICA				
Burkina Faso				
Côte d'Ivoire (Ivory Coast)				
Ethiopia				
Kenya	No	N/A	N/A	N/A
Madagascar				
Malawi	No	N/A	N/A	N/A
Mali	Yes	Combination	Official	Sheep and goats
Mozambique				
Rwanda	-	-	-	-
Senegal				
South Africa				
Tanzania	No	Not done	Not done	Not done
Uganda	No	N/A	N/A	N/A
Zambia	N/A	N/A	N/A	N/A

- Questionnaire received, but left blank by respondent.

Vaccines Available

Vaccines for the prevention of Hemorrhagic septicemia

Simple toxoid vaccines for *P. multocida* protect relatively well, as those vaccines are based on the capsule of *P. multocida. P. multocida* usually has a prominent capsule, compared to the capsule of *M. haemolytica* which is well less developed.

Bacterins:

The OIE Terrestrial Manual (HS Chapter – updated 2012), says that effective vaccines against HS are formalin-killed bacterins with adjuvants. The adjuvants enhance the level and prolong the duration of immunity. The OIE recommends to use a local isolate of *P. multocida* representing the prevalent serotype. Seed cultures for vaccine production should contain capsulated organisms.

The commonly used HS vaccines are bacterins that are either alum-precipitated vaccines (APV), aluminum hydroxide (AlOH) gel vaccines or oil-adjuvanted vaccines (OAV). To provide sufficient immunity with bacterins, repeated vaccination is required. The use of multiple emulsion (ME) vaccines has been mentioned (the viscosity of OAV may be reduced by re-emulsifying it with an equal volume of Tween 80, producing a free flowing milk-like liquid equivalent to that of a classical water-in-oil emulsion) [13], but no evidence of its use has been found. Very recently, Kumar et al published that a co-adjuvantation of an OAV with alum might be beneficial (so far, evaluated in mice). **http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4662783/pdf/IJM-7-79.pdf.**

Administration of dense bacterins can give rise to shock reactions, which are less frequent with the APV and almost nonexistent with the OAV. The oil-adjuvant vaccine has not been popular because of difficulty in syringing. The alum-precipitated-type bacterin is given at 6-month intervals, while one dose of the OAV provides protection for 9-12 months.

Live vaccines:

A live HS vaccine prepared using an avirulent *P. multocida* strain B:3,4 (Fallow deer strain) has been used for control of the disease in cattle and water buffaloes over 6 months of age in Myanmar since 1989. Safety, efficacy and cross protection data has been published [14]. It is administered by intranasal aerosol application. The vaccine has been recommended by the Food and Agriculture Organization of the United Nations (FAO) as a safe and potent vaccine for use in Asian countries. It has also been evaluated recently in Pakistan, where it showed

that the vaccine could be given safely through aerosol route to buffalo calves 4 to 6 months old. A single dose would protect for only 7 months, but animals given a booster after one month, were protected after 12 months of vaccination [15]. Challenges after 12 months were not conducted in the study. However, there is no report of its use in other countries and killed vaccines are the only preparations in use by the countries affected with HS (OIE, 2012).

Live candidate vaccines have been identified, as for example the live attenuated *aroA* mutant of *P. multocida* B:2 strain developed by Dr. Tabatabei in Iran [16]. Efficacy has been published, but no information has been found on reversion to virulence. A live *gdha* derivative *P. multocida* B:2 has been developed and evaluated with promising results in Malaysia. See Section 8 for more details.

Other HS vaccines:

There are no vaccines for HS based on biotechnology (OIE, 2012).

Comments on cross protection:

The protective immunity for Pasteurella multocida resides in the capsule. The classification based on A B D and E rests on the capsule, and there is very little, if any cross protection between capsule types. In Asia, the only isolated serotype is B:2. In Africa, the predominant serotype is E:2, but some African countries have also reported B:2 (Egypt, Sudan, Cameroon, Namibia and Zimbabwe).

Cross protection between *P. multocida* B and E is limited. For example, a study conducted with the two local Sudanese *P. multocida* strains B and E used for vaccine production, showed that there was a limited cross reaction between the two strains in the rabbit sera when tested by ELISA [17]. Vaccine of strain E protected 50% of the rabbits against challenge with strain B, while the vaccine of strain B could not protect the rabbits against the challenge with strain E (0%). Each vaccine of B and E could protect 100% of the rabbits against the challenge with the same strain.

