INTRODCTION

Foot-and-mouth disease (FMD) is a highly contagious and economically important disease caused by foot-and-mouth disease virus (FMDV) (Klein *et al*., 2008), FMD is a transboundary animal disease (FAO, 2007; Depa *et al*., 2012; Abunna *et al*., 2013), also known as aphthous fever (Mekonen *et al*., 2011; Gebregziabher *et al*., 2013; Abunna *et al*., 2013). According to the office international des epizooties (OIE), FMD ranks first among the notifiable infectious diseases of animals (Mekonen *et al*., 2011; Duguma *et al*., 2013).

Seven serotypes of the virus have been identified serologically. They were designated "O", "A", "C", "Asia1" and the "SAT1, 2 and 3". All of them were reported in Africa with the exception of Asia1 and a part from type C. Prevalent serotypes and topotypes in the continent are known for their immunological diversity (African Union Inter-African Bureau for Animal Resources, 2010), Infection with any one serotype does not confer immunity against any of the other (OIE, 2012). The 3 SAT serotypes predominate in southern Africa. They are maintained by African buffalo (*Syncerus caffer*) that can be a source of infection for susceptible livestock in close proximity. Infection in buffalo is sub-clinical and normally occurs in calves as soon as maternal antibodies wanes at 2-6 months of age (Vosloo, report; Royal Gazette).

The disease is characterized by high fever, loss of appetite, salivation and vesicular eruptions on the feet, mouth and teats (Chepkwony *et al*., 2012). Clinical signs can vary from mild to severe, FMD cannot be differentiated clinically from other vesicular diseases, such as Swine Vesicular disease, Vesicular stomatitis and Vesicular exanthema (OIE, 2012).

The disease has a high morbidity and low mortality with low occurrence in adult animals. However, myocarditis may occur in young animals resulting in death. Animals that recover remain in poor physical condition over long period of time (Jenbere *et al*., 2011). In fact, many calves die without any clinical signs of the

disease. Mortality rate is around 5% in mature animals but can run as high as 50% in young animals because of myocardial damage (Tunca *et al*., 2008).

The spread of the disease is mainly through direct and indirect contact, which former involving mechanical transfer of droplets from infected animals to other susceptible animals, while the latter route is through contaminated personnel, vehicles and fomite. Airborne transmission over long distances has been implicated under certain climatic and meteorological conditions, particularly in respect to domestic pigs that exhale the highest quantities of airborne virus, this is easily passed onto in-contact ruminants that are highly susceptible to infection by the respiratory route. FMD is endemic in most of sub-Saharan Africa, except in a few countries in Southern Africa, where the disease is controlled by the separation of infected wildlife from susceptible livestock as well as by vaccination. In most parts of Africa, FMD outbreaks are often underreported either because of its endemicity as well as the fact that it is not associated with high mortalities in adult susceptible animals. As such it is not perceived as an important livestock disease among herds men (Lazarus *et al*., 2012). In the Sudan, FMD is still endemic in the country. It occurs mostly in the cold, dry season. The extensive livestock husbandry systems adopted in the Sudan seems to favor conditions for the spread of FMD virus. Cattle reared under nomadic conditions in the Sudan wander around for grazing, which may extend for many kilometers and use their tongues while grazing, but when these functions are affected by FMD lesions in the feet and mouth, they become recumbent and mostly suffer from starvation (Habiela *et al.*, 2010).

Significant economic losses are produced by its high morbidity and the export trade restrictions imposed on affected countries (FAO, 2007). Losses were largely due to the death of newborn and suckling calves, loss of weight and milk production and a decrease in draught power and infertility (Habiela *et al*., 2010). FMD affects 27 million livestock units each year which is approximately 0.64% of the total livestock units globally. The overall economic impact was calculated

based on the costs of a vaccine and its application being US$1 and that for any livestock unit affected by FMD it would cause a loss in production equivalent to US$100. The latter estimate takes into consideration costs of diagnostics and surveillance required to prevent and control FMD. Taking into account the death of an animal, loss in weight gain, milk production and draught power, the previous loss is felt to be a conservative estimation. The total global FMD annual impacts is calculated to be US$5 billion (RVC, FAO and OIE).

Objectives:

The objectives of the present study were:

1. To estimate the seropsitivity of Foot and Mouth Disease (sero-type A) of cattle in Khartoum state, Sudan
2. To investigate the possible risk factors associated with Foot and Mouth Disease infection.

CHAPTER ONE

1. Literature Review

1.1 Definition:

Foot and mouth disease (FMD) is highly contagious and affects over 70 domestic and wild Artiodactyla species (Mohamoud *et al*., 2011), characterized by the formation of vesicles in the mouth, at coronary band and skin of interdigital cleft (Mekonen *et al*., 2011).

1.2 History of FMD:

FMD is recognized as a significant epidemic disease threatening the cattle industry since the sixteenth century and till date it is a major global animal health problem. The history of FMD may be traced to the era of Hieronymus Fracastorius, a monk who described a disease outbreak in 1546 A.D. that occurred in cattle near Verona, Italy. Almost 400 years later, in 1897, Friedrich Loeffler and Paul Frosch demonstrated that a filterable agent is responsible for FMD. This was the first demonstration that a disease of animal was caused by a filterable agent and ushered in the era of virology (Longjam *et al*., 2011).

In the Sudan, The first record of the disease was in 1903 (Abuelzein, 1983 and Habiela *et al*, 2010). Virus serotyping information has been available consistently since 1952 (The WRL at Pirbright, U.K., has been receiving FMD virus-suspected samples from the Sudan regularly since 1952. Only virus serotypes A, O, SAT 1, were isolated, until 1977 when SAT 2 was first recorded in the country) (Abuelzein, 1983).

1.3 FMDV:

1.3.1 Causative Agent:

Foot-and-mouth disease virus (FMDV), was the first recognized viral pathogen and is the sole member of the genus *Aphthovirus* belonging to the *Picornaviridae* family. Seven immunologically different serotypes of the FMD virus are known, namely, A, O, C, Asia-1, South-African Territories (SAT) -1, -2 and -3, which comprise more than 65 subtypes. The viral particle, or virion,

contains a single-stranded RNA of positive polarity, approximately 8500 nucleotides long. It is an icosahedral particle with a smooth surface and a diameter of about 30nm. (Longjam *et al*., 2011). Replication and assembly take place in the cytoplasm and the virus is released via cell lysis (Murphy *et al*, 1999). The virus is antigenically variable, and there is no cross-protection among the seven known serotypes and only limited protection between the numerous subtypes that have evolved. The RNA contained in FMDV particles encodes the viral proteins in addition to serving as a template for the synthesis of a complementary full-length negative RNA strand that in turn serves as a template for the synthesis of new viral genomes. Notwithstanding the simplicity of the mechanisms of genome replication and expression used by picorna viruses. A small number of cellular genes- mostly involved in host immunity-are known to be activated by FMDV infection. (Piccone *et al*., 2009).

1.3.2 Virus Morphology:

FMDV is a single stranded (ss) positive sense RNA virus with the whole virus particles having sedimentation coefficient of 146S and genome of~8.5 Kb size. The genome is polyadenylated at 3' end and carries a small covalently linked protein, VPg at 5' end. The 5' untranslated region (UTR) contains a short fragment called S-fragment, a poly (C) tract followed by large (L) fragment of over 700 bases. Functionally, the genome can be categorized into three main regions: (a) 5' noncoding regulatory region, (b) polyprotein coding region (subdivided into L, P1, P2, and P3), and (c) 3' non coding regulatory region. The translation initiation starts at two AUG codons separated by 84 nucleotides following the Internal Ribosome Entry Site (IRES). The viral genome is translated as a single polyprotein, which is posttranslationally cleaved by viral proteases into four structural proteins (VP1, VP2, VP3, and VP4) and several nonstructural proteins (L, 2A, 2B, 2C, 3A, 3B, 3C, and 3D). The P1 region of genome encodes the 4 structural proteins VP1, VP2, VP3, and VP4 encoded by 1D, 1B, 1C and 1A genomic regions, respectively. Sixty copies of each

structural protein (VPl-4) assemble to form the capsid. Among which VP4 is internal whereas others are exposed on virion surface. The 3 surface exposed capsid proteins carry the neutralizing antigenic sites. Among the 4 structural polypeptides, VP1 is the most immunogenic protein of FMDV having its G-H loop protruded from the surface and is maximally exposed on the capsid surface forming large part (54%) of virus surface. (Longjam *et al.*, 2011). Only VP1 (not VP2 and VP3) produces antibodies which can bind to virus and neutralise, and since VP1 alone can confer protection in animals (Crowther, 1986).

1.3.3 Phylogenetic Analysis of FMDV:

Phylogenetic analysis of the virus protein (VP) 1 region of FMD viruses has been used extensively to investigate the molecular epidemiology of the disease worldwide. These techniques have helped define genetic relationships between FMDV isolates and geographic distribution of lineages and genotypes; they have also helped establish genetically and geographically linked topotypes and trace the source of outbreaks. Topotypes are defined as geographically clustered viruses that form a single genetic lineage generally sharing >85% (O, A, C, and Asia 1) or >80% (SAT 1, SAT 2, and SAT 3) nucleotide identity in the VP1-coding region (Ayelet *et al.*, 2009).

1.3.4 Physico-Chemical Characteristics:

Picorna viruses are small RNA viruses that are enclosed with a non-enveloped protein shell (capsid). The capsid consists of poly peptides which are devoid of lipo-protein and hence is stable to lipid solvents like ether and chloroform. Foot-and-mouth disease viruses can be inactivated by a number of chemical substances at the acidic and alkaline pH ranges, however, the virus is stable between pH 7 and 9 and at 4°C and -20°C. Two percent solution of NaOH or KOH and 4% Na_2C0_3 are effective disinfectants for FMDV contaminated objects, but the virus is resistant to alcohol and phenolic and quaternary ammonium disinfectants. However, the FMD virus is also sensitive to a range of other chemicals like trypsin which causes cleavage and denaturation of the vital

capsid protein, VP1. The size of droplet aerosol also plays a role in the survival or drying out of the virus, where a droplet aerosol size of 0.5 - 0.7 IJm is optimal for longer survival of the virus in the air while smaller aerosols dry out. In dry conditions the virus also survives longer in proteins e.g. in epithelial fragments (Sahle, 2004)

1.4 Virus Classification:

FMDV is belongs to the genus Aphthovirus, one of the genera of the family Picornaviridae. The name Picornaviridae is derived from the Latin word 'pico' (small) and 'rna' (RNA) which refers to the size and genome type while the genus name 'aphtho virus' refers to the vesicular lesions produced in cloven hoofed animals (Sahle, 2004).

1.5 Strain Classification:

1.5.1 Antigenicity:

Among the capsid proteins, VP1 is the most antigenic protein and carries the domain mainly responsible for antigenic heterogeneity and cell-virus interaction. The conserved Arginine-Glycine-Aspartic acid (RGD) site within the G-H loop spanning amino acid positions 140 -160 of the VP1 protein protrudes from the virion surface and is mobile and constitutes the host cell binding motif in FMD viruses. This G-H loop often experiences a higher rate of non-synonymous substitution and greater genetic variability in the 1D gene. These independent antigenic sites were identified on the VP2 and VP3 genes was found in serotypes A, O and Asia1. Changes to the genes encoding capsid proteins can result in antigenic variation and evolvement of new subtypes. This may give rise to immunologically distinct variants that can re-infect individuals that have been previously infected by related viruses. The degree of cross protection among different subtypes of the same serotype thus varies. (Sahle, 2004 pp8-9). Type A22 does not confer immunity to type A5 virus (Crowther, 1986).

1.5.2 Genome and protein:

The genome consists of a positive sense single stranded RNA (ss RNA). The RNA genome is approximately 8500 bases long with a poly A tail at its 3' end and a viral genome protein (VPg) at its 5' end. Four polyproteins (L, P1, P2 and P3) are translated and processed into the different structural and non-structural proteins by viral encoded proteases as showen in (Figure 1.1) (Kasanga *et al*., 2012). The L protein represents the leader protein, where 2 initiation sites (AUG codons) have been identified in FMD virus, namely Lab and Lb The P1 gene product is the precursor of the capsid proteins 1D, 1B, 1C and 1A. Firstly, the intermediate P1 precursor is processed with the help of viral protease 3C pro to produce VP0, VP1 and VP3 where the products combine to form empty capsid particles. The mature virion is produced after the encapsidation of the virion RNA which is accompanied by the cleavage of VP0 to VP2 and VP4. VP1-3 are exposed on the capsid surface. The P2 (2A, 2B, 2C) and P3 (3A, 3B, 3C, 3D) regions encode for non-structural proteins that are involved in viral RNA replication and protein processing (Sahle, 2004).

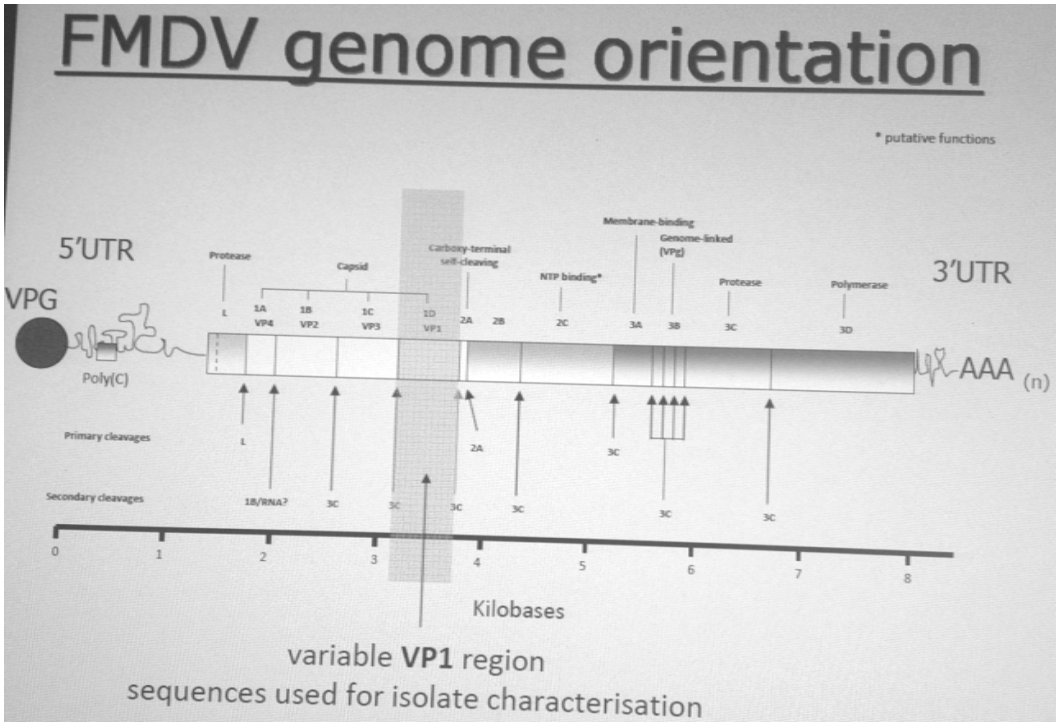


Figure 1.1: FMD genome orintation (Kasanga *et al*., 2012)

1.5.3 Genomic Variation:

1.5.3.1 Mutations:

Foot-and-mouth disease virus undergoes a high rate of mutation during replication. This is mainly due to a lack of replication error checking mechanisms. the generation of new variants is considered as one of the major problems in the control of FMD by vaccination (Sahle, 2004).

1.5.3.2 Recombination:

It has been shown that genetic recombination occurs between viruses of the same serotype as well as between serotypes. Mutations through recombination could result in the exchange of genetic material that could lead to the generation of new antigenic variants that may escape immune pressure (Sahle, 2004)

1.6 Epidemiology:

Epidemiology of FMD is complex, and it is affected by different viral, host, and environmental factors, among them, variations in virus virulence, particle stability in different microenvironments, and chances of long-term persistence. FMDV multiplication and spread also depend on the host species, nutritional and immunological status, population density, animal movements, and contacts between different domestic and wild host species and animals capable of mechanical dissemination of the virus. The environment can provide geographical barriers to virus dissemination or, alternatively, can promote virus transmission when appropriate atmospheric conditions prevail (Longjam *et al*., 2011).

1.6.1 Distribution:

There are seven recognised serotypes of FMD (O, A, C, Asia 1, SAT 1, SAT 2 and SAT 3), which differ in distribution across the world (FAO, 2007). Serotypes A and O have the widest distribution, occurring in Africa, Asia and South America. Types SAT 1, 2 and 3 are currently restricted to Africa only and Asia 1 to Asia. (Mekonen *et al*., 2011; FAO, 2007). the capacity to invade free areas is common to all types and periodically SATs are introduced into the near

East, and Asia-1 into western and eastern parts of Eurasia (FAO, 2007). In most of sub-Saharan Africa, serotypes O, A, SAT 1 and SAT 2 are predominant (Habiela *et al.*, 2010). North and Central America, New Zealand, Australia, Greenland, Iceland and western Europe are free of FMDV. Western Europe was affected by some recent outbreaks (eradication was successful), but FMD has not been reported in North America for more than 60 years. The last U.S. outbreak occurred in 1929, while Canada and Mexico have been FMD-free since 1952-1953 (Iowa State University, 2014).

1.6.2 Historical distribution:

Serotype O and A reported in France by Valee and Caree, in 1926, Waldmann and Trautwein reported serotype C. Serotypes SAT1, SAT2, and SAT3 of FMDV was observed in sample collected from the FMD outbreak in South Africa. The seventh serotype, Asia 1, was reported from Pakistan (Longjam *et al*., 2011). In the Sudan; FMD is highly endemic, Four of the 7 virus serotypes (O, A, SAT 1 and SAT 2) have been isolated but only from cattle in the country.The WRL at Pirbright, U.K., has been receiving FMD virus-suspected samples from the Sudan regularly since 1952. Only virus serotypes A, O, SAT 1, were isolated, until 1977 when SAT 2 was first recorded in the country (Abuelzein, 1983).

1.6.3 Morbidity and Mortality from FMD:

Morbidity from FMD varies with the animal's species, breed and pre-existing immunity, as well as the dose of virus and other factors. The morbidity rate can approach 100% in naive cattle or swine herds, but some FMD viruses can disappear from a sheep flock after infecting a relatively low percentage of the animals. The pattern of disease is influenced by the epidemiological situation. When more than one virus circulates in a region, there may be periodic outbreaks, due to the lack of protection between serotypes and the limited cross-protection between some strains. When there is only a single serotype in a region, the virus may cause only mild clinical signs, with cases seen mainly in

young animals as they lose their protection from maternal antibodies. Adult livestock do not usually die from FMD (the case fatality rate is approximately 1-5% for most strains), but deaths can occur in young animals. In lambs, reported mortality rates range from 5% to 94%. Mortality has also been reported to reach 80% in some groups of calves, and 100% in suckling piglets (with lower rates in older piglets). The percentage of FMDV infected animals that become carriers, with or without vaccination, is still uncertain. Estimates vary widely, with experimental and field studies reporting carrier rates ranging from less than 5% to more than 50% under different conditions (Iowa State University, 2014).