In an abstract of the Pasteruaceae conference in 2014 presented by Dr Harper, it is mentioned that while it is believed that bacterins provide excellent protection against strains of the same LPS serovar it has never been objectively tested. The group (based at Monash University in Melbourne) suggested the use of a PCR that groups the different strains into 8 LPS types. When they did the LPS structural analysis (strains 3 and 6, the most commonly isolated in poultry), it revealed that many related, but structurally distinct, LPS glycoforms are produced by different isolates from within a single genotype. Moreover, in some L3 field isolates, multiple LPS glycoforms are simultaneously produced by the one strain. The existence of genetically related field isolates producing a wide range of LPS glycoforms, with some strains producing many glycoforms simultaneously, raises significant questions about the ability of existing *P. multocida* bacterins to elicit protection against even closely related strains

http://pasteuraceae-2014.p.asnevents.com.au/days/2014-05-16/abstract/13035.

I contacted Dr Harper, who mentioned one of his PhD students has just finished looking at the sequences of HS strains, and they were all very clonal, and she did not believe there was too much difference between the strains. Unfortunately, they only had available B strains from Pakistan, and didn't have E strains from Africa, but she would think they are very clonal as well. She facilitated the contact with Dr Blackall from Queensland University who is responsible for the Australian Reference centre for Pasteurellacea. He believes that LPS polymorphism is involved in vaccine failure in poultry (associated with killed vaccines), but does not have information on HS. More details on Section 7.

Comments on vaccine potency:

Gowrakkal et al [18] say that in spite of annual vaccination, outbreaks of HS are recorded every year, and that this may be due to improper vaccination and the use of a low potent vaccine. With the focus on potency, they compared the seed culture of HS bacteria used to produce the vaccine for its antibody induction efficiency, before and after passaging it in the natural host (calf). They used the strain P-52, of the Indian Veterinary Research Institute, Izatnagar, Uttar Pradesh. The results revealed that back passaged vaccine seed, provided better immune efficiency than the original stock culture, and therefore recommend the back passaging of the seed vaccine at least once in 6 to 8 months in its natural host system for the induction of better immunity.

The OIE does mention in the Terrestrial Manual "A calf is infected with the culture, and, within 2-3 hours of its death, blood is collected aseptically from the heart and stored at -20°C in 1 ml aliquots. A fresh aliquot is used for each new batch of vaccine. It is permissible to subculture this aliquot once or twice, provided the colony size does not diminish". I have discussed this procedure with Dr Nwankpa from AU-PANVAC who believes most African laboratories produce their master seed in calves and if the seed gets too old (over 2 years), then they passage it again in calves.

For more details on the different HS vaccines, please see Table 12.

Vaccines for the prevention of Pneumonic pasteurellosis in cattle (caused by M. haemolytica and P. multocida)

The initial *M. haemolytica* vaccines, being simple toxoids had 2 limiting factors. First, there is no cross-protection between serotypes, and the more serotypes were included, the poorer the protection against all strains. Five strains were the maximum number that could be included. The second limiting factor was the endotoxin level, which induced shock reactions, and paradoxically, an exacerbation of the pneumonia.

Virulence factors were investigated, and leukotoxins (Lkt) were identified. *M. haemolytica* produces an exotoxin (Lkt) during the logarithmic phase growth, that binds to the intact signal peptide of CD18 on bovine leukocytes, and lyses them. The Lkt from cattle is relatively homogenous. It is Important to note that the leukotoxin does not protect against *P. multocida* infections.

Other vaccines have been developed based on iron regulated proteins, and outer membrane proteins (OMP) which are associated with iron regulation. The iron-regulated proteins are related to the serotype. A better

protection is shown if the iron-regulated outer proteins are derived from a number of different strains, and there is some cross-protection between unrelated strains. More strains can also be included in the vaccine than in a bacterin vaccine.

Modern vaccines use culture supernatants containing leukotoxin (Lkt) and other soluble antigens, or bacterial extracts, alone or combined with bacterins. Some people says Lkt based vaccines, protect cattle well against nearly all *M. haemolytica* infections. Some other say that these modern vaccines have 50–70% efficacy in prevention of *M. haemolytica* pneumonia [2]. There are no OIE standards for *M. haemolytica* vaccines.