1.6.4 Prevalence and risk factors of FMD in other countries:

A cross sectional study was conducted from November 2009 to April 2010 on appaerently healthy cattle in South Wollo, Dessie Zuria and Kombolcha area, Ehiopia to determine the seroprevalence of Foot and Mouth Disease in cattle, using 3ABC-ELISA test. The overall seroprevalence of FMD infection was found to be 5.59% . The study has indicated that FMD is prevalent in the study area affecting all age groups of cattle (Gebregziabher *et al.*, 2013). In study carried out from November 2010 to March 2011 at Dire Dawa and its surroundings, Ethiopia to estimate the seroprevalence of Foot and Mouth Disease in cattle using non structural protein 3ABC ELISA kit. The overall prevalence of Foot and Mouth Disease was 8.01 % (79/986). There was a statistically significant difference observed in the prevalence of FMD with the origin ($p= 0.004$) and the age of the animals ($p= 0.006$). However, there was no statistically significant difference observed in the case of sex of the study animals ($p>0.05$) (Abunna *et al.*, 2013). A total of 499 serum samples were collected (January 2007 to December 2008) and tested to determine the seroprevalence of Foot and Mouth Disease in cattle in Somali Ecosystem (SES), Kenya. The samples were screened against the five serotypes of FMD known to be in circulation in Kenya (O. A, C, SAT1, SAT2) and measured by microneutralization assay. The overall sero-prevalence of FMD in the Somali-

ecosystem was found to be 45.3% (95% CI = 40.96 to 49.66%). Twenty seven percent of all animals sampled tested positive for only one serotype while 17.6% tested positive for multiple serotypes. There was a high prevalence ($p \leq 0.05$) in the circulation of serotype O (23 and 95% CI = 20.13- 7.57%) as compared with the other serotypes, while the prevalence of serotype C was significantly lower ($p \leq 0.05$) compared to the other four serotypes (1.6 and 95% CI = 0.82-3.12). Wajir district recorded the highest prevalence (24.8 and 95% CI = 16.71 to 27.54) while Garissa district recorded the least (6.2%). There was no significant sero-prevalence variation in relation to sex while older animals had higher sero-prevalences. The pastoral mode of livestock production, porous borders and wildlife inter-phase are significant factors (Chepkwony *et al.*, 2012). An epidemiological study was conducted in Rajshahi, Bngladesh between July2010 and February 2011 with the objective of determining the perevalence of foot -and- mouth disease (FMD) in cattle and identifying the potensial risk factors associated with the disease. In total, 347 skin diseased cattle were examiend in the veterinary Clinic of the University of Rajshahi. The overall prevalence of FMD rajshahi was found to be 25.07% (n=87) and the potential risk factor for FMD in this study area were assessed by questionnaire, there was a statistically significant difference ($p<0.01$) in prevalence associated with age of cattle. The prevalence of FMD was significantly ($p<0.01$) higher in male (36.53%) than female (16.06%). A significant ($p<0.01$) variation in breed susceptibility was observed affecting mostly indigenous cattle(41.46%) compared to cross breed (16.07%) (Sarker *et al.*, 2011). A serological survey was conducted between 2009 and 2011 in some states in Nigeria to determine the seroprevalence of foot and mouth disease in cattle and demonstrate the evidence of antibodies in sheep, goats and pigs. (448) sera of cattle, sheep and goats were screened for FMD antibodies using the Enzyme-linked immunosorbent assay (ELISA). overall prevalence of FMD was 64.73%; CI95%: 60.20 to 69.02%. Specific seroprevalence in sheep and goats were 41.66 and 21.81%, respectively. The

result confirmed that FMD is still an important cattle disease (Lazarus *et al*., 2012). A cross-sectional seroprevalence study of cattle foot and mouth disease (FMD) was conducted in Somalia Regional State, western Ethiopia. to determine the individual seroprevalence of FMD in cattle. A total of 384-serum samples were collected from cattle herds. A 3ABC bovine ELISA kit was used. The overall individual animal antibody seroprevalence was 14.05% (95% CI= 11.2 to 18.13%). Statistically no significant variation (P>0.05) was observed between districts. There was no significant variation (P>0.05) in seroprevalence among male (19.4%) and female (13.6%) animals, age groups of animals showed a significant variation ($\chi 2$= 8.45, DF= 2, P= 0.01) (Mohamoud *et al*., 2011). A cross sectional study was conducted from October 2007 to April 2008 to determine sero-prevalence and associated risk factors for seropositivity of FMD. Using 3ABC ELISA. The overall seroprevalence was 5.6% (48.4% at herd level) for FMD was found differences in giographical locations, age groups and herd sizes were risk factors found statistically ($p<0.05$) associated with occurrence of FMD (Jenbere *et al*., 2011). A sero-epidemological study was conducted in Southwestern Ethiopia between November 2007 and February 2008 to determining the sero-prevalence of Foot and Mouth Disease (FMD) in cattle and identifying the potential risk factors associated with the disease. a total of 273 sera samples were collected from cattle in 98 herds. The sera samples were screened using the FMD-3ABC-ELISA kit. The overall prevalence of FMD was 12.08% (33/273). There was statistically significant difference between districts. From the various risk factors analyzed peasant associations, cross boundary movement and herd size were seen to be statistically associated ($p<0.05$) with the seroprevalence of FMD. There was no significant variation in seroprevalence among sex, age and herd type. (Gelaye *et al*., 2009).

1.6.5 Prevalence of the Disease in Sudan:

In Sudan, a survey was conducted between 2006 and 2008. A total of 1,069 sera were randomly collected from cattle (469), sheep (319), goats (88) and dromedary camel (193) from seven states in the Sudan; for the detection of antibodies to FMDV. Application of liquid phase blocking (LPB) ELISA revealed that antibodies to four serotypes were present in ruminants; namely O, A, SAT 1 and SAT 2. No antibodies to FMDV were detected in camel sera. The results differed from early reports regarding the prevalence of serotype specific antibodies in different species; for instance, in cattle, the antibodies to type A (78.13%) surpassed that of type O (69.39%) and the antibodies to type SAT 2 (44%) surpassed that of type SAT 1 (20.2%). This work elucidates the current epidemiology of FMD in some parts of the Sudan (Habiela *et al.*, 2010).

1.6.6 Host Range of FMD:

Cattle, sheep, goats, and pigs are the main domesticated species infected. The Water Buffalo (*Bubalus bubalis*) can become infected and may also transmit infection to other species. Camelids, experimentally infected, contract the disease but there is no evidence of transmission to other domestic livestock and there seems to be some doubt as to whether they play any role in the epidemiology of the disease in domestic livestock.

A wide range of wild cloven-footed animals contract FMD including deer and pigs. The African Buffalo (*Syncercus caffer*) appears to be particularly susceptible to infection and may act as a reservoir host (Davies, 2002). Although FMD is known as a disease of cloven footed animals it can occur naturally in other animals, infection has been established experimentally in a number of other species. However, it is doubtful whether these animals play any part in the epidemiology of the disease. FMD is not considered zoonotic. Although clinical cases have been proven in human, these are extremely rare in relation to human exposure during outbreaks (Depa *et al.*, 2012; Davies, 2002).

1.6.7 Susceptibility:

Cattle with FMD, especially the highly productive breeds found in developed countries, often have severe clinical signs. In water buffalo, the clinical signs are reported to be milder than in cattle, and lesions may heal more rapidly. Young pigs up to 14 weeks of age may die suddenly from heart failure; piglets less than 8 weeks of age are particularly susceptible. FMD tends to be mild in sheep and goats Dromedary camels do not seem to be susceptible to FMD severe outbreaks have been documented in wild populations of some species such as mountain gazelles (*Gazella gazelle*), impala and saiga antelope (*Saiga tatarica*), and high mortality or severe clinical signs have been reported in some captive wildlife species. Young animals can die suddenly of myocarditis. (Iowa State University, 2014). Susceptability was observed affecting mostly indigionus cattle (Sarker *et al*., 2011). It is not associated with high mortalities in adult susceptible animals (Lazarus *et al*., 2012).

1.6.8 Pathogenicity of FMD:

Replication of the infectious particles is extremely rapid after entry through the upper respiratory tract or lung, with viraemia seeding infection into the epithelium where secondary virus multiplication results in vesicles and shedding from the udder in milk. The incubation period, from infection to clinical signs, may be as short as 2/3 days or as long as 14 days and infected animals may become infectious before showing clinical signs. The virus is excreted during viraemia for some days; thereafter as serum antibody develops viraemia decreases, and the animal ceases to be infectious as the lesions heal. The disease is characterised by vesicular lesions on the coronary band of the hooves and in the mucosa of the mouth including the tongue and palate. The vesicles typically contain clear or straw-coloured fluid before they burst and heal. There is a rise in body temperature of some 3–4˚C.

In sheep The lesions are often difficult to find and may be confused with other conditions. The disease varies considerably in its severity. It may result in death

or severe morbidity particularly in neonates but in areas where the infection is endemic the disease may be mild and the few vesicles that appear may heal without further damage (Davies, 2002).

1.6.9 Immunity:

1.6.9.1 Passive Immunity:

It has been found that the antibody titers in the case of passive immunity in calves from cows that have recovered from foot-and-mouth disease or have been vaccinated against it rise immediately following the intake of colostrum. They have been found to reach a peak level, displaying maximal values for a long period of time. Later on the antibody titers decline gradually. Specific antibodies have also been demonstrated at the age of more than two months following birth. Higher values of passive immunity have been established in calves from survivals and from cows that have been vaccinated twice and this is explained by the presence of a higher content of foot-and-mouth disease antibodies in the dams (Mitev *et al*., 1975).

1.6.9.2 Active Immunity:

Antibodies to FMDV structural proteins could be detected in both sheep and cattle at day 6 post vaccination by ELISA (Australian Veterinary Journal). A high level of immunity can be induced by potent vaccines within a few days in both cattle and pigs but the interval between vaccination and protection may be some 14 days with the usual commercial vaccines. The current generation of FMD vaccines protect animals for periods up to 12 months but the immunity conferred is not absolute and FMD wild virus may multiply to a greater or lesser extent in a vaccinated animal. Vaccine programme failures may be attributable to challenge as in large intensive units or to inadequate population cover that leaves sufficient unvaccinated and therefore susceptible animals for the virus to maintain itself and continue circulating. Vaccines can be employed prophylactically to protect a population against future challenge, or in emergency, to deal with a current epidemic. Prophylactic vaccination on a

national scale is usually confined to cattle and such programmes successfully eliminated FMD from Continental post-war Europe. It is widely assumed that for vaccine to be used on a national scale, 80% cover (Davies, 2002).

1.6.10 Risk factors for FMD infection:

The most important factors that could be associated with FMD are; age, sex, breed, farming system, seasonal influence, previous disease and preventive measures during examination. (Sarker *et al*., 2011). FMD occurs usually during the dry season when feed is not available (Duguma *et al*., 2013). FMDV multiplication and spread also depends on the host species, nutritional and immunological status, population density, animal movements, and contacts between different domestic and wild host species and animals capable of mechanical dissemination of the virus. The environment can provide geographical barriers to virus dissemination or, alternatively, can promote virus transmission when appropriate atmospheric conditions prevail. In this multifactorial scenario (Longjam *et al*., 2011). Wildlife have been shown to play a role as a maintenance host for FMDV,when fences and vaccination zones around the national parks are absent. Thus, uncontrolled animal movements are still a major risk for spreading FMD. Transboundary mobility of FMDV has been proven between East African countries (Namatovu *et al*., 2013).

1.6.11 Transmission:

The disease spreads rapidly by movement of infected animals or mechanically on fomites such as clothing, shoes, vehicles and veterinary instruments. The reasons for the rapidity of spread to fully susceptible population is due to the highly infectious nature of the virus, the production of high titer in respiratory secretions, and the large volumes of droplets and aerosols of virus shed by infected animals, the stability of virus in such droplets, the rapid replication cycle with very high virus yeilds and the short incubation period (Gebregziabher *et al*., 2013).

1.6.11.1 Direct contact:

In tropicl areas the most important method of spread is by direct contact between animals moving freely across states and national boundaries as trade or nomadic cattle. Disease outbreaks occur mostly with the onset of the hot humid season. The climatic stress suppresses the existing immunity in cattle population which at first leads to sporadic, and subsequently to severe and wide spread disease outbreak (Abunna *et al*., 2013).

1.6.11.2 Air borne Transmission:

Airborne trans-mission over long distances has been implicated under certain climatic and meteorological conditions, particularly in respect to domestic pigs that exhale the highest quantities of airborne virus (Lazarus *et al*., 2012). The disease is notoriously contagious that it can spread as much as 50(fifty) miles downwind from one outbreak area to another (Mekonen *et al*., 2011). Wind borne aerosol virus produced by infected animals are carried over 250kms. Survival of virus in aerosols depends on relative humidity. Cattle are mainly infected by inhalation, often from pigs, which excrete large amount of virus by respiratory aerosols and are considered highly important in disease spread. Large amounts of virus are excreted by infected animals before clinical signs are evident and wind may spread the virus over long distances (Depa *et al*., 2012).

1.6.11.3 Indirect contacts Transmission:

The indirect most common transmission pathway is the consumption of contaminated hay (USAID, 2007-2008), through contaminated personnel, vehicles and fomites (Lazarus *et al*., 2012).

1.6.12 Incubation period:

The incubation period of FMD virus infection is 2 to 14 days (Beyi, 2012). however it can be as short as 24 hours in pigs challenged with a high dose. The incubation period can be highly variable depending on host, agent and environmental factors including husbandry management factors (Senawi, 2012). It is reported to be one to 12 days in sheep, 4 days in wild boar, 2 days in feral

pigs, 2-3 days in elk, 2-14 days in Bactrian camels and possibly up to 21 days in water buffalo infected by direct contact (Iowa state University, 2014)

1.6.13 Survival on fomites:

Foot and Mouth Disease virus is considered to be a moderately stable virus. At a pH between 7.0 and 8.5 most strains are stable, especially at lower temperatures. Unlike other picorna viruses, the FMDV capsid dissociates at a pH of 6.5 or below. It is stable at a humidity above 55 to 60% but is sensitive to heat and desiccation. The survival of FMDV is also influenced by the nature of the materials as a high concentration of organic material helps the survival of the virus. The virus can be recovered from the blood, pharynx, vagina and rectum up to 97 hours prior to the onset of vesicular lesions. It can also persist in mammary tissue for 3 to 7 weeks after infection. The virus can survive outside the host, and potential sources of virus include excretions and secretions of infected livestock such as saliva, semen, milk, faeces, urine, and vaginal secretions. The virus can also survive in skim milk, cream and the pelleted cellular debris components of milk obtained from FMD infected cows after the milk had been pasteurised at 72°C for 0.25 minutes and in cream components after heat treatment at 93°C for 0.25 minutes. The FMDV can also survive for long periods in meat and animal products including frozen bone marrow, lymph nodes and offal The average period of survival of FMDV on wool at 4°C is approximately 2 months with the period of survival decreasing considerably as the temperature increases to 18°C. The maximum estimated survival period of FMDV outside the host is approximately three months in regions with daily temperatures greater than 20°C (Senawi, 2012).

1.6.14 Carrier state:

A carrier animal is defined as one from which the virus can be recovered 28 days or more after infection (Depa *et al.*, 2012; Davies, 2002). Also it is an inapparent infection or have been recorded in cattle, African buffalo, sheep , and goats but not in pigs. It occurs with all serotypes and has been identified in both

experimentally and naturally infected animals. It is well established that a proportion of animals infected by FMD virus eventually become ‘‘carriers’’ with equal facility irrespective of whether they have been vaccinated, passively immunised or are immunologically naive cattle.The carrier period appears to vary between species, being in excess of 12 months in cattle, up to 9 months in sheep and goats and at least 5 years in African Buffalo (Davies, 2002).

1.7 The Disease:

1.7.1 Clinical Signs of FMD:

The disease is characterized by high fever, loss of appetite, salivation (Gelaye *et al*, 2009) Infected animals exhibit blisters and ulcers in the mouth, feet and udder, lose weight and stop producing milk. Although the disease is rarely fatal in adult animals, high mortality can result in the young. On recovery ruminants can become ‘carriers’ with persistent sub-clinical infection (Australian Governmen, 2007).

1.7.2 Post-Mortem Findings and Histopathology:

The primary lesions in the young animals are observed in the heart at necropsy. The myocarditis of young animals is acute, with hyaline degeneration and necrosis of muscle fibers and an intense infiltration of mainly lymphocytes (Tunca *et al*., 2008). Degenerative muscle fiber cells were swollen, and the cross striations disappeared. Cardiac myocyte nuclei within the foci of myocarditis were perchromatic, pyknotic, or karyorrhectic. With development of necrosis, the nuclei disappeared and muscle fibers disintegrated into irregular masses of eosinophilic amorphous that invaded the damaged fibers contained large eosinophilic granules of necrotic debris within their cytoplasm. Fine mineral granules were scattered indiscriminately among the necrotic contents of some fibers. A focal acute vasculitis affected a few small vessels (Tunca *et al*., 2008).

The characteristic lesions of foot and mouth disease are single or multiple, fluid-filled vesicles or bullae; however, these lesions are transient and may not be observed. The earliest lesions can appear as small pale areas or vesicles, while

ruptured vesicles become red, eroded areas or ulcers. Erosions may be covered with a gray fibrinous coating, and a demarcation line of newly developing epithelium may be noted. Loss of vesicular fluid through the epidermis can lead to the development of “dry” lesions, which appear necrotic rather than vesicular. Among domesticated animals, dry lesions are particularly common in the oral cavity of pigs. The location and prominence of FMD lesions can differ with the species In young animals, cardiac degeneration and necrosis can result in irregular gray or yellow lesions, including streaking, in the myocardium; these lesions are sometimes called “tiger heart” lesions. Piglets can have histological evidence of myocarditis without gross lesions in the heart. Signs of septicemia, abomasitis and enteritis, as well as myocarditis, have been reported in lambs. Only nonspecific gross lesions were described in infected fetuses from experimentally infected sheep. They included petechial hemorrhages in the skin, subcutaneous edema, ascites with blood-tinged peritoneal fluids and epicardial petechiae. Vesicles were not found, and the placenta did not appear to be affected. Some infected fetuses had no gross lesions. In another study, infected fetuses were generally autolyzed (Iowa State University, 2014).

1.7.3 Sample Collection:

The samples for laboratory diagnosis are epithelium or vesicular fluid collected from FMD suspected animals. The samples of choice in cattle are lesions from tongue tissue, buccal mucosa, wounds from feet and hoofs. In pig, fluid filled vesicles wounds from tongue, snout, coronary band and hoof shall be collected. At least one gram of epithelial tissue shall be collected from each animal. Additional samples shall be collected if the weight of each sample is not enough. The samples are kept in screw cap bottle, contained 50% glycerine buffer, capped tightly and sealed with adhesive tape to prevent leakage of the buffer. The bottles are clearly labeled and wrapped with many layers of paper before putting in suitable container or can and tightly sealed. For transportation of the samples to laboratory, the samples are kept cool in strong, unbreakable ice

container. In case of sending samples via post, the samples shall be wrapped with many layers of paper, to prevent from breaking and leaking, before putting into unbreakable container or box together with detailed history of the samples. For serum samples, serum is collected by using sterile glass syringe or dried and cleaned container. After blood clotting, the cleared serum is transferred to plastic vial. If serum containing red blood cell, centrifuge the serum to separate red blood cell and then collect clear serum in plastic vial and seal tightly to prevent leaking. Then the serum shall be kept cool in ice cube container or -20 °C refrigerator while transport to laboratory (Thai AgriculturalSTandard, 2004).

1.7.4 Diagnosis:

The accurate diagnosis of infection with FMDV is of prime most importance for both control and eradication campaigns in FMD endemic areas and as a supportive measure to the stamping out policy in FMD-free areas (Longjam *et al*., 2011). Numerous diagnostic techniques have been developed for FMD with the aim of developing a rapid, sensitive, specific and reliable method to effectively diagnose the disease. The diagnosis of FMD can be divided into 3 categories based on the detection of clinical signs, virus, or antibody to the virus (Senawi, 2012).

1.7.4.1 Clinical signs:

The disease is often initially diagnosed based on clinical signs therefore requires vigilance by the farming community and veterinary profession and the infrastructure to allow early reporting of disease (Senawi, 2012).

1.7.4.2 Virological:

viral diagnosis of FMD is carried out on epithelial tissue or vesicular fluid from clinical samples using specific laboratory diagnostic techniques.

1.7.4.2.1 Viral isolation:

This test requires cell culture to detect the presence of live virus in suitable samples. There are many types of cell lines used including calf thyroid cells, 2 - 7 day old suckling mice cells, IBR – S2 cells, Lamb kidney cells, Pig cells and

baby hamster kidney (BHK 21), (Senawi, 2012). The cell cultures should be examined for cytopathic effect (CPE) for 48 hours If no CPE is detected the cells should be frozen and thawed, used to inoculate fresh cultures and examined for CPE for another 48 hours. Unweaned mice are an alternative to cell cultures and should be 2–7 days old and of selected inbred strains. Some field viruses may require several passages before they become adapted to ice. In the case of OP fluids, pretreatment with an equal volume of chloro-fluoro-carbons may improve the rate of virus detection by releasing virus from immune complexes. (OIE, 2012).

1.7.4.2.2 Immuonological methods:

1.7.4.2.2.1 Complement fixation test (CFT):

In 1929, Ciuca was first to use CFT for typing antiserum and FMDV of guinea pig origin. Although CFT was a fast method it needed high virus load and results were sometimes affected by pro-and anticomplementary activities of the test sample (Longjam *et al.*, 2011). CFT an alternative test for international trade (OIE, 2012). It has disadvantages which are its relatively low sensitivity. The sensitivity and specificity of the test is also dependent upon the animal species tested and is not sufficiently sensitive to detect infection (Senawi, 2012).

1.7.4.2.2.2 Nucleic acid recognition:

This method is dependent upon detection of viral RNA by a PCR. It is a diagnostic method of choice and several tests have been developed for FMDV. The PCRs are very sensitive as they require only a small quantity of sample. The fundamental requirements for PCRs are selection of the primers and an efficient extraction method. The primers selected should specifically target all FMDV types and subtypes without reacting with other unrelated viruses of the same family. RT-PCR can be used to amplify genome fragments of FMDV in diagnostic materials including epithelium, milk, serum and OP samples (Senawi, 2012). However, with the introduction of molecular techniques in the field of diagnosis, several techniques based on viral genome detection such as

hybridization using DNA probes and the advent of Polymerase Chain Reaction (PCR) technique in the recent past have led to development of several reverse transcription PCR (RT-PCR) procedures for specific detection of FMDV RNA. Because of the reported sensitivity and specificity, RT-PCR has been evaluated as a diagnostic tool for FMDV detection in parallel with ELISA and virus isolation. Another form of PCR, multiplex PCR (mPCR), has also been evaluated for differentiating FMDV serotypes as well as for differential diagnosis with other vesicular diseases such as Vesicular' Stomatitis, Swine Vesicular Disease. The most recent development in the field of diagnosis by nucleic acid detection is the use of thermal cyclers capable of measuring fluorogenic PCR amplification in real-time have become available, making precise quantitation of nucleic acids possible over a wide concentration range. The fluorogenic RTPCR provides relatively fast result, enables a quantitative assessment to be made of virus amount, and can handle more samples and/or replicates of samples in a single assay than the conventional RT-PCR procedure.Therefore it is seen as a valuable tool to complement the routine diagnosis procedure for FMD virus diagnosis (Longjam *et al*., 2011).

1.7.4.3 Serological Diagnosis:

Serological tests for FMD are of two types; those that detect antibodies to viral structural proteins (SP) and those that detect antibodies to viral nonstructural proteins (NSPs) (OIE, 2012).

1.7.4.3.1 Testing antibody to structural proteins:

The structural proteins (SP) tests are serotype-specific and detect antibodies by vaccination and infection.They are highly sensitive, the virus or antigen used in the test is closely matched to the strain circulating in the field. (OIE, 2012).

1.7.4.3.1.1 Virus neutralization test (VNT):

It is a prescribed test for international trade (OIE, 2012). VNT detect antibodies to FMDV structural proteins and require separate testing for each of the seven serotypes of FMDV These tests are time consuming to perform, require virus

containment facilities and cannot differentiate vaccinated from convalescing animals (Bronsvoort *et al*., 2006).

1.7.4.3.1.2 Enzyme linked immunosorbent assy (ELISA):

ELISA is the preferred procedure for the detection of FMD viral antigen and identification of viral serotype, it is preferable to the complement fixation test because it is more sensitive and it is not affected by pro- or anti-complementary factors (OIE, 2012). The ELISAs are also considered to be more reliable than the VNT and are useful for evaluating the immunological response of animals following infection and vaccination. However they have some disadvantages including low specificity and the lack of stability of inactivated antigens (Senawi, 2012). The ELISAs are blocking- or competition-based assays that use serotype-specific polyclonal antibodies (PAbs) or monoclonal (MAbs), are quicker to perform and are not dependent on tissue culture systems and the use of live viruses (OIE, 2012).

1.7.4.3.2 Testing antibody to non-structural proteins:

The detection of antibody to the NSPs of FMDV can be used to identify past or present infection with any of the seven serotypes of the virus, whether or not the animal has also been vaccinated (OIE, 2012).

1.7.4.3.2.1 Agar gel Immunodiffusion test:

useful tool for epidemiological surveys of livestock, however the interpretation of results in animals that have been repeatedly vaccinated can be confusing (Senawi, 2012).

1.7.4.3.2.2 Non-structural protein (NSP) ELISA:

The ELISA-based diagnostic techniques for NSP antibody detection provide many advantages, such as objectivity compared with gel diffusion tests for NSP, high sensitivity and specificity, and the capability for large-scale screening. The NSP ELISA tests have been recommended by the OIE to be used for serological surveillance in regions or countries that practice vaccination against FMD and for monitoring virus circulation in the field (Senawi, 2012).

1.7.4.4 Differential Diagnosis:

Erosive lesions in the buccal cavity are characteristic of other diseases such as mucosal disease and rinderpest, whilst foot lesions in sheep may be caused by foot rot. However salivation, lameness in all four feet, and a sharp rise in temperature are characteristics of FMD and should always be a cause for suspicion (Davies, 2002).

1.8 Treatment:

There is no specific treatment for FMD, other than supportive care. Treatment is likely to be allowed only in countries or regions where FMD is endemic (Iowa State University, 2014).

1.9 Control of FMD:

The control strategies vary from country to country based on their epidemiologic conditions (Beyi, 2012). Vaccination build up immunity for the animals. FMDV vaccines must be closely matched with the serotype or strain of FMDV in the field outbreaks. Control by movement restriction and quarantine animals for at least 21 days is another method that can limit spreading FMDV to neighboring area. Ring vaccination at 5 – 10 km radius shall be done (Thai Agriculural Standard, 2004). FMD endemic countries do not follow stamping out policy and use only vaccination as a measure of control. For effective control of FMD about 60-80% of animals need to be covered under vaccination so as to control the outbreak of diseases (Depa *et al*., 2012).

1.9.1 Control by Vaccination:

In the absence of the capacity to control FMD through animal movement restrictions and other biosecurity measures, vaccination remains the only practical control strategy (Namatovu *et al*., 2013). The first FMD vaccine was produced in 1938 using tongue epithelium harvested from cattle deliberately infected with FMD virus. Oil adjuvant single and double emulsions are used to produce vaccine for immunisation of all species of animals including pigs. Oil adjuvant vaccine should have potency of at least 3 PD_{50} and provide protective

immunity within 7 days in cattle, swine and sheep. Revaccination must be carried out every 6 months. After multiple doses of vaccines in older animals vaccination frequency could be decreased to once a year provided that no new strains are not covered by the vaccine formulation emerge or are introduced. (Depa *et al*., 2012).

The (O.I.E.) trade regulations at the time required 12 months of disease freedom following the end of emergency vaccination before a country could be declared free. However, in 2004 the regulations were altered and countries can now regain FMD free status six months after the end of vaccination providing they have carried out surveillance with an NSP test this is three months later than could be achieved following a slaughter policy without vaccination. (Bronsvoort *et al*., 2006), Many countries use routine vaccination against local FMD virus stains (Haratian *et al*., 2000). Vaccination can be used to help restrict the spread of the infection, reduce the susceptibility of animals to infection with FMD virus, protect from clinical illness and reduce shedding of virus and onward transmission. However, the protective effect takes time to develop and may be overwhelmed by a high level of challenge or a poor antigenic match between the vaccine strain and the challenge virus. Furthermore, animals that are clinically protected may still become infected and in the case of ruminants, go on to become subclinical carriers (Arnold *et al*., 2007).

1.9.2 Control by Movement Restriction:

During the phase of an outbreak, infected farms are quarantined and animal movement is restricted. Also, ring vaccination is done around the infected area to break the spread of the outbreak. In peace time, massive vaccination, restriction of animal movement to disease-free areas and surveillance are carried out (Beyi, 2012).

1.9.3 Control by Stamping out:

Stamping out (slaughter of all infected and in contact animals) (Depa *et al*., 2012). Control of the disease encompasses an exclusion and slaughter policy,

particularly for the FMD-free countries (Aggarwal *et al*., 2002) The United Kingdom, Ireland, countries of Scandinavia, Japan, Canada and the United States of America were able to control the disease by stamping-out. In Europe, FMD has been successfully controlled for several decades by extensive vaccination of the cattle population. Most of the European countries have agreed to a policy of non-vaccination and In the case of an outbreak, infected as well as in contact animals will be slaughtered (Sahle, 2004).

1.9.4 Progressive Control Pathway for FMD (PCPFMD):

FAO/OIE assist endemic countries to reduce progressively the impact of FMD through the Progressive Control Pathway for FMD (PCPFMD) tool It consists of six stages (0–5). The main activities of the PCP–FMD tool include; monitoring circulating serotypes, vaccination and enhancing bio-security. In Eastern Africa, quarantine and vaccination are among the existing FMD control strategies , however, the effectiveness of quarantine is limited by inadequate facilities and very weak law enforcement against animal movements. Restriction of animal movements is complicated by social customs (communal grazing, dowry and pastoralism) and both legal and illegal cross-border animal movements. In addition, although, wildlife have been shown to play a role as a maintenance host for FMDV, fences and vaccination zones around the national parks are absent. Thus, uncontrolled animal movements are still a major risk for spreading FMD mobility of FMDV has been proven between East African countries Hence, there is a need for an integrated regional approach to FMD control (Namatovu *et al*., 2013).

The main constraints in controlling this disease and why it is considered as the most dreaded viral disease are its high contagiousness, wide geographical distribution, broad host range, ability to establish carrier status, antigenic diversity leading to poor cross-immunity, and relatively short duration of immunity(Longjam *et al*., 2011).

CHAPTER TWO

2. Materials and Methods

2.1 Study area:

The study was conducted in Khartoum State. The State lies between longitude 31˚-34˚ east and latitude 15˚-16˚ north in an area of about 28.165 square kilometres. It is bordered to the north and the east side by the River Nile State, to North Western by the Northern State, and to the east and south-eastern and south by Kassala, Gedaref and Gezira state respectively and to west by North Kordofan (Figure 2.1). Most of Khartoum state lies in the climatic semi-desert region, while northern areas lie in desert zones. The climate of the state is ranging from hot to very hot. The weather is rainy in summers, cold and dry in winters. Average rainfall reaches 100-200 mm in the north-eastern areas and 200-300 mm in the North Western areas. Temperature ranges in summer between 25-40˚C degrees in the months from April to June, and 20-35˚C in the months from July to October. In winter, however, temperatures continue to decline between November to March from 25-15 degrees.

Geographically, Khartoum state is divided into three blocks (Figure 2. 2).

A. First block: it starts from the Mugran, i.e. the confluence of the two rivers (the blue and white Niles). Being confined between them, this block extends southwards to the boundaries of the Gezira state. Administratively, it is divided into two localities, Khartoum and Gabal Owlia localities.
B. Second block: is limited between the Blue Nile and the River Nile. It includes the localities of Bahari and Sherg Elneel which.
C. Third block: namely, the one located west of the White Nile and the River Nile, includes three localities; Omdurman, Um Badda and Karari.

Figure 2.1: The study area Khartoum state, Sudan (MARFR, 2014)

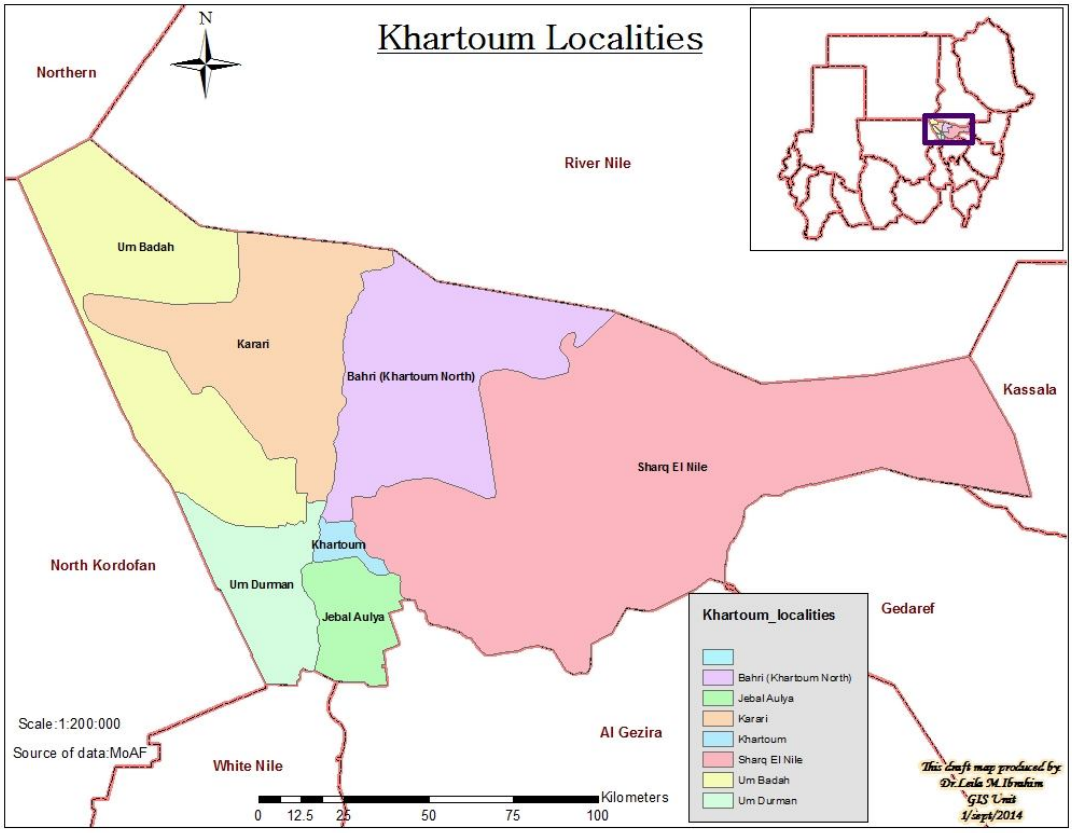


Figure 2. 2: Localities in Khartoum state (MARFR, 2014)

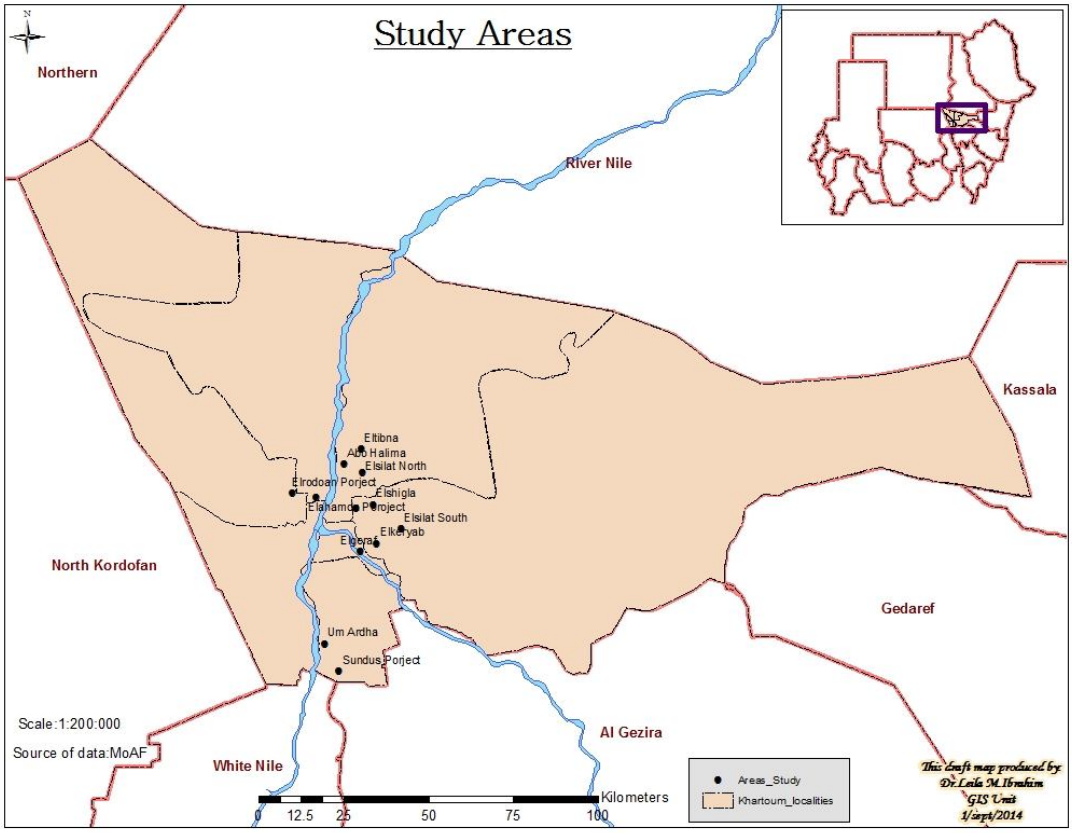


Figure 2. 3: Selected localities and location (MARFR, 2014)

2.2 Sample size:

The actual sample size for determining the prevalence rate of FMD in cattle in Khartoum state was calculated based on the following parameters: 95% level of confidence, ±5% desired level of precision and the expected prevalence rate of FMD in cattle. The prevalence was calculated with the help of the following formula as described by (Thrusfield, 2005-2007):

$$n = \frac{1.96^2 P_{exp} (1- P_{exp})}{d^2}$$

where: n = required sample size

P_{exp} = expected prevalence

d = desired absolute precision

From a previous study carried out in Ethiopia (November 2009 to April 2010) of FMD, the prevalence estimation was found to be 5.59% (Gebregziabher *et al*., 2013). Then the sample size calculated as fallow:

$$n = \frac{1.96^2 \times 0.0559 \times (1- 0.0559)}{0.05^2} = 81.0964679616$$

So the Sample size was 82 serum samples from cattle in Khrtoum state but 85 sample was collected as per available kits.

2.3 Samples:

Blood samples were collected - from 9/4/2014 to 22/5/2014 - from jugular vein of individual animal - healthy cattle - using 10 ml capacity of non-heparinized vacutainer tube, each tube was labeled and kept at room temperature overnight. serum was harvested into another sterile eppendorph tube and stored at -20°C until the day of dispatch. Finally, sera were transported to the Veterinary Research Institute (VRI), FMD Department at Soba Khartoum by using an icebox containing icepacks and then stored again at -20°C until the virus neutralization test was conducted (Duguma *et al*., 2013). Samples were collected from different breeds and different ages. From a total of 85 sera collected as shown in (Table 2.1) only 77 samples were tested.

Table 2.1: Blood samples taken from farms in different localities of Khartoum State

No.	Locality	Location	breed		Sex		Production purpose		Grazing system		Total of sera
			No. of cross	No. of local	No. of female	No. of male	Milk	Milk & Meat	Resident	Transhuman	
1	Khartoum	Elgeraf	2	0	2	0	Milk		Resident		2
2	Jabal Elawliaa	Sundus Project	5	0	5	0		Milk & Meat	Resident		5
3	Jabal Elawliaa	Um Ardha	7	0	5	2	Milk		Resident		7
4	Bahry	Eltibna	12	0	12	0	Milk		Resident		12
5	Bahry	Abo Halima	2	0	2	0	Milk		Resident		2
6	Bahry	Elsillat North	5	0	4	1	Milk		Resident		5
7	Sherg Elneel	Hilat koko	2	0	2	0	Milk		Resident		2
8	Sherg Elneel	Elkeryab	3	0	3	0	Milk		Resident		3
9	Sherg Elneel	Elshigla	4	0	4	0	Milk		Resident		4
10	Sherg Elneel	Elsillat South	7	1	6	2	Milk		Resident		8
11	Umbadda	Elrodoan project	14	1	15	0	Milk		Resident		15
12	Karary	Elahamda project	18	2	19	1	Milk			Transhuman	20
Total No. of serum											85

2.4 Sampling Strategy and Study design:

A cross-sectional study was conducted to estimate sero-positivity of Foot and Mouth Disease virus sero-Type A in cattle and to investegate posible risk factors associated with the disease in Khartoum state using multi-stage sampling strategy.