Vaccines for the prevention of pasteurellosis in small ruminants (caused by M. haemolytica and B. trehalosi)

Sheep are affected by all 17 of the original *P. haemolytica* serotypes (now known as *M. haemolytica*, and some as *B. trehalosi*) and because their strains show much more variability between different strains of any single serotype, they are not protected as well as cattle are, by leukotoxin type vaccines. Serotyping does not indicate whether a particular strain isolated from sheep would protect that sheep if a Lkt vaccine would be used. The leukotoxin vaccines are based on the cattle *M. haemolytica* A1 strain of the Lkt, which may be the same or may differ from the Lkt of the strain which is infecting the sheep. This explains the variability in protective effect when sheep are vaccinated with Lkt vaccines (commercial vaccines for sheep are still usually based on the serotype, and not on Lkt).

This can be exemplified by a contact I had from an African vaccine manufacturer requesting help for a case of pasteurellosis in goats, in which the vaccine was not effective, and due to their limited diagnostic capability, they were not able to serotype.

A large multinational pharmaceutical company produces a vaccine for sheep based on iron-regulated proteins which contains 9 serotypes and is available in South Africa. This vaccine provides a broader protection. The amount of cross protection cannot be determined precisely, as there is a complex combination or virulence factors in sheep strains.

Main vaccine needs

There is a need for a vaccine that has/is:

- Longer duration of immunity.
- Easier to administer
- More cross-protective (especially in some African countries, where both strains have been identified)

Table 13: Comparison of the different Hemorrhagic septicemia vaccines

	Alum-precipitated vaccines (APV)	Aluminum hydroxide gel	Oil adjuvant vaccines (OAV)	B:3, 4
Status	Extensively used in different parts of the world	Thailand and Laos, but also other countries in Asia and Africa.	Extensively commercialized in Asia. Also sold in Africa.	Used in Myanmar
Type	Killed	Killed	Killed	Live
Origen	Bacterin to which potash alum has been added to have a 1% final concentration	Aerated cultures blended with AlOH gel at a final concentration of 3%		Strain isolated from a fallow deer in England (*P. multocida* strain B: 3,4)
Target species	Cattle, sheep and goats	Cattle and buffaloes	Cattle and buffaloes	Cattle and water buffaloes
Indications	Vaccinate animals 6 months of age and above.	6 months of age or above, followed by booster dose after 1 month. Other manuf say from 2 weeks of age.	Vaccinate at 3-4 months of age with 2 doses at 3 months interval	For use in animals over 6 months of age
Immunity	6 months	4 - 6 months.	1 year but has been reported 18 months in buffaloes, and 26 months in cattle.	1 year
Route	SC	SC	IM	IN (fine aerosol)
Dose & volume	Approx 0.75 µg dry weight per ml, so that a 3 ml dose has minimum 2 µg bacteria. (5 - 10 ml for cattle & buffaloes depending on the size of the animal)	2 ml for all age groups of cattle and buffaloes. 1 ml for all age groups sheep and goats.	If a 3-mL dose is to be used, the minimum requirement of 2 µg dry bacteria should be present in 1.5 mL . Some manufacturers use a 2 ml dose.	2×10^7 viable organisms
Serology on standard tests				
Withdrawal period		7 days		
Efficacy				Cross protection serotypes E:2, F:3,4 and A;3,4.
Zoonotic characteristics	No	No	No	No
Use in pregnant animals		Just as precaution, it is recommended to try in some animals first		
Other side effects	Shock reactions can occur	Small lump may develop at injection site, and animals may show temporary lameness.	Difficult to inject due to high viscosity. Abscess at the site of injection and vaccination reactions often reported	
First used	1950s			1984
Large scale use	Extensively used in Southern India and Central Africa. Most popular is some Asian countries.			In Myanmar has been used since 1989
Others	Important to mix thoroughly because alum causes the cells to settle at the bottom	Shake well before use.	Superior in terms of storage at room temperature for 1 year without deterioration	FAO has recommended it for use in other Asian countries, however, there are no reports of it being used.

Characteristics of Ideal Vaccine Candidates for Smallholders

The Target Product Profiles (TPPs) reflect the availability and utility of current agents and incorporate features that will be necessary to improve on the current products and to address unmet needs, taking into account the particular requirements of the poorest livestock keepers.

The TPPs are more robust when they include the opinions and consider the needs of the different stakeholders. While efforts have been made to encompass them, the TPP showed in Table 12 below, should be considered a proposal, a live document subject to improvements.