Using the probability sampling methods to select cattle, a single visit to farms was made to collect samples and fill out the questionnaire. Twelve locations were visited from the 6 localities of Khartoum state (Figure 2.3). Within the selected cattle farms cattle were selected using simple random sampling, thus 85 blood samples were collected.

2.5 Serum Neutralization Assay Description and Principle:

Virus Neutralization is a micro test for FMD antibodys performed with BHK-21 cells in flat bottomed tissue culture microtitre plates. The sera were inactivated at 56˚C for 30 minute before testing (Chepkwony *et al.*, 2012). Serum neutralization has been the gold standard, when available, for the detection and quantitation of antiviral antibodies. Neutralizing antibody also attracts great interest because it is considered a direct correlate of protective antibody in vivo. For the assay of neutralizing antibody virus are set up, usually in a 96-well microtiter plate. a constant amount of the test serum, usually diluted serum from the test animal is added. The assay is based on the difference in virus titer between the two titrations. A constant amount of virus, usually 100 TCID50, obtained by previous titration and dilution of a stock virus, is mixed with serial, usually twofold dilutions, of the test serum. The highest dilution of serum that neutralizes the test dose of virus is the titer of the serum., end points indicated by cytopathology. The serum-virus mixtures were inoculated into disposable, nontoxic, sterile, plastic plates with 96 flat-bottomed wells in each of which a cell monolayer has been established. Plates are then incubated for 48 hours until the wells containing the "virus only" controls develop evidence of infection By neutralizing the infectivity of the virus, antibody protects the cells against viral

destruction the highest dilution of antibody that protects cells from the virus represents the titer of neutralizing antibody contained in the serum specimen. (Murphy *et al*, 1999).

2.5.1 Equipments:

Microscope	Inverted microscope (Olympus CK×31)
Micro-plates	Nunc TC microwell 96 FSI cat no 168055
Digital pipettes	Tips multichannel pipette and one channel pipette
Pipettes	sterile, glass
Bottles	sterile, glass bijoux and universals
Tips	Finntips, sterilised
Water bath	for heat inactivation - kept between 54˚C and 58˚C
Shaker	Mini orbital shaker
Mixer	Vortex mixer
Incubator	kept at +35˚C to +39 ˚C
Refrigerator	kept between +1˚C and +8 ˚C
Freezer	-20˚C
Micro- plate Seals	sterile, loose fitting lids (adhesive tape) approx 18x133mm
Reservoir	Plastic reservoir sterilised

2.5.2 Chemicals:

Antibiotics	To prevent bacterial growth
Disinfectants	Detol

2.5.3 Reagents and Organisms:

Reference Sera	Convalescent serum against standard virus isolates (-ve) control serum and (+ve) control serum
Diluent	GMEM - pH is 7.4-7.6
FMDV	Sudanese FMDV sero- type A isolated from Khartoum state 2011
Cell culture	BHK-21 cell line
Tested Sera	convalecent

2.5.4 procedure:

Steps were done in a Class II cabinet and freshly makeup detol was used for disinfection.

2.5.4.1 Inactivation of sera samples:

Serum samples were been thawed at room temprature and heat inactivated in water bath at 56˚C for 30 minute and allowed to cool at room temprature.

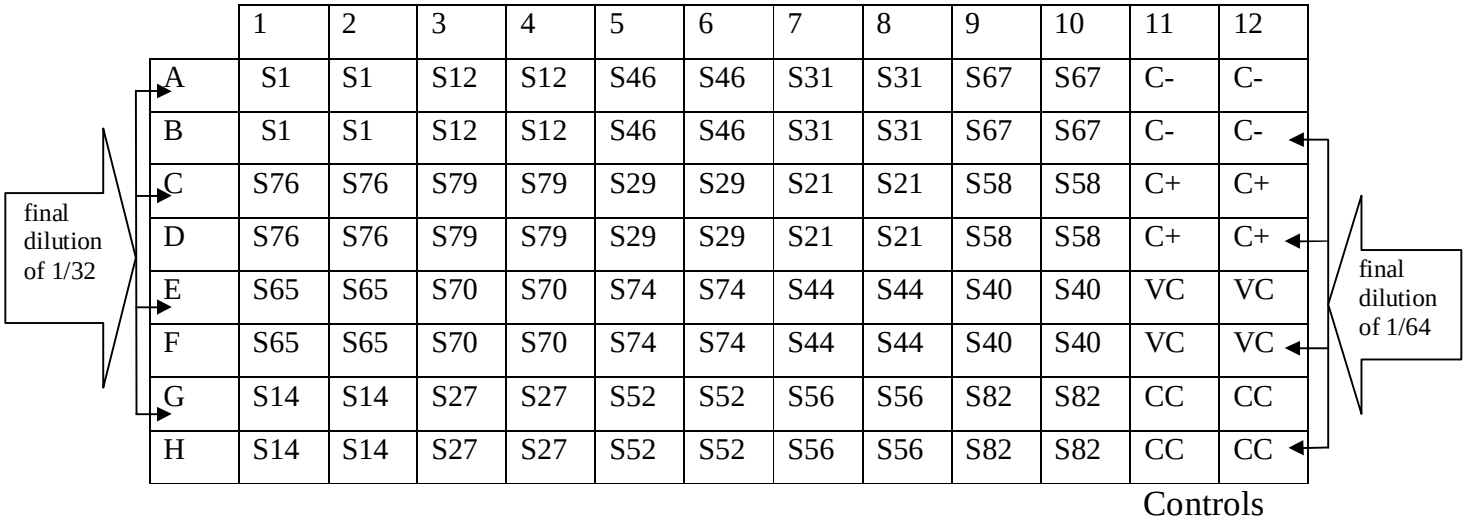
To prevent bacterial growth antibiotics were added (6 microliter of penicilline - sterptomicine compination and 6 microliter of Gentamicine), the compintion of penicillene – strptomicine was prepared according to the manufactures instructions and stored at -20˚C in freezer for 3-6 month while the Gentamicine stored at 4˚C in refregrator. After additon of antibiotics samples were stored in freezer at -20˚C til testing.

2.5.4.2 Dispense diluent into micro- plate (Test plates):

Serum diluent (complete GMEM containing 10% tris-buffer) was pour in a sterile reservoir and covered with a lid when not required. multichannel pipette was used to distribute 50µls diluent all through a u-bottomed microtitre plate then 46µls was added as follows: wells A1: A 12, C1: C12, E1: E12 and G1: G12. Plates were labled with tested sero-type and date of test, all plates were covered with Micro- plate seal to keep sterile between steps. Each microtitre plate tested 24 or 20 test sera in addition to controls.

2.5.4.3 Dispense serum samples into micro- plate:

Stored samples were thawed at room temprature, chosen randomly for plates then mixed by Vortex mixer. One channel pipette was used to poure serum samples in the micro- plate, a new tip was then used for each sample. Each serum was tested in 4 wells; 2 wells for each dilution. 6µls of each samples was added to each well of one pair (to detect adverse quality of test sera, their incomplete inactivation or presence of other agents affecting cell appearance), as Shown in (Figure 2. 4).



	1	2	3	4	5	6	7	8	9	10	11	12
A	S1	S1	S12	S12	S46	S46	S31	S31	S67	S67	C-	C-
B	S1	S1	S12	S12	S46	S46	S31	S31	S67	S67	C-	C-
C	S76	S76	S79	S79	S29	S29	S21	S21	S58	S58	C+	C+
D	S76	S76	S79	S79	S29	S29	S21	S21	S58	S58	C+	C+
E	S65	S65	S70	S70	S74	S74	S44	S44	S40	S40	VC	VC
F	S65	S65	S70	S70	S74	S74	S44	S44	S40	S40	VC	VC
G	S14	S14	S27	S27	S52	S52	S56	S56	S82	S82	CC	CC
H	S14	S14	S27	S27	S52	S52	S56	S56	S82	S82	CC	CC

Figure 2. 4: Test Plate Layout of VNT for FMDV sero-type A, each serum was tested in 4 wells (2 wells for final dilution of 1/32 and 2 wells for final dilution of 1/64).

S = Sample

C- = Negative serum control

C+ = Positive serum control

VC = Virus control

CC = Cell control

2.5.4.4 Dispense controls into micro- plate:

One channel pipette has used and a new tip was used for each control. Each of 4 wells, in columns 11 and 12 of a micotitre plate were done as follow: 6µls of (-ve) control was added to wells A 11-12, 6µls of (+ve) control was added to wells C 11-12, 6µls of diluent only was added to Virus control wells E 11-12 and 6µls of diluent only was added to cell control wells G11-12, as shwon in (Figer 2.4).

2.5.4.5 Dilution of samples and dispense FMDV sero-type A into micro-plate:

In the microtitre system contents of each well have been mixed, new tips were put onto a multichannel pipette, in row A by pipetting contents up and down few times (25 times and more), the row would test 6 sera at dilution 1/16 (final

dilution of 1/32). then transferd 50µls from each well and place it into row B for s erial twofold dilutions. Mixed as above then discarded 50µls from raw B leaving 50 µl of dilution 1/32 (final dilution of 1/64). same steps were done for raw C to dilute it in raw D, raw E to dillute it in F and raw G to dilute it in raw H (new tips were put onto a multichannel pipette before diluting different samples for each 2 rows).

Multistepper pipette was used 50µls of previously titrated FMDV sero-type A stock, containing 100 $TCID_{50}$, was dispensed into every serum dilutions wells of the micro-plate. Exccept the 4 wells of the cell control wells (E11-12 and F11-12) which received in place virus diluent (complete GMEM).The virus control received no serum but serum diluent. The positive and negative serum controls were reference sera or local bovine sera of known positivity. Plates were tightly covered and shaked with mini orbital shaker for 10 minute then kept at room temprature for an hour.

2.5.4.6 Dispense cell's suspension into micro- plate:

50µls of BHK-21 cell suspension, in the above described growth medium, has been dispensed to all wells of the plate exccept virus control wells(G11-12 and H11-12), suspension was agitated frequently to prevent sedimentation. Finally plates were covered with adhesive tape then tightly coverd with micro-plate seals and kept in Incubator at 37 ˚C, and with a source of humidity for about 48 hours, (all working surfaces and hands were disinfected) and plates were read microscopically 48 hours later. On the 3rd day post-seeding, the monolayers were stained with 0.1 % crystal violet in 10% formol-saline. CPE was indicated by cell lysis and, therefore, only uninfected monolayers were fixed and took up stain. Virus neutralization was marked by the presence of residual monolayers as shwon in (Figure 2.5).

2.6 Interpretation of results:

Reading of plates as follows: Score "+ve" for the well showing CPE which means FMDV sero type A not binded to its specific antibody so the virus

entered the BHK-21 cells for replication and cell death occured show the CPE which meant (-ve) for the VNT.

Score "-ve" for wells that showed no CPE which means the virus binded to its specific antibody (antigen antibody complex), that means (+ve) result of the VNT.

as shwon in (Figer 2.6).

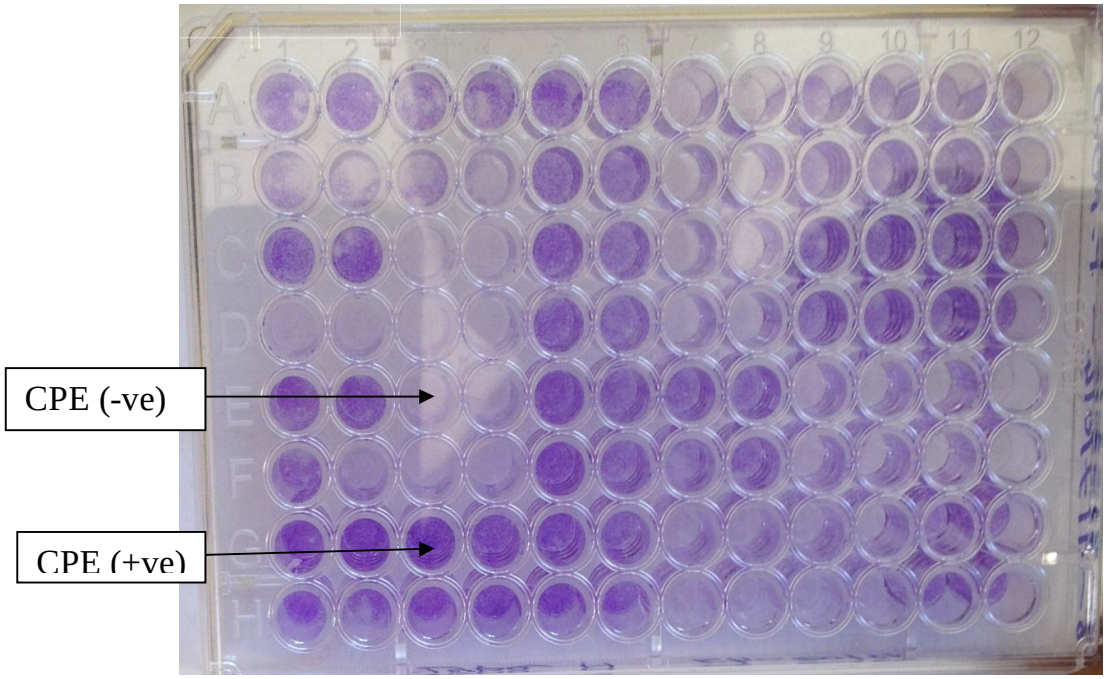


Figure 2.5: Screening format of VNT

Stained CPE (-ve) and CPE (+ve)

2.7 Data collection:

The primary data were collected through observations, structured questionnaires that target herdsmen. Moreover, the samples (85) were collected using probability sampling methods.

2.8 Questionnaire survey:

The questionnaire has been used to collect information from herdsmen, emphasizing data on hosts and environment. Cattle were chosen randomly from each farm then the questionnaire was filled out by interviewing the owners.

Host attributes included breed, species, age, sex, and body condition. Environment and management attributes included herd size, farm hygiene and type of husbandry system, history of FMD in the herd and the general management factors included manure disposal.

2.9 Data management and analysis:

For data analysis, data from quesionnare was entered, coded, and stored electronically in Microsoft Excel Spread sheet for Windows 2007. Analytical statistics were computed using Statistical Package for Social Sciences (SPSS) stastical softwae for Windows version 11.5.

All descriptive statistics computed for each variable eg; (age, breed, and locations) to summarize the data for seroprevalence. Frequency table (number of observations within variable), prevalence rates by cross tabulation (2 × 2 tables) was constructed to show seropositivity among groups of cattle (number of positive valid samples/number of individuals sampled in the variable).

Hypothesis testing for association between disease and potential risk factors were firstly, tested by univariate analysis by Chi-square (χ^2) test to test the significant diffference between the risk factors and FMD (the risk factor was considered significant in the univariate analysis at (P-value $\leq$0.30), these risk factors were entered in the multivariate analysis.

secondly, a multivariate analysis was performed by logistic regression model, to assess significant risk factors associated with sero-positivity of FMDV sero-type A in the study area. In the multivariate analysis, confidence level was at 95% and P-value $\leq$0.05 was considered significant.

Maps were produced using Arc GIS version 9.3 to show selected study states, localities, sero-prevalence rates of FMD by state and sero-prevalence rates of FMD by locality.

CHAPTER THREE

3. Results

3.1 Virus nutralization test results:

During the study period 85 animals were sampled, for the presence of antibodies against Foot and Mouth Disease Virus (FMDV) sero-type A in their blood samples using (VNT). Only 77 serum samples were tested because one serum has been discarded for heamolysis and the other 7 for laboratory complications. The overall seroprevalence of FMD seor-type A in Khartoum state was 68.8% (53/77) as in table 3.1.

Table 3.1: The overall prevalence rate of FMD sero- type A in Khartoum state

		Frequency	Percent	Cumulative Percent
Valid	Negative	24	31.2	31.2
	Positive	53	68.8	100.0
	Total	**77**	**100.0**	

The result showed that high seroprevalence of FMD Sero-type A in the tested sera collected from cattle in Khartoum state.

3.2 Descriptive statistical analysis frequency tables:

Frequency of FMD Sero-type A in result is shown in (table 3.2). Twenty four of cattle were negative and fifty three were positive. Therefore the overall prevalence rate was 68.8% (Table 3.1).

3.3 Analysis of risk factor for FMDV:

3.3.1 localities:

Results of frequency of FMD in locality is shown in (Table 3.2). The distribution of positive animals according to localities were in Khartoum 1 (100%), in Jabal Elawliaa 4 (40%), in Sherg Elneel 13 (81.3%), in Bahry 13

(76.5%), in Umbadda 8 (57.1%) and in Karary 14 (73.7%). The result of distribution and prevalence of FMD in locality were shown in (Table 3.3).

The results of association between localities and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysis is shown in (Table 3.4). Statistically there was no significant difference between the localities of Khartoum state ($p>0.05$).

3.3.2 Locations:

Results of frequency of FMD in location is shown in (Table 3.2). The distribution of positive animals according to locations were shown in (Table 3.3), they were from Elgeraf one sample and the prevalence was (100%), one from Sundus Project the prevalence was (33.3%), 3 from Um Ardha the prevalence was (42.9%), 2 from Hilat koko Mahleb area prevalence was (100%), 2 from Elkeryab the prevalence was (55.7%), 6 from Elsillat South prevalence was (85.7), 3 from Elshigla prevalence was (75%), 7 from Eltibna prevalence was (63.6%), 2 from Abo Halima prevalence was (100%), 4 from Elsillat North prevalence was (100%), 8 from Elrodoan project prevalence was (57.1%) and 14 from Elahamda projec prevalence was (73.7%).

The results of association between locations and VNT of FMDV sero-type (A) seropositvity are shown in (Table 3.4). Statistically there was no significant difference between locations of Khartoum state ($p>0.05$).

3.3.3 Sex

Results of frequency of FMD in sex is shown in (Table 3.2). 72 of cattle were female and 2 were male. Infection was higher in female (69.4%) compared with male (60%), results of distribution and prevalence of FMD in sex were shown in (Table 3.3).

The results of association between sex and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant variation in the seroprevalence of FMD in females and males ($p>0.05$).

3.3.4 Age

On the basis of age, animals were divided into three groups less than 2 Year (young), from 2 - 4 Year (medium age) and more than 4 Year (old), results of frequency of FMD in cattle is shown in (Table 3.2). 9 of cattle were young, 11 were mediun ages and 57 were old. Infection was higher in old cattle (77.2%) compared with medium (54.5%) and young cattle (33.3%) respectively.

the results of distribution and prevalence of FMD sero-type (A) in age were shown in (Table 3.3).

The results of association between age and VNT of FMDV sero-type (A) seropositivity is shown in (Table 3.4). Statistically there was significant association between age and FMD occurrence ($p<0.05$).

3.3.5 Breed

Results of frequency of FMD in breed is shown in (Table 3.2). 4 of cattle were local breed and 73 of cattle were cross breed . Infection was higher in local breed (100%) compared with cross breed (67.1%). For distribution and prevalence of FMD sero-type (A) in breed the results were shown in (Table 3.3). Results of association between breed and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysis is shown in (Table 3.4). Statistically there was no significant difference between breed categories ($p>0.05$).

3.3.6 Body condition

Result of frequency of FMD in body codition is shown in (Table 3.2). 71 cattle were categorized as good body condition and 6 were emacited. Infection was higher in cattle with good body condition (69%) compared with emaciated cattle (66.7%). For distribution and prevalence of FMD sero-type (A) in body condition, the results were shown in (Table 3.3).

The results of association between body codition and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the categories ($p>0.05$).

3.3.7 Herd size

Result of frequency of FMD sero-type (A)in herd size is shown in (Table 3.2). 8 of cattle were categorized in small herd size (less han 50 cattle), 50 cattle were in medium herd size (50 - 150 cattle) and 19 were in big herd size (more than 150 cattle). Infection was higher in small herd size (87.5 %) compared with big herd size(78.9%) and medium herd size (62%). For distribution and prevalence of FMD in Herd size, the results were shown in (Table 3.3).

The results of association between herd size and VNT of FMDV seropositvity were significantlly variable in the univariate analysis is shown in (Table 3.4). Statistically there was no significant difference between the categories ($p>0.05$).

3.3.8 Production purpose

Results of frequency of FMD in production purpose is shown in (Table 3.2). 74 of cattle were categorized as mik prodution and the other groop as milk and meat production. Infection was higher in cattle used for milk production (70.3%) compared with cattle used for milk and meat production (33.3%). For distribution and prevalence of FMD in production purpose, the results were shown in (Table 3.3).

The results of association between production purpose and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4).

Statistically there was no significant difference between the two groop ($p>0.05$).

3.3.9 Herd type mixed with other species:

Regarding herd type mixed with other spcies results of frequency of FMD is shown in (Table 3.2). 27 cattle were not mixed with other species and 50 of cattle were mixed. Infection was higher in cattle not mixed cattle (74.1%) compared with cattle mixed with other species (66%). For the prevalence of FMD in Herd type mixed with other species, the results were shown in (Table 3.3). The results of association between Herd type mixed with other species and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4).

Statistically there was no significant difference between the two groop ($p>0.05$).

3.3.10 Mixed species:

Results of frequency of FMD in mixed species is shown (Table 3.2). 27 of cattle were not mixed with other species,7 of cattle were mixed with goat, 3 of cattle were mixed with sheep and 40 of cattle were mixed with sheep and goat. Infection was higher in cattle not mixed (74.1%) compared with cattle mixed with sheep and goat (70%), cattle mixed with sheep (66.7) and cattle mixed with goat (42.9%). For distribution and prevalence of FMD results were shown in (Table 3.3).

The results of association between mixed species and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4).

Statistically there was no significant difference between the catigories ($p>0.05$).

3.3.11 Previous history of FMD in the herd:

Results of frequency of FMD in previous history of FMD in the herd is shown in (Table 3.2). 20 of cattle did not have previous history of FMD in their herd and 57 of cattle had previous history of FMD in their herd. Infection was higher in cattle that have previous history of FMD (71.9%) compared with cattle that did not have previous history of FMD (60%). For distribution and prevalence of FMD sero-type (A) in previous history of FMD in the herd, the results were shown in (Table 3.3).

The results of association between previous history of FMD in the herd and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4).

Statistically there was no significant difference between the categories ($p>0.05$).

3.3.12 Other diseases:

Results of frequency of FMD in other diseases is shown in (Table 3.2). 54 of cattle did not affected with other disease, 14 cattle were affected with Theileriasis, 7 cattle have abortion and 2 of cattle were suspected with John's. Infection was high in cattle which had suspected cases of John's (100%) compared with other categories. Prevalence of FMD sero-type (A) in cattle affected with other diseases the results were shown in (Table 3.3).

The results of association between other diseases and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the catigories ($p>0.05$).

3.3.13 Production System:

Results of frequency of FMD in production system is shown in (Table 3.2). 59 of cattle were resident and 18 were transhumant. Infection was higher in transhumant cattle (72.2%) compared to resident cattle (67.8%). For distribution prevalence of FMD sero-type (A) in production system, the results were shown in (Table 3.3).

The results of association between herd type mixed with other species and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the catigories ($p>0.05$).

3.3.14 Farm construction:

Results of frequency of FMD in farm construction is shown in (Table 3.2). 32 of cattle were kept in closed sheds mud and the throne of mats, 15 cattle were kept in brick stables and zinc roof, 26 cattle were kept in pens of iron with roof of iron and mats, 1 cow was kept in pens of zinc and iron roof and floor mats and 3 cattle were kept in pens of firewood. Infection was higher in cattle kept in pens of zinc and iron roof and floor mats (100%) compared with other cattle. For distribution prevalence of FMD sero-type (A) in construction of farm, the results were shown in (Table 3.3).

The results of association between construction of farm and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the categories ($p>0.05$).

3.3.15 Ventilation:

Results of frequency of FMD in ventilation is shown in (Table 3.2). all of the cattle were kept in good ventilation; 53 out of 77 cattle had FMD (68.8%). For distribution prevalence of FMD sero-type (A) in ventilation, the results were shown in (Table 3.3).

The results of association between ventilation and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant associtionbetween the ventilation and the ocurrence of FMD.

3.3.16 Grazing system:

Results of frequency of FMD in grazing system is shown in (Table 3.2). 73 of cattle were have private grazing system, 4 were categorized as private and common grazing. Infection was higher in cattle category graze private grazing system (69.9%) compared with the category private and common grazing (50%). For distribution prevalence of FMD sero-type (A) in grazing system, the results were shown in (Table 3.3).

The results of association between grazing system and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between private and private and common grazing ($p>0.05$).

3.3.17 Water resources:

Results of frequency of FMD in water resources is shown in (Table 3.2). 8 of cattle waterd from Private tap water (well in the area project), 13 cattle waterd from private tap water, 8 cattle waterd from private water resources, 3 waterd from private tap water and common from Nile canal in the area and 45 of cattle waterd from private tap water from well in the area. Infection was higher in cattle watered private; tap water from well in the area as well as cattle watered privatly (75%) compared with other cattle. Prevalence of FMD sero-type (A) in water resources, the results were shown in (Table 3.3).

The results of association between water resources and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the water resources categories ($p>0.05$).

3.3.18 Use of concentrates:

Results of frequency of FMD in cattle fed with concentrates is shown in (Table 3.2). all of the cattle were fed with concentrates; 53 out of 77 cattle had FMD

(68.8%). Distribution prevalence of FMD sero-type (A) in using of concentrates, the results were shown in (Table 3.3).

For the association between using concentrates and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant associtionbetween using concentrates and the ocurrence of FMD.

3.3.19 Artificial insemination:

Results of frequency of FMD in artificial insemination is shown in (Table 3.2). 54 of cattle were not artificially inseminated and 23 were artificially inseminated. Comparatively the percentage was higher in cattle not artificially inseminated (74.1%) compared with cattle artificially inseminated (56.5%). For distribution prevalence of FMD sero-type (A) in artificial insemination, the results were shown in (Table 3.3).

The results of association between artificial insemination and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysi is shown in (Table 3.4). Statistically there was no significant difference between the cattle used artificial insemination and not used it ($p>0.05$).

3.3.20 Where stay in dry season:

Results of frequency of FMD in where stay in dry season is shown in (Table 3.2), all of the cattle were stayed in the farm in dry season; 53 out of 77 cattle had FMD (68.8%). For distribution prevalence of FMD sero-type (A) in where stay in dry season the results were shown in (Table 3.3).

The results of association between where cattle stay in dry season and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant associtionbetween where stay in dry season and the ocurrence of FMD.

3.3.21 Where stay in rainy season:

Result of frequency of FMD in where stay in rain season is shown in (Table 3.2). 58 of cattle were stayed in the farm at rain season and 19 were stayed out of the farm. Infection was higher in cattle stayed out farm in rain season (73.7%)

compared to cattle stayed in farm in rain season (67.2%). For distribution and prevalence of FMD sero-type (A) in where stay in rain season, the results were shown in (Table 3.4).

The results of association between where stay in rain season and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between the two categories ($p>0.05$).

3.3.22 Contact with neighboring herd:

Result of frequency of FMD in contact with neighboring herd is shown in (Table 3.2). 38 of cattle were not in contact with neighboring herds and 39 of cattle were in contact with neighboring herd. Infection was higher in cattle in contact with neighboring herds (71.8%) compared with cattle that were not incontact with neighboring herds (65.8%). For distribution prevalence of FMD sero-type (A) in contact with neighboring herd, results were shown in (Table 3.3).

The results of association between contact with neighboring herd and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically no significant difference between cattle in contact with neighboring herd and cattle not in contact ($p>0.05$).

3.3.23 Test of new animals before placement in the herd:

Results of frequency of FMD in test of new animals before placement in the herd is shown in (Table 3.2). 14 of cattle owners test new animals before placement in the herd and 63 did not test new animals before placement in the herd. Infection was higher in that not tested before placement in the herd (71.4%) compared to that tested before placement in the herd (57.1%). For prevalence distribution of FMD in test of new animals before placement in the herd, the results were shown in (Table 3.3).

The results of association between test of new animals before placement in the herd and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysis is shown in (Table 3.4). Statistically there was no

significant difference between test of new animals before placement in the herd or not test of new animals before placement in the herd ($p>0.05$).

3.3.24 Isolation pen for sick animals:

Results of frequency of FMD in isolation pen for sick animals is shown in (Table 3.2). 27 of cattle; owners have isolation pens for sick animals and 50 of cattle; owners not have isolation pens for sick animals. Infection were higher in category cattle; owner have not isolation pens for sick (72%) compared with the category cattle; owner have Isolation pens for sick animals (62.9%). For distribution and prevalence of FMD in isolation pen for sick animals, the results were shown in (Table 3.3).

The results of association between isolation pen for sick animals and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between cattle with isolation pen for sick or not have isolation pens for sick animals ($p>0.05$).

3.3.25 Visit of workers to other farms:

Results of frequency of FMD in visit of workers to other farms is shown in (Table 3.2). 30 of cattle; workers not visit other farms and 47 of cattle; workers visit other farms. Infection were higher in workers visit other farms (72.3%) compared with workers not visit other farms (63.3%). For distribution and prevalence of FMD in visit of workers to other farms, the results were shown in (Table 3.3). The results of association between Isolation pen for sick animals and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between workers visit other farms or workers not visit other farms ($p>0.05$).

3.3.26 Awareness of farmer with FMD:

Results of frequency of FMD in awareness of farmer with FMD is shown in (Table 3.2). all cattle owners were aware with FMD althow 53 cattle were positive of FMD (68.8%). For distribution prevalence of FMD sero-type (A) in awareness of farmer with FMD the, results were shown in (Table 3.3).

The results of association between awareness of farmer with FMD and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically There was not any role of awareness of farmer and FMD ocurrence.

3.3.27 Disposal of carcasses:

Result of frequency of FMD in disposal of carcasses is shown in (Table 3.2). 24 of cattle; for disposal of carcasses have burning in incinerator of the project, 22 of cattle; for disposal of carcasses burning out of the farm and 31 of cattle; for disposal of carcasses throw out farm. Infection was higher in categor disposal of carcasses throw out farm (77.4%) compared with other categories. For distribution prevalence of FMD sero-type (A) in disposal of carcasses, the results were shown in (Table 3.3).

The results of association between disposal of carcasses and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.5). Statistically there was no significant difference between disposal of carcasses categories ($p>0.05$).

3.3.28 Manure disposal:

Results of frequency of FMD in manure disposal is shown in (Table 3.2). 10 of cattle; for manure disposal sell it to bricks makers weekly, 2 of cattle; for manure disposal store out farm till selling and 65 of cattle; for manure disposal store in the farm till selling. Infection was higher in category store out farm till selling (100%) compared to sell to bricks makers weekly and store out farm till selling (40%) and (72.3%) respectivly. For distribution prevalence of FMD sero-type (A) in manure disposal, the results were shown in (Table 3.3).

The results of association between manure disposal and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysis is shown in (Table 3.4). Statistically there was no significant difference between manure disposal categories ($p>0.05$).

3.3.29 Veterinary services:

Results of frequency of FMD in veterinary services is shown in (Table 3.2). 33 of cattle were supervised daily by veterinarian, 43 of cattle were supervised by

veterinary public and private hospital or clinic in the area and 1 cattle was supervised regularly by veterinarian. Infection was higher in cattle regularly supervised by veterinarian (100%) compared to cattle supervised by public and private hospital or clinic in the area (72.1%) and to cattle supervised daily by veterinarian (27.3%) and regular veterinary supervision (63.6%). For distribution prevalence of FMD sero-type (A) in veterinary services, the results were shown in (Table 3.3).

The results of association between veterinary services and VNT of FMDV sero-type (A) seropositvity were significantlly variable in the univariate analysi is shown in (Table 3.4). Statistically there was no significant difference between veterinary services categories ($p>0.05$).

3.3.30 Use of FMD vaccine to control the disease:

Result of frequency of FMD in use of FMD vaccine to control the disease is shown in (Table 3.2). 9 of cattle vaccinated against FMD and 68 of cattle were not vaccinated. Infection was higher in unvaccinated cattle (70.6%) compared with cattle vaccinated againist FMD (55.6%). For distribution and prevalence of FMD in use of FMD vaccine to control the disease, the results were shown in (Table 3.3).

The results of association between use of FMD vaccine to control the disease and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between use of FMD vaccine or not use it to control the disease ($p>0.05$).

3.3.31 Use of treatment to control FMD:

Results of frequency of FMD in use of treatment to control FMD is shown in (Table 3.2). 19 of cattle treated with antibiotics, 3 of cattle treated traditionaly with pure bee honey for three days, 31 of cattle treated traditionaly with *cassia nilotica* and glycerin and 24 of cattle were not treated. Infection was higher in the category treated with antibiotics (73.7%) compared to cattle treated with *Acassia nilotica* and glycerin (70.9%), un treated cattle (66.7%) and cattle

treated with pure bee honey for three days (33.3). For distribution prevalence of FMD sero-type (A) in other treatment to control FMD, the results were shown in (Table 3.3). The results of association between other treatment to control FMD and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between other treatment to control FMD categories ($p>0.05$).

3.3.32 Other information:

Results of frequency of FMD in other information is shown in (Table 3.2). 2 of cattle affected with CBPP, tick infestation and suspected with John's disease, 3 cattle affected with Brucellosis, tick infestation and calves diarrhea, 7 of cattle affected with Brucelloces, tick infestation and mastitis, 11 of cattle affected with Theileriasis, tick infestation and House flies, 4 of cattle affected with Theileriasis and mastitis, 3 of cattle affected with CBPP, tick infestation and Theileriasis.7 of cattle had Pneumonia, calves diarrhea and suspected with Brucella, 4 of cattle affected with CBPP, calves diarrhea, suspected with Brucella, tick infestation, 2 of cattle affected with calves diarrhea and tick infestation, 19 of cattle had tick infestation and Theileriasis, 3 of cattle affected with mastitis and Lumpy skin disease, 1 of cattle affected with mastitis and 5 of cattle affected with Theileriasis, tick infestation and mastitis. Infection was higher in the category cattle afected with CBPP, tick infestation and suspected with John's disease, Theileriasis and mastitis category, calves diarrhea tick infestation and Theileriasts category, mastities and calves diarrhea category, calves diarrhea and Tick infestation category and mastities category (100%) compared to other categories. For distribution prevalence of FMD sero-type (A) in other information, the results were shown in (Table 3.3).

The results of association between other information and VNT of FMDV sero-type (A) seropositvity is shown in (Table 3.4). Statistically there was no significant difference between other information categories ($p>0.05$).

3.3.33 Summary of frequencies for the distributions of FMDV according to potential risk factors; locality, location, sex, age, breed, body condition, herd size, production purpose, herd type mixed with other species, mixed species, other diseases, previous history of FMD in the herd, other disease, production system, farm construction, ventilation, grazing system, water resources, use of concentrates, artificial insemination, where stay in dry season, where stay in rainy season, contact with neighboring herd, test of new animals before placement in the herd, isolation pen for sick animals, visit of workers to other farms, awareness of farmer with FMD, disposal of carcasses, manure disposal, veterinary services, use of FMD vaccine to control the disease, use of treatment to control FMD and other information) in Khartoum state, 2014 as showen in (Table 3.2).

Table 3.2: Summary of frequencies for the distribution of 77 cattle examined in Khartoum State, 2014 according to potential risk factors

Risk Factors and its levels	Number of tested samples	Relative Frequency (%)	Cumulative Frequency (%)
Localities			
- Khartoum	1	1.3	1.3
- Jabal Elawliaa	10	13.0	14.3
- Sherg Elneel	16	20.8	35.1
- Bahry	17	22.1	57.1
- Umbadda	14	18.2	75.3
- Karary	19	24.7	100.0
Locations			
- Elgeraf	1	1.3	1.3
- Sundus Project	3	3.9	5.2
- Um Ardha	7	9.1	14.3
- Hilat koko Mahleb area	2	2.6	16.9
- Elkeryab	3	3.9	20.8
- Elsillat South	7	9.1	29.9
- Elshigla	4	5.2	35.1
- Eltibna	11	14.3	49.4
- Abo Halima	2	2.6	51.9
- Elsillat North	4	5.2	57.1
- Elrodoan project	14	18.2	75.3
- Elahamda projec	19	24.7	100.0
Sex			
- Female	72	93.5	93.5
- Male	5	6.5	100.0
Age			
- <2 year young	9	11.7	11.7
- 2 – 4 year (medium age)	11	14.3	26.0
- > 4 year (old)	57	74.0	100.0
Breed			
Local	4	5.2	5.2
Cross	73	94.8	100.0
Body condition			
- good body condition	71	92.2	92.2
- emaciated	6	7.8	100.0
Herd size			
- <50 cattle (small herd)	8	10.4	10.4
- 50 - 150 cattle (medium herd)	50	64.9	75.3
- >150 cattle (big herd)	19	24.7	100.0

Table 3.2: continued

Risk Factors and its levels	Number of tested samples	Relative Frequency (%)	Cumulative Frequency (%)
Production purpose			
- Milk	74	96.1	96.1
- Milk and meat	3	3.9	100.0
Herd type mixed with other species			
- No	27	35.1	35.1
- Yes	50	64.9	100.0
Mixed specie:			
- Not mixed	27	35.1	53.1
- Goat	7	9.1	44.2
- Sheep	3	3.9	48.1
- Sheep and Goat	40	51.9	100.0
Previous history in the herd			
- No	20	26.0	26.0
- Yes	57	74.0	100.0
Other diseases			
- No others	54	70.1	70.1
- Theileriasis	14	18.2	88.3
- abortion	7	9.1	97.4
- Suspected with John's disease	2	2.6	100.0
Production system			
- Resident	59	76.6	76.6
- Transhumant	18	23.4	100.0
Farm construction			
- Closed sheds of mud with roof of mats	32	41.6	41.6
- Brick stables and zinc roof	15	19.5	61.0
- pens of iron with roof of iron and mats	26	33.8	94.8
- pens of zinc with roof of iron and mats	1	1.3	96.1
- pens of firewood	3	3.9	100.0
Ventilation			
- Good	77	100.0	100.0
- Bad	0	0	100.0
Grazing system			
- private	73	94.8	94.8
- private and common	4	5.2	100.0

Table 3.2: continued

Risk Factors and its levels	Number of tested samples	Relative Frequency (%)	Cumulative Frequency (%)
Water resources			
- Private tap water from well in the area (project)	8	10.4	10.4
- Private tap water	13	16.9	27.3
- Private	8	10.4	37.7
- Private tap water and common from Nile canal in the area	3	3.9	41.6
- Private tap water from well in the area	45	58.4	100.0
Use of concentrate			
- Yes	77	100.0	100.0
- No	0	0	100.0
Artificial insemination			
- No	54	70.1	70.1
- Yes	23	29.9	100.0
Where stay in dry season			
- In the farm	77	100.0	100.0
- Out of the farm	0	0	100.0
Where stay in rainy season			
- In the farm	58	75.3	75.3
- Out of the farm	19	24.7	100.0
Contact with neighboring herd			
- No	38	49.4	49.4
- Yes	39	50.6	100.0
Test of new animals before placement in the herd			
- Yes	14	18.2	18.2
- No	63	81.8	100.0
Isolation pen for sick animals			
- Yes	27	35.1	35.1
- No	50	64.9	100.0
Visit of workers to other farms			
- No	30	39.0	39.0
- Yes	47	61.0	100.0

Table 3.2: continued

Risk Factors and its levels	Number of tested samples	Relative Frequency (%)	Cumulative Frequency (%)
Awareness of farmer with FMD			
- Yes	77	100.0	100.0
- No	0	0	100.0
Disposal of carcasses			
- burning in incinerator of the project - burning in incinerator	24	31.2	31.2
- Burning out of the farm	22	28.6	59.7
- Throw out farm	31	40.3	100.0
Manure Disposal			
- sell to bricks makers weekly	10	13.0	13.0
- store out farm till selling	2	2.6	15.6
- store in the farm till selling	65	84.4	100.0
Veterinary services			
- daily supervision of a veterinarian	33	42.9	42.9
- Veterinary, public and private hospital or clinic in the are	43	55.8	98.7
- regular veterinary supervision	1	13	100.0
Use of FMD vaccine to control the disease			
- Yes	9	11.7	11.7
- No	68	88.3	100.0
Use of treatment to control FMD			
- Antibiotics	19	24.7	24.7
- pure bee Haney three days	3	3.9	28.6
- *Acacia nilotica* and glycerin	31	40.3	68.8
- No	24	31.2	100.0

Table 3.2: continued

Risk Factors and its levels	Number of tested samples	Relative Frequency (%)	Cumulative Frequency (%)
Other information			
- CBPP, Tick infestation and suspected with John's disease.	2	2.6	2.6
- Brucellla, Tick infestation and calves diarrhea.	3	3.9	6.5
- Brucellla, Tick infestation and mastitis	7	9.1	15.6
- Theileriasis Tick infestation and House flies	11	14.3	29.9
- Theileriasis and mastitis	4	5.2	35.1
- CBPP, Tick infestation and Theileriasis	3	3.9	39.0
- calves diarrhea, Tick infestation and theileriasts	4	5.2	44.2
- Pneumonia, calves diarrhea and suspected with Brucella.	7	9.1	53.2
- CBPP, calves diarrhea, suspected with Brucella, Tick infestation	4	5.2	58.4
- mastitis and calves diarrhea	2	2.6	61.0
- calves diarrhea and Tick infestation	2	2.6	63.6
- Tick infestation and Theileriasis.	19	24.7	88.3
- mastitis and lumpy skin disease	3	3.9	92.2
- mastitis	1	1.3	93.5
- Theileriasis, Tick infestation and mastitis	5	6.5	100.0
Result			
- Negative	24	31.2	31.2
- positive	53	68.8	100.0
Total	77	100.0	

3.3.34 Summary of cross tabulation for the distributions of FMDV according to potential risk factors; locality, location, sex, age, breed, herd size, body condition, production purpose, herd type mixed with other species, mixed species, other diseases, previous history of FMD in the herd, production system, farm construction, ventilation, grazing system, water resources, use of concentrates, artificial insemination, where stay in dry season, where stay in rainy season, contact with neighboring herd, test of new animals before placement in the herd, isolation pen for sick animals, visit of workers to other farms, awareness of farmer with FMD, disposal of carcasses, manure disposal, veterinary services, use of FMD vaccine to control the disease, use of treatment to control FMD and other information) in Khartoum state, 2014. (Table 3.3).