Information on current vaccines has been obtained from the datasheet of different products as per links below:

APV: **http://ivpm.webs.com/products.htm**

AlOH: **http://www.brilliantbiopharma.com/hs-vaccine-haemorrhagic-septicaemia-vaccine/19**

AlOH: **http://www.obpvaccines.co.za/Cms_Data/Contents/OBPDB/Folders/Product/~contents/5HRJ2K3SLYQ7WA9V/1196%20Pasteurella_Cattle_PI.pdf**

OAV: **http://iahvb.kerala.gov.in/index.php/products/vaccines/livestock-vaccines/haemorrhagic-septicaemia-vaccine**

OAV: **http://vsvri-eg.com/Products/ProductsAnimal%202-1%20Haemorrhagic%20Septicaemia%20Oil.%20Adj.%20Vaccine%20(%20for%20cattle%20).html**

Table 14: Target Product Profile (TPP) Hemorrhagic septicemia vaccine – Proposal:

	Attribute	Minimum (current available vaccine)	Ideal
1	Antigen	Immunogen with protective antigens for *P. multocida* B (Asia) or E (Africa)	Immunogen with protective antigens for *P. multocida* B (Asia) **and** B &E (Africa)
2	Indication for use	For active immunization of cattle, buffaloes, sheep and goats.	For active immunization of cattle, buffalo, sheep and goats of all ages and sexes.
3	Recommended species	Cattle, buffalo, sheep and goats	Cattle, buffalo, sheep and goats.
4	Recommended dose	APV: 5 – 10 ml (depending animal size) AlOH: 5 ml OAV: 2 – 3 ml Live: 2 x 10^7 viable organisms/dose	Same dose for all species (2 ml)
5	Pharmaceutical form	Alum-precipitated AlOH gel Oil adjuvanted Fine aerosol	Ready to use solution/suspension
6	Route of administration	APV: SC AlOH: SC OAV: IM Live: IN	SC, IM or fine aerosol
7	Regimen - primary vaccination	APV: single vaccination AlOH: Booster 1 month. AOV: Booster 3 months Live: single vaccination	Single lifetime dose
8	Regimen - booster	APV: AlOH: 6 – 12 months (one month before expected occurrence)	Lifelong immunity after primary vaccination

		AOV: Annual (one month before monsoon)	
9	Epidemiological relevance	Protection against Hemorrhagic septicemia	Protection against Hemorrhagic septicemia
10	Recommended age at first vaccination	APV: 6 months AlOH: 6 months. Some manufacturers say from 2 weeks. OAV: 3- 4 months Live: Over 6 months of age	From 1-2 months of age, when other vaccines are applied.
11	Onset of immunity	APV: AlOH: 2 weeks OAV: 21 days Live:	One week following primary vaccination
12	Duration of immunity	APV: 6 months AlOH: 4 – 6 months OAV: 1 year Live strains: 1 year	Lifelong immunity
13	Expected efficacy	To prevent disease & prevent mortality.	To prevent infection and transmission in 100% of the animals.
14	Expected safety	APV: AlOH: Occasionally, transient swelling at site of injection. OAV: Small swelling at injection site. Live: reactions when used SC or IM. Safe when used IN.	No post-vaccinal reactions at any age.
15	Withdrawal period	APV: AlOH: 7 days OAV: Live strains:	Nil

16	Special requirements for animals	Do not vaccinate un-healthy animals or during an outbreak.	Vaccinate all animals
17	Special requirements for persons		None
18	Package size	APV: 50 doses AlOH: 20 – 50 doses OAV: 150 doses Live strains:	Multiple pack size from 5 doses
19	Price to end user		
20	Storage condition and shelf-life as packaged for sale	APV: 21 days at room temperature, 6 months at 4°C. AlOH: Stable at 2-8°C for 2 years OAV: Stable at 4-8°C for 1- 2 years Live strains:	Stable at 30°C for 24 months
21	In-use stability	APV: AlOH: OAV: Live strains:	24 hours or greater
22	Other:		

Limitations

Scientific quality: The publications and data from the different research groups, should be carefully evaluated. The use of good science and good experimental design with use of proper controls, adequate numbers, suitable challenge model, reproduction of results by them and by independent groups, and appropriate analysis has not been verified for this monograph. If any of these projects were to be pursued, a detailed peer review taking into account the above considerations is strongly recommended.

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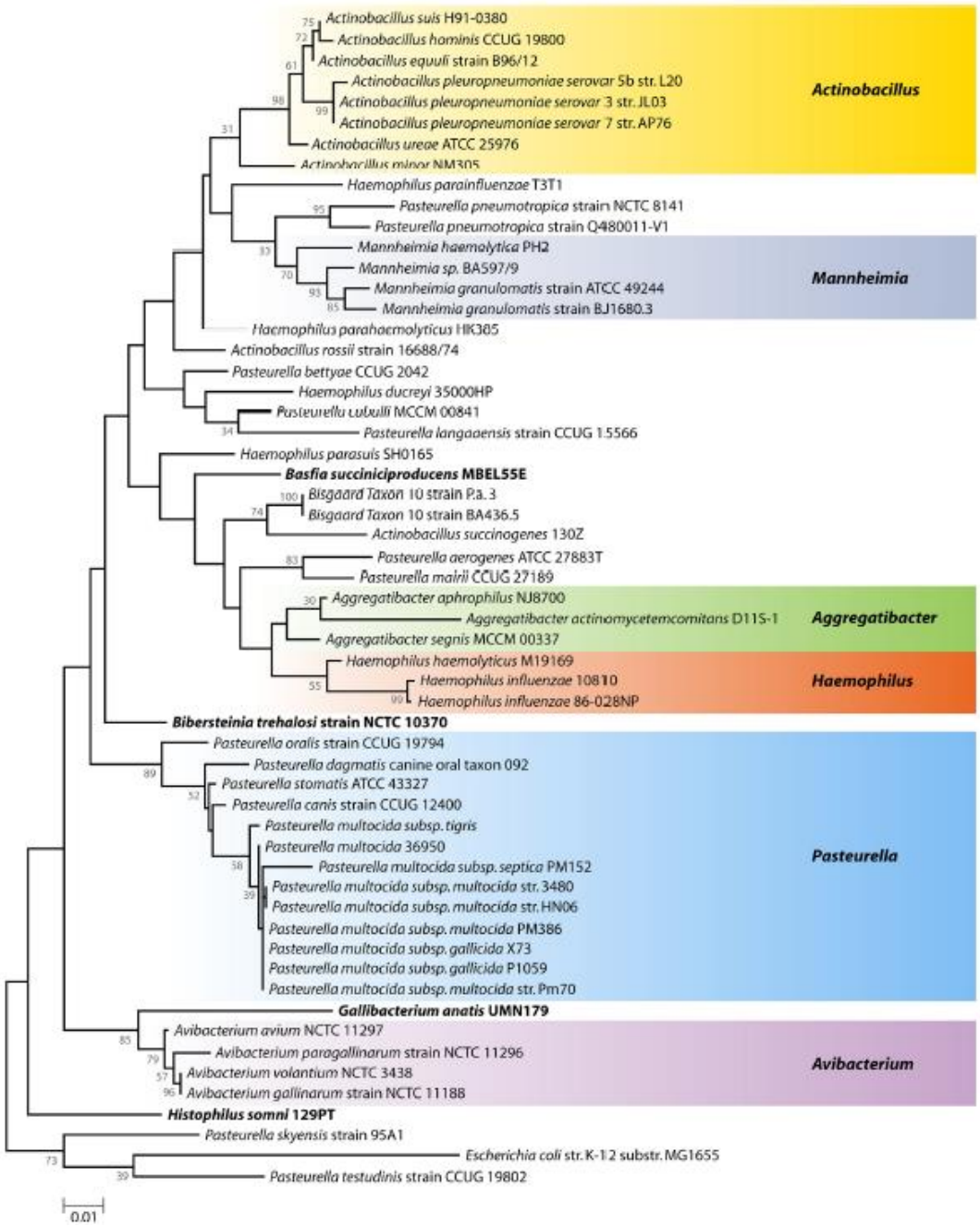
ANNEX 1: Phylogenetic relationships of *Pasteurella multocida* and related *Pasteurellaceae* bacteria based on 16S rRNA genes.

Source: Wilson and Hoi, 2013 [1].



ANNEX 2: Revised names for genus Pasteurellaceae

New Name	Old Name	Old Serotype
Mannheimia haemolytica	*Pasteurella haemolytica*	1, 2, 5, 6, 7, 8, 9, 12, 13, 14, 16
Mannheimia glucosida	*Pasteurella haemolytica*	11
Mannheimia rumenalis	*Pasteurella haemolytica*	Untypable
Mannheimia granulomatis	*Pasteurella granulomatis*	Untypable
Mannheimia varigena	*Pasteurella haemolytica*	6, 14
Mannheimia taxon 7	*Pasteurella haemolytica*	Untypable
Mannheimia taxon 8 A, B & C	*Pasteurella haemolytica*	Untypable
Mannheimia taxon 9	*Pasteurella haemolytica*	Untypable
Mannheimia taxon 10	*Pasteurella haemolytica*	Untypable
Mannheimia taxon 12	*Pasteurella haemolytica*	Untypable
Biberstenia trehalosi	*Pasteurella haemolytica*	3, 4, 10 & 15

Pasteurella multocida did not change the name.