Table 3.3: Summary of cross tabulation for the prevalence of FMD sero-type A with potential risk factors in 77 cattle in Khartoum State 2014

Risk Factors and its levels	Number of tested samples	No. (+ve)	(+ve) (%)
- Localities			
- Khartoum	1	1	100.0
- Jabal Elawliaa	10	4	40.0
- Sherg Elneel	16	13	81.3
- Bahry	17	13	76.5
- Umbadda	14	8	57.1
- Karary	19	14	73.7
Locations			
- Elgeraf	1	1	100.0
- Sundus Project	3	1	33.3
- Um Ardha	7	3	42.9
- Hilat koko Mahleb area	2	2	100.0
- Elkeryab	3	2	66.7
- Elsillat South	7	6	85.7
- Elshigla	4	3	75.0
- Eltibna	11	7	63.6
- Abo Halima	2	2	100.0
- Elsillat North	4	4	100.0
- Elrodoan project	14	8	57.1
- Elahamda projec	19	14	73.7
Sex			
- Female	72	50	69.4
- Male	5	3	60
Age			
- <2 year young	9	3	33.3
- 2 – 4 year (medium age)	11	6	54.5
- > 4 year (old)	57	44	77.2
Breed			
- Local	4	4	100.0
- Cross	73	49	67.1
Body condition			
- good body condition	71	49	69.0
- emaciated	6	4	66.7
Herd size			
- <50 cattle (small herd)	8	7	87.5
- 50 - 150 cattle (medium herd)	50	31	62.0
- >150 cattle (big herd)	19	15	78.9

Table 3.3: continued

Risk Factors and its levels	Number of tested samples	No. (+ve)	(+ve) (%)
Production purpose			
- Milk	74	52	70.3
- Milk and meat	3	1	33.3
Herd type mixed with other species			
- No	27	20	74.1
- Yes	50	33	66.0
Mixed specie:			
- Not mixed	27	20	74.1
- Goat	7	3	42.9
- Sheep	3	2	66.7
- Sheep and Goat	40	28	70.0
Previous history in the herd			
- No	20	12	60.0
- Yes	57	41	82.0
Other diseases			
- No others	54	38	70.4
- Theileriasis	14	9	64.3
- abortion	7	4	57.1
- Suspected with John's disease	2	2	100.0
Production system			
- Resident	59	40	67.8
- Transhumant	18	13	72.2
Farm construction			
- Closed sheds of mud with roof of mats	32	22	68.8
- Brick stables and zinc roof	15	11	73.3
- pens of iron with roof of iron and mats	26	17	65.4
- pens of zinc with roof of iron and mats	1	1	100.0
- pens of firewood	3	2	66.7
Ventilation			
- Good	77	53	68.8
- Bad	0	0	0
Grazing system			
- private	73	51	69.9
- private and common	4	2	50.0

Table 3.3: continued

Risk Factors and its levels	Number of tested samples	No. (+ve)	(+ve) (%)
Water resources			
- Private tap water from well in the area (project)	8	6	75.0
- Private tap water	13	8	61.5
- Private	8	6	75.0
- Private tap water and common from Nile canal in the area	3	1	33.3
- Private tap water from well in the area	45	32	71.1
Use of concentrate			
- Yes	77	53	68.8
- No	0	0	0
Artificial insemination			
- No	54	40	74.1
- Yes	23	13	56.5
Where stay in dry season			
- In the farm	77	53	68.8
- Out of the farm	0	0	0
Where stay in rainy season			
- In the farm	58	39	67.2
- Out of the farm	19	14	73.7
Contact with neighboring herd			
- No	38	25	65.8
- Yes	39	28	71.8
Test of new animals before placement in the herd			
- Yes	14	8	57.1
- No	63	45	71.4
Isolation pen for sick animals			
- Yes	27	17	63.0
- No	50	36	72.0
Visit of workers to other farms			
- No	30	19	63.3
- Yes	47	34	72.3

Table 3.3: continued

Risk Factors and its levels	Number of tested samples	No. (+ve)	(+ve) (%)
Awareness of farmer with FMD			
- Yes	77	53	68.8
- No	0	0	0
Disposal of carcasses			
- burning in incinerator of the project - burning in incinerator	24	15	62.5
- Burning out of the farm	22	14	63.6
- Throw out farm	31	24	77.4
Manure Disposal			
- sell to bricks makers weekly	10	4	40.0
- store out farm till selling	2	2	100.0
- store in the farm till selling	65	47	72.3
Veterinary services			
- daily supervision of a veterinarian	33	21	63.6
- Veterinary, public and private hospital or clinic in the are	43	31	72.1
- regular veterinary supervision	1	1	100.0
Use of FMD vaccine to control the disease			
- Yes	9	5	55.6
- No	68	48	70.6
Use of treatment to control FMD			
- Antibiotics	19	14	73.7
- pure bee Haney for three days	3	1	33.3
- *Acacia nilotica* and glycerin	31	22	71.0
- No	24	16	66.7

Table 3.3: continued

Risk Factors and its levels	Number of tested samples	No. (+ve)	(+ve) (%)
Other information			
- CBPP, Tick infestation and suspected with John's disease.	2	2	100.0
- Brucellla, Tick infestation and calves diarrhea.	3	1	33.3
- Brucellla, Tick infestation and mastitis	7	3	42.9
- Theileriasis Tick infestation and House flies	11	7	63.6
- Theileriasis and mastitis	4	4	100.0
- CBPP, Tick infestation and Theileriasis	3	2	66.7
- calves diarrhea, Tick infestation and theileriasts	4	4	100.0
- Pneumonia, calves diarrhea and suspected with Brucella	7	4	57.1
- CBPP, calves diarrhea, suspected with Brucella, Tick infestation	4	3	75.0
- mastitis and calves diarrhea	2	2	100.0
- calves diarrhea and Tick infestation	2	2	100.0
- Tick infestation and Theileriasis.	19	12	80.0
- mastitis and lumpy skin disease	3	2	66.7
- mastitis	1	1	100.0
- Theileriasis, Tick infestation and mastitis	5	4	80.0

3.3.35 Hypothesis testing for association between FMD sero- type A and potential risk factors in univariate analysis were tested by Chi-square Tests (χ^2) to test the significant difference between the risk factors and FMD - in 77 tested cattle in Khartoum State (the risk factor was considered significant at (P-value $\leq$0.3). The result showed that there were 7 risk factors statistically significant. These were locality (p- value = 0.218), age (p- value = 0.017), breed (P- value = 0.167), herd size (P- value = 0.193), Artificial insemination (P- value = 0.128), test of new animal befor placement in the herd (P- value = 0.297) and Manure disposal (P- value = 0.076), (Table 3.4).

Table 3.4: Univariate analysis of potential risk factors with VNT- in 77 tested cattle in Khartoum State using Chi – square (χ^2) test

Risk Factor	Total No. of tested samples	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Locality				7.0	5	0.218
- Khartoum	1	1	100.0			
- Jabal Elawliaa	10	4	40.0			
- Sherg Elneel	16	13	81.3			
- Bahry	17	13	76.5			
- Umbadda	14	8	57.1			
- Karary	19	14	73.7			
Location				10.2	11	0.505
- Elgeraf	1	1	100.0			
- Sundus Project	3	1	33.3			
- Um Ardha	7	3	42.9			
- Hilat koko Mahleb area	2	2	100.0			
- Elkeryab	3	2	55.7			
- Elsillat South	7	6	85.7			
- Elshigla	4	3	75.0			
- Eltibna	11	7	63.6			
- Abo Halima	2	2	100.0			
- Elsillat North	4	4	100.0			
- Elrodoan project	14	8	57.1			
- Elahamda projec	19	14	73.7			
Sex				0.194	1	0.659
- Female	72	50	69.4			
- male	5	3	60.0			
Age				8.190	2	0.017
- <2 year (young)	9	3	33.3			
- 2 – 4 year (medium age)	11	6	54.5			
- > 4 year (old)	57	44	77.2			
Breed				1.911	1	0.167
- local	4	4	100.0			
- cross	73	49	67.1			

Table 3.4: continued

Risk Factor	Total No. of tested samples	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Body condition				0.014	1	0.905
- good body condition	71	49	69.0			
- emaciated	6	4	66.7			
Herd size				3.294	2	0.193
- <50 cattle (small herd)	8	7	87.5			
- 50 - 150 cattle (medium herd)	50	31	62.0			
- >150 cattle (big herd)	19	15	78.9			
Production purpose				0.533	1	0.465
- milk	74	52	70.3			
- milk and meat	3	1	33.3			
Herd type mixed with other species				533	1	0.465
- No	27	20	74.1			
- Yes	50	33	660			
Mixed species				2.579	3	0.461
- Not mixed	27	20	74.1			
- Goat	7	3	42.9			
- Sheep	3	2	66.7			
- Sheep and Goat	40	28	70.0			
Previous history of FMD in the herd				0.982	1	0.322
- No	20	12	60.0			
- Yes	57	41	71.9			
Other diseases				1.546	3	0.672
- No disease	54	38	70.4			
- Theileriasis	14	9	64.3			
- abortion	7	4	57.1			
- Suspected with John's disease	2	2	100.0			
Production System				0.126	1	0.723
- Resident	59	40	67.8			
- Transhumant	18	13	72.2			

Table 3.4: continued

Risk Factor	Total No. of tested samples	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Farm construction				0.745	4	0.946
- Closed sheds of mud with roof of mats	32	22	68.8			
- Brick stables and zinc roof	15	11	73.3			
- pens of iron with roof of iron and mats	26	17	65.4			
- pens of zinc with roof of iron and mats	1	1	100			
- pens of firewood	3	2	66.7			
Ventilation				(a)		
good	77	53	68,8			
Grazing system				0.697	1	0.404
- private	73	51	69.9			
- private and common	4	2	50.0			
Water resources				2.477	4	0.649
- Private tap water from well in the area (project)	8	6	75.0			
- Private tap water	13	8	61.5			
- Private	8	6	75.0			
- Private tap water and common from Nile canal in the area	3	1	33.3			
- Private tap water from well in the area	45	32	71.1			
Use of concentrates				(a)		
Yes	77	53	68.8			
Artificial insemination				2.316	1	0.128
- No	54	40	74.1			
- Yes	23	13	56.5			

Table 3.4: continued

Risk Factor	Total No.	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Where stay in dry season				(a)		
in the farm	77	53	68.8			
Where stay in rainy season				0.277	1	0.599
- in the farm	58	39	67.2			
- out of the farm	19	14	73.7			
Contact with neighboring herd				0.324	1	0.569
- No	38	25	65.8			
- Yes	39	28	71.8			
Test of new animals before placement in the herd				1.090	1	0.297
Yes	14	8	57.1			
No	63	45	71.4			
Isolation pen for sick animals				0.667	1	0.414
- Yes	27	17	62.9			
- No	50	36	72.0			
visit of workers to Other farms				0.692	1	0.405
- No	30	19	63.3			
- Yes	47	34	72.3			
Awareness of farmer with FMD				.(a)		
Yes	77	53	68.8			
Disposal of carcasses				1.791	2	0.408
- burning in incinerator of the project - burning in incinerator	24	15	62.5			
- Burning out of the farm	22	14	63.6			
- Throw out farm	31	24	77.4			

Table 3.4: continued

Risk Factor	Total No. of tested samples	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Manure disposal				5.146	2	0.076
- sell to bricks makers weekly	10	4	40.0			
- store out farm till selling	2	2	100.0			
- store in the farm till selling	65	47	72.3			
Veterinary services				1.081	2	0.582
- daily supervision of a veterinarian	33	21	63.6			
- Veterinary, public and private hospital or clinic in the are	43	31	72.1			
- regular veterinary supervision	1	1	100.0			
Use of FMD vaccine to control the disease				0.837	1	0.360
- Yes	9	5	55.6			
- No	68	48	70.6			
Use of treatment to control FMD				2.089	3	0.554
- Antibiotics	19	14	73.7			
- pure Bee haney three days	3	1	33.3			
- *Acacia nilotica* and glycerin	31	22	70.9			
- No	24	16	66.7			

Table 3.4: continued

Risk Factor	Total No. of tested samples	No. (+ve)	% of (+ve)	χ^2	D.F.	P-Value
Other information				12.000	14	0.606
- CBPP, Tick infestation and Suspected with John's disease.	2	2	100.0			
- Brucellla, Tick infestation and calves diarrhea.	3	1	33.3			
- Brucellla, Tick infestation and mastitis	7	3	42.9			
- Theileriasis Tick infestation and House flies	11	7	63.6			
- Theileriasis and mastitis	4	4	100.0			
- CBPP, Tick infestation and Theileriasis	3	2	66.7			
- calves diarrhea, Tick infestation and theileriasis	4	4	100.0			
- Pneumonia, calves diarrhea and suspected with Brucella.	7	4	57.1			
- CBPP, calves diarrhea, Suspected with Brucella , Tick infestation	4	3	75.0			
- mastitis and calves diarrhea	2	2	100.0			
- calves diarrhea and Tick infestation	2	2	100.0			
- Tick infestation and Theileriasis.	19	12	63.2			
- mastitis and Lumpy skin disease	3	2	66.7			
- mastitis	1	1	100.0			
- Theileriasis, Tick infestation and mastitis	5	4	80.0			

(a) No stats coz variable is constant

3.3.36 Multivariate analysis was done by logistic regression model to assess significant risk factors associated with sero-positivity of the FMDV sero-type A in Khartoum state. (Exp(B)) express Odd Ratio (OR) (= the increased or decreased probability (OR $\neq 1$) of occurrence in comparison to the reference (OR = 1)., In the multivariate analysis confidence level was at 95% and P-value at ≤ 0.05. The result showed that one of the age categorry was found to be significant (P-value <0.05) so the age was significantly associated with FMD occurrence (Table 3.5).

Table 3.5: Multivariate analysis of risk factors with seropostivity in 77 tested cattle in Khartoum State, Sudan

Risk Factor	No. tested	No. (+ve) (%)	Exp(B)	P-Value	95% CI for Exp(B)	
					Lower	Upper
Locality				0.755		
- Jabal Elawliaa	10	4 (40.0)	ref			
- Umbadda	14	8 (57.1)	546395191.044	1.000	.000	-
- Karary	19	14 (73.7)	165507472.097	1.000	.000	-
- Bahry	17	13 (76.5)	1.787	.536	.284	11.226
- Sherg Elneel	16	13 (81.3)	.941	1.000	.000	-
- Khartoum	1	1 (100.0)	.385	.262	.072	2.045
Age				.065		
- <2 year (young)	9	3 (33.3)	ref			
- 2 – 4 year (medium age)	11	6 (54.5)	.099	.023	.013	.731
- > 4 year (old)	57	44 (77.2)	.451	.282	.106	1.926
Breed						
- cross	73	49 (67.1)	ref			
- local	4	4 (100.0)	231014417.815	.999	.000	-
Herd size						
- 50 - 150 cattle (medium herd)	50	31 (62.0)	ref			
- >150 cattle (big herd)	19	15 (78.9)	.000	.999	.000	
- <50 cattle (small herd)	8	7 (87.5)	.000	.999	.000	-
Artificial insemination						
- No	54	40 (74.1)	ref			
- Yes	23	13 (56.5)	791131072.693	.999	.000	-
Test of new animals before placement in the herd						
- Yes	14	8 (57.1)	ref			
- No	63	45 (71.4)	2.801	1.000	.000	-
Manure disposal						
- sell to bricks makers weekly	10	4 (40.0)	ref			
- store in the farm till selling	65	47 (72.3)	-	.999	-	
- store out farm till selling	2	2 (100.0)	241884573078598600.000	.999	.000	-

CHAPTER FOUR

4. Discussion

This study was carried out to estimate the sero-positivity of FMD sero type A in cattle by VNT and to investigate the potential risk factors associated with the occurrence of FMD in Khartoum state, Sudan. It was predicted using univariate analysis (χ^2) and multivariate analysis (logistic regression). Based on the results of this study the overall sero-prevalence of FMD sero- type A in Khartoum state was (68.8%), an indication of its importance in the study area. The sero-prevalence documented in this study showed lower value when compared to the previous reports of Habiela *et al*., (2010) which was 78.13% in cattle in Sudan. The reduction in prevalence could be due to the fact that some farms in Khartoum state applied vaccination as a measure of control. The high sero- prevalence of FMD in Khartoum in the present study (68.8%) might be due to lack of effective control policy for FMD. Furthermore, the high prevalence in our study confirmed that FMD is still endemic in Khartoum state.