ANNEX 3: Additional data on disease presence and incidence

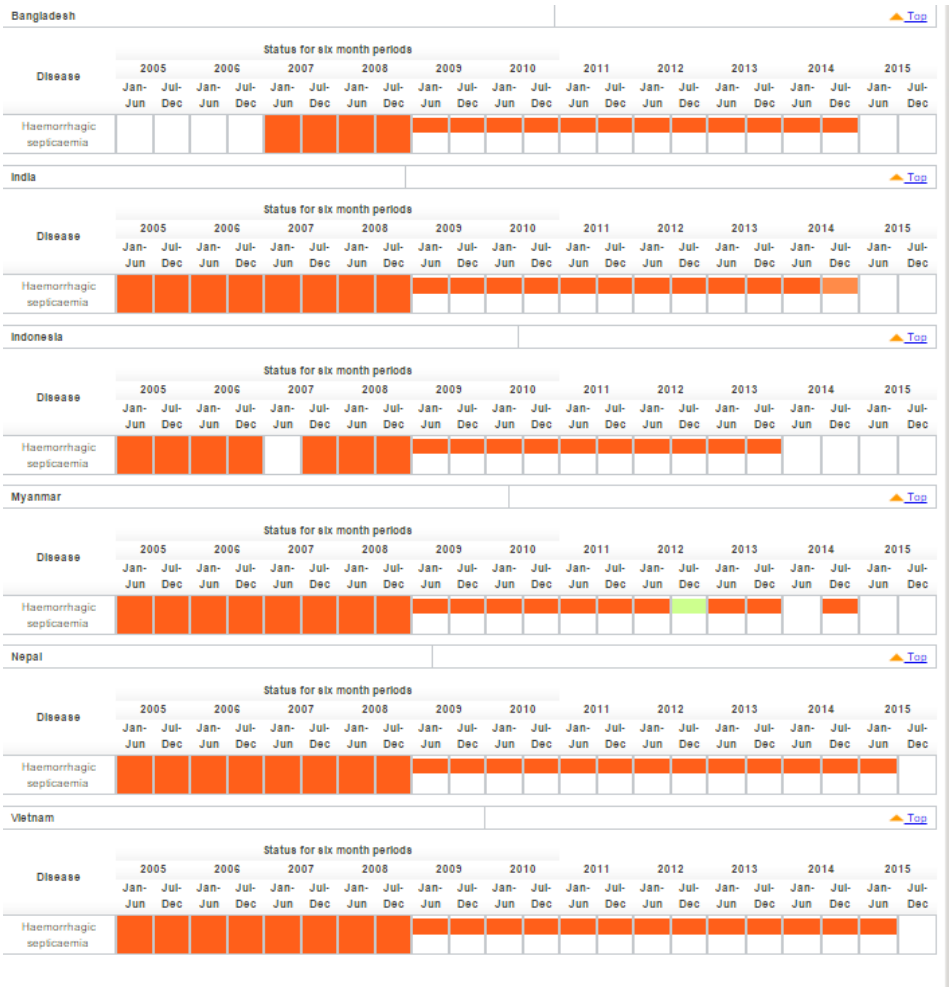
Reports to OIE on Hemorrhagic septicemia:

Key to colours

- There is no information available on this disease
- Never reported
- Disease absent
- Disease suspected but not confirmed
- Infection/infestation
- Disease present
- Disease limited to one or more zones
- Infection/infestation limited to one or more zones
- Disease suspected but not confirmed and limited to one or more zones

When different animal health statuses between domestic and wild animal population are provided, the box is split in two: the upper part for domestic animals, and the lower part for wild animals.

Hemorrhagic septicemia in Asia: Bangladesh, India, Indonesia, Myanmar, Nepal and Vietnam



Hemorrhagic septicemia in Western Africa: Burkina Faso, Ivory Coast, Mali and Senegal

Hemorrhagic septicemia in Eastern Africa: Ethiopia, Kenya, Rwanda, Tanzania and Uganda

Hemorrhagic septicemia in Southern Africa: Madagascar, Malawi, Mozambique, South Africa and Zambia