In this study when we have investegated the risk factors associated with the occurrence of FMD by univariate analysis; the study revealed a significant variation (χ^2 = 8.19, p=0.017) on sero-positivity of foot and mouth disease among the three age groups, an increasing prevalence as the age increases. The higher sero-prevalence of FMD in (>4 year) cattle was (77.2 %) and in (2-4 year) cattle was (54.5 %) and in (<2 year) cattle was (33.3%). The finding observed in the current study is in agreement with the previous reports of Abunna *et al*. (2013); at Dire Dawa and its surroundings, eastern Ethiopia who claime there is a tendency of progressively increase in prevalence with advaning age (p= 0.006), the odds of animals in age band of 3 to 4 years and above 4 years of age was 3.46 and 2.43 times at more risk of infection than young animals (age group less than 3 years), also this study is in agreement with Chepkwony *et al*. (2012); in the Somali eco-system in Kenya who reported significant difference between ages (p<0.05) with the older animals showing a

higher risk of infection with FMD virus compared to younger animals, Jenbere *et al*., (2011); in selected district of Afar pastoral area, northeast Ethiopia who reported that age groups were found to be statistically significant (P= 0.007), Mohamoud *et al.* (2011); in Awbere and Babille districts of Jijiga zone, Somalia regional state, eastern Ethiopia reported that age groups; (< 2 years), young (2 to 4 years) and adults (> 4 years) of animals showed a significance variation ($\chi 2$ = 8.45, DF = 2, P = 0.01)., Mekonen *et al.* (2011); in Borana and Guji zones, southern Ethiopia who claime that significant difference was observed between different age groups; ($\chi 2$= 6.68, P<0.05) and Sarker *et al.* (2011); in Bangladesh who claime there was statistically significant different between the three age categories; <2 years, 2-4 years and >4 years. On the other hand Duguma *et al.* (2013); who had done their research in Bale Zone, Oromiya regional state, Ethiopia found that despite the prevalence was higher in adults than in youngs, with 22.31% and 18.64% respectively although there was no significant association between sero-positivity of FMD and age of cattle (P= 0.599), also Gelay *et al.* (2009); who has done their research in Bench Maji zone of southern Ethiopia said that there was no significant variation in seroprevalence among age which were grouped into three categories: calf (<1 year), young (1-3 years) and adult (>3 years) and Gebregziabher *et al.* (2013); in Dessie Zuria and Kombolcha area, suoth Wollo, Ethiopia who claime that seroprevalence recorded among age groups and in adult cattle was found to be statistically not significant (P= 0.8259, χ^2=0.05). The relatively low prevalence in young animal might indicate low frequency of exposure to risk factors, the low exposure in immature age groups was as a result of keeping young animals around homestead separately from the adult animals and the reasons of increased susceptibility to old cattle might be due to malnutrition, poor immunity, poor management system and stress of production.

In the univariate analysis a significant difference was observed in local breed (100%) and cross breed (67.1%). animals that showed a significance

variation among breed groups (χ^2 = 1.911 p=0.167) this finding is in agreement with the previous finding of Sarker *et al.* (2011); in Bangladesh who said breed significantly associated with the prevalence of FMD (P< 0.01) indigenous cattle were mostly affected by FMD compared to cross breed also, finding of this study was not in agreement with Gebregziabher *et al.* (2013); Ethiopia who said that seroprevalence recorded among local breed and cross breed was found to be statistically not significant (P= 0.728, χ^2=3.39) and with the previous finding of Duguma *et al.* (2013); in Ethiopia who said the difference in prevalence between local and cross breeds was statistically not significant (P= 0.599, χ^2=0.31). However, the higher prevalence in local breed might be due to uncontrolled movement unlike that of relatively controlled movement in cross breeds, sub optimum management practices implemented on indigenous cattle as they were supplemented with minimum inputs due to their low production and body weight gain, indigenous cattle were let to graze nearby therefore the higher level of incidence might be due to higher frequency of contact with infected animals of the nearby farmers which increase the degree of acquiring FMD. Moreover, the proportion of samples taken from local breeds might also contribute to this high prevalence.

The sero-prevalence rate of FMD in cattle serum samples collected from the six surveyed localities of Khartoum state of the Sudan was higher in Khartoum (100%) followed by Sherg Elneel (81.3%), Bahry (76.5%), Karary (73.2%), Umbadda (75.1%) and then Jabal Elawliaa (40%). there was significant difference in geographical locations in the univariate analysis, this finding is in agreement with the previous reports of Abunna *et al.* (2013); Ethiopia who said there was a statistically significant difference observed in the prevalence of FMD with the origin (p= 0.004), also the study is in agreement with Gelay *et al.* (2009); in Ethiopia who found difference was statistically significant (P<0.05; OR=4.95%; CI= 1.7823 - 8.9774), Jenbere *et al.*, (2011); northeast Ethiopia who claime that district were found to be statistically significant (P= 0.004, χ^2=13.4),

Mekonen et *al.* (2011); in southern Ethiopia who said geographical distribution was statistically significant (χ^2=104.26, P<0.05) but the study is not in agreement with Duguma *et al.* (2013); in Ethiopia who said that the difference in prevalence between studed sites was statistically non-significant (P= 0.066), also the study is not in agreement with the finding of Gebregziabher *et al.* (2013); in Ethiopia who claime that the difference in prevalence between locations was statistically not significant (P> 0.1234, χ^2=2.37), Lazarus *et al.* (2012), sero-epidemiology of foot-and-mouth disease in some border states of Nigeria said there is no difference in seropositivity between cattle sampled at different sites (p = 0.274), Mohamoud *et al.* (2011); Somalia regional state, eastern Ethiopia said that statistically no significant variation (P > 0.05) was observed in the prevalence of FMD in Districts. The variation of investigated areas could be a point of difference, considering the fact that each area has its specific and unique indigenous components and risk factors. Furthermore, divergent results could probably be explained by differences in the investigated various risk factors in each area, the variation in sero-prevalence could probably be attributed to the small sample size for estimating the sero-prevalence rate of FMD which did not reflect epidemiological status of the disease in the study area.

In this study herd size was categorized into <50 cattle (small) (87.5%), >150 cattle (big herd) (78.9%) and 50 - 150 cattle (medium herd) (62%) was statistically significant which was in agreement with Jenbere *et al.*, (2011); in northeast Ethiopia who said herd sizes statistically were found to be significant (P= 0.193, χ^2=3.294). Gelay *et al.*, (2009), southern Ethiopia said no significant difference was observed between small (4.85%) and medium (8.33%) herds; however, the differences observed between large (64.52%) and small (4.85%) and large (64.52%) and medium (8.33%) were statistically significant (P<0.05, OR=35) FMD prevalence tended to increase with herd size this might be attrebted to highly infectious nature of the disease and mode of transmission

which is enhanced by crowding and frequency of contact in addition takeoff from big herd size were in significant hence there was high chance of an individual animal to stay in the herd for life- long therefore, there was a high chance for the virus to circulate in the herd one entered into a large sized herd generally herd size increase the seroprevalence increases .

In this study there was significant difference in artificial insemination (P= 0.128, χ^2= 2.316), test of new animals before placement in the herd (P= 0.297, χ^2=1.090) and manure disposal (P= 0.076, χ^2=5.146).

The multivariate analysis, was done using logistic regression, with confidence interval of 95% and p-value of ≤0.05 was used to assess the association between identified significant risk factors in the univariate analysis and FMD occurrence. All potential risk factors (with p-value ≤0.3) thought to be important in the univariate analysis were entered into the multivariate analysis. This analysis showed no association between the six risk factors (locality, breed, herd size, artificial insemination, test of new animal before placement in the herd and manure disposal) and occurrence of FMD. The only significant risk factor in the multivariate analysis was age (p-value = 0.023). There was no significant difference between young and old cattle (p-value = 0.282); however, the differences observed between young and madiun age cattle (p-value = 0.023) and were statistically significant (P<0.05, OR=0.99), This finding is in agreement with the previous reports of Abunna *et al*. (2013); in eastern Ethiopia who reported that animal factors play significant role in the occurrence of the disease, also the finding in agreement with Jenbere *et al*., (2011); in northeast Ethiopia reported age groups were found to be statistically significant (P<0.05), Sarker *et al*., (2011); in Bangladesh found significant difference in different age groups (p<0.01) Which were analyzed in three categories; <2 years, 2-4 years and >4 years. On the contrary Duguma *et al*. (2013); in Ethiopia found that the prevalence was higher in adults than in young's although there was no significant association between sero-positivity of FMD and age of cattle (P=

0.599), also Gelay *et al.* (2009); southern Ethiopia found no significant difference in age categories. Logicaly; The relatively low prevalence in young animal might be indicative of low frequency of exposure to risk factors The low exposure in immature age groups was as a result of keeping young animals around homestead and around camps where there is less contact with other herds, the reasons of increased susceptibility to median aged cattle might be due to malnutrition, poor immunity, poor management system and stress of physiological condition which were known to affect their resistance to infection. In addition, the median age cattle herds follow seasonal patterns in search of good pasture and water and the herds are usually composed of adult males hence higher prevalence in median cattle is likely due to constant re-exposure to FMD, non-lactating and non-pregnant female cows and hence more exposed to FMD than younger age group. For the old cattle (>4 years) no significant difference was observed between it and young but there is significant difference between it and mediun age category and this might be attributed to old cattle composed of adult females lactating and pregnant cattle were being herded in homestead areas and hence having less chance of exposure median age animals may have acquired infection from multiple serotypes and / or infections also repeated exposure and close contact with other animal due to lack of control of animal movement

Conclusion:

Results of the present study have added to our knowledge on the epidemiology of FMD sero-type A in cattle in Khartoum state of the Sudan. They showed that the sero-prevalence rate was considerably high in the study area (68.8%). This fact justifies the need for attention and subsequent investigation for identification of the FMD virus circulating in the area, which helps in the implementation of effective control measures.

According to the study results in univariate and multi-variate analysis: age were found to be a significant risk factor for the occurrence of FMD (P-value ≤ 0.05).

Recommendations:

1. More studies on potential risk factors that enhance the spread and transmission of FMD in the Sudan, are needed.
2. Control strategy against this contagious and economically important disease based on vaccination against the four circulating serotpypes (A, O, SAT1 and SAT2) in the field should be carried out.
3. Improvement of management systems and tight biosecurity measures.

REFERENCES

Abuelzein, E. M. E. (1983). Foot and mouth disease in the Sudan, Rev. sci. Tech. Off. int. Epiz., 2 (1). Pp: 178-179

Abunna, F., Fikru, S. and Rufael, T. (2013). Sero-prevalence of Foot and Mouth Disease (FMD) at Dire Dawa and Its Surroundings, Eastern Ethiopia, IDOSI Publications, Global Veterinaria 11 (5), pp: 575 -576.

African Union Inter-African Bureau for Animal Resources (2010). Bulletin of Animal Health and Production in Africa, Volume 58 No 4, pp:309.

Aggarwal, N. and Barnett, P. V. (2002). Antigenic sites of foot-and-mouth disease virus (FMDV): an analysis of the specificities of anti-FMDV antibodies after vaccination of naturally susceptible host species, Journal of General Virology83, pp: 775.

Arnold, E. M., Paton, J. D., Ryan, E., Cox, J. S. and Wilesmith, W. J. (2007). Modelling studies to estimate the prevalence of foot-and-mouth disease carriers after reactive vaccination, Proceeding the Royal of Society Biological science, Downloaded from rspb.royalsocietypublishing.org, pp: 107.

Australian Government, 2007. Veterinary Journal Volume 85, No 8, August, Compiled by the Disease Preparedness Section Office of the Chief Veterinary Officer. Exotic Animal Diseases Bulletin Foot and Mouth Disease vaccine, pp:18, 20.

Ayelet, G., Mahapatra, M., Gelaye, E., Berhe, G. Egziabher, Rufeal, T., Sahle, M., Nigel, P. Ferris, Wadsworth, J., Geoffrey, H. Hutchings, and Knowles, N. J. (2009). Genetic Characterization of Foot-and-Mouth Disease Viruses, Ethiopia, 1981–2007. Emerging Infectious Diseases www.cdc.gov/eid Vol. 15, No. 9, pp: 1410.

Beyi, A., MSc thesis Wageningen University (2012). Costs and benefits of foot and mouth disease vaccination practices in commercial dairy farms in Central Ethiopia, pp: 6,8.

Bronsvoort, B. M. D., Toft, N., Bergmann, I. E., Sørensen, K. J., Anderson, J., Malirat, V., Tanya, N. V. and Morgan, K. (2006). Evaluation of three 3ABC ELISAs for foot-and-mouth disease non-structural antibodies using latent class analysis, BMC Veterinary Research, pp: 2.

Chepkwony, E. C., Gitao, C. G. and Muchemi, G. M. (2012). Seroprevalence of Foot and Mouth Disease in the Somali Eco-System in Kenya. International Journal of Animal and Veterinary Advances, 4(3), pp: 198, 200.

Crowther, J. R. (1986). Rev. sci. tech. Off. int. Epiz., 5 (2). Antigenic structure of foot-and-mouth disease virus, pp: 299, 301

Davies G. (2002), Foot and mouth disease, Research in Veterinary Science, 73, pp: 195–198.

Depa, P. M., Dimri, U., Sharma, M. C., Tiwari R. (2012). Update on epidemiology and control of Foot and Mouth Disease - A menace to international trade and global animal enterprise. Vet. World,Vol.5(11), pp: 694,696,698-700.

Duguma, M., Jibril,Y., Issa, A. and Hunde, A. (2013). Sero-Prevalence of Foot and Mouth Disease of Cattle in Bale Zone, Oromiya Regional State, Ethiopia, Global Veterinaria 11 (1), pp: 59, 61- 62.

FAO, EMPRES Emergancy prevention system **(2007).** Focus on Foot -and-Mouth disease situation world wide and major epidemiological events in 2005-2006, Issue No 1, pp:1

Gebregziabher, A. and Issa, A. (2013). Sero-prevalence of Foot and Mouth Disease in Dessie Zuria and Kombolcha Area, Soth Wollo. Ethiopia Veterinary Research, 6 (1), pp:1

Gelaye, E., Ayelet, G., Abera, T. and Asmare, K. (2009). Seroprevalence of foot and mouth disease in Bench Maji zone, Southwestern Ethiopia. Journal of Veterinary Medicine and Animal Health, pp: 005.

Habiela, M., Alamin, M. A. G., Abdelraouf Y. and Ali H.Y. H. (2010). Epizootiological study of foot and mouth disease in the Sudan: the situation after two decades. VETERINARSKI ARHIV 80 (1), 11-26, pp: 11-12, 20-21.

Haratian, K., Roustaei, M. H., Mahravani, H., Salehizadeh, M. and Akhavizadegan, M.A. (2000). Neutralizing Antibody Response in Cattle Following Vaccination against Foot-and-Mouth Disease Arch. Razi Ins, pp:130.

IOWA State University press (2014), Foot and Mouth Disease. the center for foot security and puplic health, Institute for international cooperatio in animal biologics, pp:2-6.

Jenbere, T. Sh., Etana, M., and Negussie, H. (2011). Study on the risk factors of Foot and Mouth disease in selected district of Afar pastoral area, northeast Ethiopia. Journal of Animal and Veterinary Advances,10(11), pp:1368.

Kasanga, C., Sallu, R., Ngeleja, C. M., Wadsworth, J., Ferris, N., Hutchings, G., Cleaveland, S., Lembo, T., Wambura, P., Yongolo ,M., Knowles, N., King, D. and Rweyemamu, M. (2012). Molecular Epidemiology of Foot-and-Mouth Disease Virus in Tanzania, GFRA Scientific Workshop, Hazyview, RSA: 17th April.

Klein, J., Hussain, M., Ahmad, M., Afzal, M. and Alexandersen, S. (2008). Epidemiology of foot-and-mouth disease in Landhi Dairy Colony, Pakistan, the world largest Buffalo colony.Virology Journal, Bio Med Central, pp: 2.

Lazarus, D. D., Schielen, W. J. G., Wungak, Y., Kwange, D.and Fasina, F. O. (2012), Sero-epidemiology of foot-and-mouth disease in some Border States of Nigeria. African Journal of Microbiology Research Vol. 6(8), pp. 1756 -1761.

Longjam, N., Deb, R., Sarmah, A. K., Tayo, T., Awachat, V. B., and Saxena V. K. (2011). A Brief Review on Diagnosis of Foot-and-Mouth Disease of Livestock: Conventional to Molecular Tools. Veterinary Medicine International Volume Article ID 905768, pp:1-4.

Mekonen, H., Beyene, D., Rufael, T., Feyis, A. and Abunna, F. (2011). Study on the prevalence of Foot and Mouth Disease in Borana and Guji Zones, Southern Ethiopia. Veterinary World, Vol.4(7), pp: 293.

Mitev, G., Tekerlekov, P. and Genchev, G. (1975). Passive immunity in calves born of cows that have recovered from foot-and-mouth disease or been vaccinated against it, vet Med Nauki;12(7): 3-7.

Mohamoud, A., Tessema, E. and Degefu, H. (2011). Seroprevalence of bovine foot and mouth disease (FMD) in Awbere and Babille districts of Jijiga zone, Somalia Regional State, Eastern Ethiopia. African Journal of Microbiology Research Vol. 5(21), pp: 3559.

Murphy, F. A., Gibbs, E. P. J., Horzinek, M. C. and Studdert, M. J. (1999) Foot and mouth disease in veterinary epidemiology 3rd edition California, Academic press USA, pp:38.

Mwiine, F. N., Ayebazibwe, Ch., Alexandersen, S., Olaho-Mukani,W., Ademun, A. R. O. and Tjornehoj, K. (2010). Seroepidemiological investigation of foot-and-mouth disease virus serotypes in cattle around Lake Mburo National Park in South-Western Uganda. Journal of Veterinary Medicine and Animal Health Vol. 2(4), pp: 46.

Namatovu, A., Wekesa, S. N., Tjørnehøj, K., Dhikusooka, M. T., Muwanika, V. B., Siegsmund, H. R. and Ayebazibwe Ch. (2013). Laboratory capacity for diagnosis of foot-and-mouth disease in Eastern Africa: implications for the progressive control pathway, http://www.biomedcentral.com/1746-6148/9/19**.**

OIE, Terrestrial Manual (2012), Foot and Mouth Disease, chapter 2.1.5. Version adopted by the World Assembly of Delegates of the OIE, pp: 1-2,5-6,9-10,22.

Piccone, M. E., Feng,,Y., Chang, A. C. Y., Mosseri, R., Lu, Q ., Kutish, G. F., Lu, Zh., Burrage, Th. G., Gooch, Ch., Rock, D. L. and Cohen S. N. (2009). Identification of Cellular Genes Affecting the Infectivity of Foot-and-Mouth Disease Virus. Journal of virology, pp: 6681.

Royal veterinary Collage (RVC), FAO and OIE. The impact of foot and mouth disease, pp:15.

Sahle, M. (2004). an epidemiological study on the genetic relationships of foot and mouth disease viruses in east Afric, a thesis submitted in partial fulfilment of the requirements for the degree of Doctor of Philosophy in the Department of Veterinary Tropical Diseases, Faculty of Veterinary Science, University of Pretoria, South Africa, pp:3-8, 22.

Sarker, S., Talukder, S., Haque, H. M., Islam, H. M. and Gupta, D. S. (2011). Epidemiological study on Foot-and-mouth disease in cattle: Prevalence and risk factor assessment in Rajshahi, Bangladesh. Wayamba Journal of Animl science, pp:71.

Senawi, J. (2012). Epidemiology of Foot and Mouth Disease in Cattle in Pahang, Malaysia, research Masters with Training (RMT), School of Veterinary and Biomedical Sciences, Faculty of Health Sciences, Murdoch University, pp:13-14,16, 19-22, 24-25.

Thai AgriculturalSTandard TAS 10400 (2004). Diagnostic test of Foot and mouth disease, National Bureau of Agricultural Commodity and Food Standards Ministry of Agriculture and Cooperatives, pp1-3.

Thrusfield, M., 2005, 2007 veterinary Epidimiology Third edition, by Blackwell Science Ltd, a Blackwell Publishing company, pp: 248.

Tunca, R., Sozmen, M., Erdogan, H., Citil, M., Uzlu, E., Ozen, H. and Gokçe, E. (2008). Determination of cardiac troponin I in the blood and heart of calves with Foot-and-Mouth Disease, Journal of Veterinary Diagnostic Investigation, pp:598, 600- 601.

US AID, from the American people **(2007-2008).** Foot and Mouth Disease surveillance and Sero-epidemiological Situation in Ethiopia, pp:3.

Vosloo, W., Foot-and-mouth disease in Africa, ARC-Onderstepoort Veterinary Institute, South Africa, sourced from http://www.wrlfmd.org/ Published in the Royal Gazette Vol.121 Section 120D.

APPENDIX 1

Frequency table for the distribution of 77 cattle examined for FMD sero-type A according to potential risk factors in Khartoum state, Sudan:

Appendix 1.1: locality

		Frequency	Percent	Cumulative Percent
Valid	- Khartoum	1	1.3	1.3
	- Jabal Elawliaa	10	13.0	14.3
	- Sherg Elneel	16	20.8	35.1
	- Bahry	17	22.1	57.1
	- Umbadda	14	18.2	75.3
	- Karary	19	24.7	100.0
	Total	**77**	**100.0**	

Appendix 1.2: location

		Frequency	Percent	Cumulative Percent
Valid	- Elgeraf	1	1.3	1.3
	- Sundus Project	3	3.9	5.2
	- UmAraha	7	9.1	14.3
	- Hilat Koko mahleb area	2	2.6	16.9
	- Elkeryab	3	3.9	20.8
	- Elsillat South	7	9.1	29.9
	- Elshigla	4	5.2	35.1
	- Eltibna	11	14.3	49.4
	- Abo Halima	2	2.6	51.9
	- Elsillat North	4	5.2	57.1
	- Elrodoan Project	14	18.2	75.3
	- Elahamda Project	19	24.7	100.0
	Total	**77**	**100.0**	

Appendix 1.3: Sex

		Frequency	Percent	Cumulative Percent
Valid	- Female	72	93.5	93.5
	- Male	5	6.5	100.0
	Total	**77**	**100.0**	

Appendix 1.4: Age

		Frequency	Percent	Cumulative Percent
Valid	- <2 year young	9	11.7	11.7
	- 2-4 year medium	11	14.3	26.0
	- >4 year old	57	74.0	100.0
	Total	**77**	**100.0**	

Appendix 1.5: Breed

		Frequency	Percent	Cumulative Percent
Valid	- Local	4	5.2	5.2
	- Cross	73	94.8	100.0
	Total	**77**	**100.0**	

Appendix 1. 6: Body condition

		Frequency	Percent	Cumulative Percent
Valid	- Good	71	92.2	92.2
	- Emaciated	6	7.8	100.0
	Total	**77**	**100.0**	

Appendix 1.7: Herd size

		Frequency	Percent	Cumulative Percent
Valid	- <50 cattle (small herd)	8	10.4	10.4
	- 50- 150 cattle (medium herd)	50	64.9	75.3
	- >150 cattle (big herd)	19	24.7	100.0
	Total	**77**	**100.0**	

Appendix 1.8: Production purpose

		Frequency	Percent	Cumulative Percent
Valid	- Milk	74	96.1	96.1
	- Milk and meat	3	3.9	100.0
	Total	**77**	**100.0**	

Appendix 1. 9: Herd type (mixed with other species)

		Frequency	Percent	Cumulative Percent
Valid	- No	27	35.1	35.1
	- Yes	50	64.9	100.0
	Total	**77**	**100.0**	

Appendix 1.10: Mixed specie

		Frequency	Percent	Cumulative Percent
Valid	- Not mixed	27	35.1	53.1
	- Goat	7	9.1	44.2
	- Sheep	3	3.9	48.1
	- Sheep and Goat	40	51.9	100.0
	Total	**77**	**100.0**	

Appendix 1.11: History of FMD in the herd

		Frequency	Percent	Cumulative Percent
Valid	- No	20	26.0	26.0
	- Yes	57	74.0	100.0
	Total	**77**	**100.0**	

Appendix 1.12: Other diseases

		Frequency	Percent	Cumulative Percent
Valid	- No others	54	70.1	70.1
	- Theileriasis	14	18.2	88.3
	- Abortion	7	9.1	97.4
	- Suspected with Johns disease	2	2.6	100.0
	Total	**77**	**100.0**	

Appendix 1.13: Production system

		Frequency	Percent	Cumulative Percent
Valid	- Resident	59	76.6	76.6
	- Transhumant	18	23.4	100.0
	Total	**77**	**100.0**	

Appendix 1.14: Farm construction

		Frequency	Percent	Cumulative Percent
Valid	- Closed sheds of mud with roof of mats	32	41.6	41.6
	- Brick stables and zinc roof	15	19.5	61.0
	- Pens of iron with roof of iron and mats	26	33.8	94.8
	- Pens of zinc and with roof of iron and mats	1	1.3	96.1
	- Pens of firewood	3	3.9	100.0
	Total	**77**	**100.0**	

Appendix 1.15: Ventilation

		Frequency	Percent	Cumulative Percent
Valid	Good	77	100.0	100.0
	Bad	0	0	100.0
	Total	**77**	**100.0**	

Appendix 1.16: Grazing system

		Frequency	Percent	Cumulative Percent
Valid	- Private	73	94.8	94.8
	- Private and common	4	5.2	100.0
	Total	**77**	**100.0**	

Appendix 1.17: Water resources

		Frequency	Percent	Cumulative Percent
Valid	- Private tap water in the area (project)	8	10.4	10.4
	- Private tap water	13	16.9	27.3
	- Private	8	10.4	37.7
	- Private tap water and common from Nile canal in the area	3	3.9	41.6
	- Private tap water from well in the area	45	58.4	100.0
	Total	**77**	**100.0**	

Appendix 1.18: Use of concentrate

		Frequency	Percent	Cumulative Percent
Valid	Yes	77	100.0	100.0
	No	0	0	100.0
	Total	**77**	**100.0**	

Appendix 1.19: Artificial insemination

		Frequency	Percent	Cumulative Percent
Valid	- No	54	70.1	70.1
	- Yes	23	29.9	100.0
	Total	**77**	**100.0**	

Appendix 1.20: Where stay in dry season

		Frequency	Percent	Cumulative Percent
Valid	- In the farm	77	100.0	100.0
	- Out of the farm	0	0	100.0
	Total	**77**	**100.0**	

Appendix 1.21: Where stay in rainy season

		Frequency	Percent	Cumulative Percent
Valid	- In the farm	58	75.3	75.3
	- Out of the farm	19	24.7	100.0
	Total	**77**	**100.0**	

Appendix 1. 22: Contact with neighboring herds

		Frequency	Percent	Cumulative Percent
Valid	- No	38	49.4	49.4
	- Yes	39	50.6	100.0
	Total	**77**	**100.0**	

Appendix 1.23: Test of new animals before placement in the herd

		Frequency	Percent	Cumulative Percent
Valid	- Yes	14	18.2	18.2
	- No	63	81.8	100.0
	Total	**77**	**100.0**	

Appendix 1. 24: Isolation pen for sick animals

		Frequency	Percent	Cumulative Percent
Valid	- Yes	27	35.1	35.1
	- No	50	64.9	100.0
	Total	**77**	**100.0**	

Appendix 1. 25: Visit of workers to other farms

		Frequency	Percent	Cumulative Percent
Valid	- No	30	39.0	39.0
	- Yes	47	61.0	100.0
	Total	**77**	**100.0**	

Appendix1.26: Awareness of farmer with FMD

		Frequency	Percent	Cumulative Percent
Valid	Yes	77	100.0	100.0
	No	0	0	100.0
	Total	**77**	**100.0**	

Appendix 1.27: Disposal of carcasses

		Frequency	Percent	Cumulative Percent
Valid	- Burning in incinerator of the project	24	31.2	31.2
	- Burning out of the farm	22	28.6	59.7
	- Throw out farm	31	40.3	100.0
	Total	**77**	**100.0**	

Appendix 1.28: Manure Disposal

		Frequency	Percent	Cumulative Percent
Valid	- Sell to bricks makers weekly	10	13.0	13.0
	- Store out farm till selling	2	2.6	15.6
	- Store in the farm till selling	65	84.4	100.0
	Total	**77**	**100.0**	

Appendix 1.29: Veterinary services

		Frequency	Percent	Cumulative Percent
Valid	- Daily supervision of a veterinarian	33	42.9	42.9
	- Veterinary public and private hospital or clinic in the area	43	55.8	98.7
	- Regular veterinary supervision	1	13	100.0
	Total	**77**	**100.0**	

Appendix 1.30: Use of FMD vaccine to control the disease

		Frequency	Percent	Cumulative Percent
Valid	- Yes	9	11.7	11.7
	- No	68	88.3	100.0
	Total	**77**	**100.0**	

Appendix 1.31: Use of treatment to control FMD

		Frequency	Percent	Cumulative Percent
Valid	- Antibiotics	19	24.7	24.7
	- Pure bee honey three days	3	3.9	28.6
	- Acacia nilotica and glycerin	31	40.3	68.8
	- No	24	31.2	100.0
	Total	**77**	**100.0**	

Appendix 1.32: Other information

		Frequency	Percent	Cumulative Percent
Valid	- CBPP, Tick infestation and John's disease suspicious.	2	2.6	2.6
	- Brucellla, Tick infestation and calves diarrhea.	3	3.9	6.5
	- Brucellla, Tick infestation and mastitis	7	9.1	15.6
	- Theileriasis Tick infestation and House flies	11	14.3	29.9
	- Theileriasis and mastitis	4	5.2	35.1
	- CBPP, Tick infestation and Theileriasis	3	3.9	39.0
	- calves diarrhea, Tick infestation and theileriasts	4	5.2	44.2
	- Pneumonia, calves diarrhea and Brucella suspicious	7	9.1	53.2
	- CBPP, calves diarrhea, Brucella suspicious,Tick infestation	4	5.2	58.4
	- mastitis and calves diarrhea	2	2.6	61.0
	- calves diarrhea and Tick infestation	2	2.6	63.6
	- Tick infestation and Theileriasis	19	24.7	88.3
	- mastitis and lumpy skin disease	3	3.9	92.2
	- mastitis	1	1.3	93.5
	- Theileriasis tick infestation and mastitis	5	6.5	
	Total	**77**	**100.0**	

APPENDIX 2

Cross tabulation for the distribution of FMD sero-type A in cattle examined in Khartoum state with potential risk factors:

Appendix 2.1: Result of distribution of prevalence of FMD in locality

Locality * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Locality	Khartoum	Count	0	1	1
		% of locality	.0%	100.0%	100.0%
		% of result	.0%	1.9%	1.3%
		% of total	.0%	1.3%	1.3%
	Jabal Elawliaa	Count	6	4	10
		% of locality	60.0%	40.0%	100.0%
		% of result	25.0%	7.5%	13.0%
		% of total	7.8%	5.2%	13.0%
	Sherg Elneel	Count	3	13	16
		% of locality	18.8%	81.3%	100.0%
		% of result	12.5%	24.5%	20.8%
		% of total	3.9%	16.9%	20.8%
	Bahry	Count	4	13	17
		% of locality	23.5%	76.5%	100.0%
		% of result	16.7%	24.5%	22.1%
		% of total	5.2%	16.9%	22.1%
	Umbadda	Count	6	8	14
		% of locality	42.9%	57.1%	100.0%
		% of result	25.0%	15.1%	18.2%
		% of total	7.8%	10.4%	18.2%
	Karary	Count	5	14	19
		% of locality	26.3%	73.7%	100.0%
		% of result	20.8%	26.4%	24.7%
		% of total	6.5%	18.2%	24.7%
Total		Count	24	53	77
		% of locality	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.2: Result of distribution of prevalence of FMD in location

Location * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Location	Elgeraf	Count	0	1	1
		% of location	.0%	100.0%	100.0%
		% of result	.0%	1.9%	1.3%
		% of total	.0%	1.3%	1.3%
	Sundus Project	Count	2	1	3
		% of location	66.7%	33.3%	100.0%
		% of result	8.3%	1.9%	3.9%
		% of total	2.6%	1.3%	3.9%
	Um Ardha	Count	4	3	7
		% of location	57.1%	42.9%	100.0%
		% of result	16.7%	5.7%	9.1%
		% of total	5.2%	3.9%	9.1%
	Hilat koko Mahleb area	Count	0	2	2
		% of location	.0%	100.0%	100.0%
		% of resut	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	Elkeryab	Count	1	2	3
		% of location	33.3%	66.7%	100.0%
		% of result	4.2%	3.8%	3.9%
		% of total	1.3%	2.6%	3.9%
	Elsillat South	Count	1	6	7
		% of location	14.3%	85.7%	100.0%
		% of result	4.2%	11.3%	9.1%
		% of total	1.3%	7.8%	9.1%
	Elshigla	Count	1	3	4
		% of location	25.0%	75.0%	100.0%
		% of result	4.2%	5.7%	5.2%
		% of total	1.3%	3.9%	5.2%
	Eltibna	Count	4	7	11
		% of location	36.4%	63.6%	100.0%
		% of result	16.7%	13.2%	14.3%
		% of total	5.2%	9.1%	14.3%
	Abo Halima	Count	0	2	2
		% of location	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	Elsillat North	Count	0	4	4
		% of location	.0%	100.0%	100.0%
		% of result	.0%	7.5%	5.2%
		% of total	.0%	5.2%	5.2%
	Elrodoan project	Count	6	8	14
		% of location	42.9%	57.1%	100.0%
		% of result	25.0%	15.1%	18.2%
		% of total	7.8%	10.4%	18.2%

Appendix 2.2: continued

			Result of VNT		
			Negative	positive	Total
Location	Elahamda projec	Count	5	14	19
		% of location	26.3%	73.7%	100.0%
		% of result	20.8%	26.4%	24.7%
		% of total	6.5%	18.2%	24.7%
Total		Count	24	53	77
		% of location	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.3: Result of distribution of prevalence of FMD according to sex

Sex* Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Sex	Female	Count	22	50	72
		% of sex	30.6%	69.4%	100.0%
		% of result	91.7%	94.3%	93.5%
		% of total	28.6%	64.9%	93.5%
	male	Count	2	3	5
		% of sex	40.0%	60.0%	100.0%
		% of result	8.3%	5.7%	6.5%
		% of total	2.6%	3.9%	6.5%
Total		Count	24	53	77
		% of sex	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.4: Result of distribution of prevalence of FMD according to age

Age * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Age	<2Year young	Count	6	3	9
		% of age	66.7%	33.3%	100.0%
		% of result	25.0%	5.7%	11.7%
		% of total	7.8%	3.9%	11.7%
	2 - 4 Year (medium age)	Count	5	6	11
		% of age	45.5%	54.5%	100.0%
		% of result	20.8%	11.3%	14.3%
		% of total	6.5%	7.8%	14.3%
	>4 Year (old)	Count	13	44	57
		% of age	22.8%	77.2%	100.0%
		% of result	54.2%	83.0%	74.0%
		% of total	16.9%	57.1%	74.0%
Total		Count	24	53	77
		% of age	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.5: Result of distribution of prevalence of FMD according to breed

Breed * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Breed	local	Count	0	4	4
		% of breed	.0%	100.0%	100.0%
		% of result	.0%	7.5%	5.2%
		% of total	.0%	5.2%	5.2%
	cross	Count	24	49	73
		% of breed	32.9%	67.1%	100.0%
		% of result	100.0%	92.5%	94.8%
		% of total	31.2%	63.6%	94.8%
Total		Count	24	53	77
		% of breed	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.6: Result of distribution of prevalence of FMD according to body condition

Body condition * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Body conditi	good body condition	Count	22	49	71
		% of body condition	31.0%	69.0%	100.0%
		% of result	91.7%	92.5%	92.2%
		% of total	28.6%	63.6%	92.2%
	emaciated	Count	2	4	6
		% of body condition	33.3%	66.7%	100.0%
		% of result	8.3%	7.5%	7.8%
		% of total	2.6%	5.2%	7.8%
Total		Count	24	53	77
		% of body condition	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.7 Results of distribution of prevalence of FMD according to herd size

Herd size * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Herd size	<50 cattle (small herd)	Count	1	7	8
		% of herd size	12.5%	87.5%	100.0%
		% of result	4.2%	13.2%	10.4%
		% of total	1.3%	9.1%	10.4%
	50 - 150 (medium herd)	Count	19	31	50
		% of herd size	38.0%	62.0%	100.0%
		% of result	79.2%	58.5%	64.9%
		% of total	24.7%	40.3%	64.9%
	>150 (big herd)	Count	4	15	19
		% of herd size	21.1%	78.9%	100.0%
		% of result	16.7%	28.3%	24.7%
		% of total	5.2%	19.5%	24.7%
Total		Count	24	53	77
		% of herd size	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.8: Result of distribution of prevalence of FMD according to production purpose

Production purpose * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Production purpose	milk	Count	22	52	74
		% of production purpose	29.7%	70.3%	100.0%
		% of result	91.7%	98.1%	96.1%
		% of total	28.6%	67.5%	96.1%
	milk and meat	Count	2	1	3
		% of production purpose	66.7%	33.3%	100.0%
		% of result	8.3%	1.9%	3.9%
		% of total	2.6%	1.3%	3.9%
Total		Count	24	53	77
		% of production purpose	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.9: Result of distribution of prevalence of FMD according to Herd type mixed with other species

Herd type mixed with other species * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Herd type mixed with other species	No	Count	7	20	27
		% of herd type mixed with other species	25.9%	74.1%	100.0%
		% of result	29.2%	37.7%	35.1%
		% of total	9.1%	26.0%	35.1%
	Yes	Count	17	33	50
		% of herd type mixed with other species	34.0%	66.0%	100.0%
		% of result	70.8%	62.3%	64.9%
		% of total	22.1%	42.9%	64.9%
Total		Count	24	53	77
		% of herd type mixed with other species	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.10: Result of distribution of prevalence of FMD according to mixed species

Mixed species * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Mixed species	Not mixed	Count	7	20	27
		% of mixed species	25.9%	74.1%	100.0%
		% of result	29.2%	37.7%	35.1%
		% of total	9.1%	26.0%	35.1%
	Goat	Count	4	3	7
		% of mixed species	57.1%	42.9%	100.0%
		% of result T	16.7%	5.7%	9.1%
		% of total	5.2%	3.9%	9.1%
	Sheep	Count	1	2	3
		% of mixed species	33.3%	66.7%	100.0%
		% of result	4.2%	3.8%	3.9%
		% of total	1.3%	2.6%	3.9%
	Sheep and Goat	Count	12	28	40
		% of mixed species	30.0%	70.0%	100.0%
		% of result	50.0%	52.8%	51.9%
		% of total	15.6%	36.4%	51.9%
Total		Count	24	53	77
		% of mixed species	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.11: Result of distribution of prevalence of FMD according to previous history of FMD in the herd

Previous history of FMD in the herd * Result of (VNT)

			Result of VNT		
			Negative	positive	Total
Previous history of FMD in the herd	No	Count	8	12	20
		% of previous history of FMD in the herd	40.0%	60.0%	100.0%
		% of result	33.3%	22.6%	26.0%
		% of total	10.4%	15.6%	26.0%
	Yes	Count	16	41	57
		% of previous history of FMD in the herd	28.1%	71.9%	100.0%
		% of result	66.7%	77.4%	74.0%
		% of total	20.8%	53.2%	74.0%
Total		Count	24	53	77
		% of previous history of FMD in the herd	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.12: Result of prevalence of FMD according to other diseases

Other diseases * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Other diseases	No others	Count	16	38	54
		% of other diseases	29.6%	70.4%	100.0%
		% of result	66.7%	71.7%	70.1%
		% of total	20.8%	49.4%	70.1%
	Theileriasis	Count	5	9	14
		% of other diseases	35.7%	64.3%	100.0%
		% of result	20.8%	17.0%	18.2%
		% of total	6.5%	11.7%	18.2%
	Abortion	Count	3	4	7
		% of other diseases	42.9%	57.1%	100.0%
		% of result	12.5%	7.5%	9.1%
		% of total	3.9%	5.2%	9.1%
	Suspected withJohn's diseas	Count	0	2	2
		% of other diseases	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
Total		Count	24	53	77
		% of other diseases	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.13: Result of distribution of prevalence of FMD according to production system

Production system * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Production System	Resident	Count	19	40	59
		% of production System	32.2%	67.8%	100.0%
		% of result	79.2%	75.5%	76.6%
		% of total	24.7%	51.9%	76.6%
	Transhumant	Count	5	13	18
		% of production System	27.8%	72.2%	100.0%
		% of result	20.8%	24.5%	23.4%
		% of total	6.5%	16.9%	23.4%
Total		Count	24	53	77
		% of production System	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.14: Result of distribution of prevalence of FMD according to farm construction

Farm construction * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Farm construction	Closed sheds of mud with roof of mats	Count	10	22	32
		% of farm construction	31.3%	68.8%	100.0%
		% of result	41.7%	41.5%	41.6%
		% of total	13.0%	28.6%	41.6%
	Brick stables and zinc roof	Count	4	11	15
		% of farm construction	26.7%	73.3%	100.0%
		% of result	16.7%	20.8%	19.5%
		% of total	5.2%	14.3%	19.5%
	pens of iron with roof of iron and mats	Count	9	17	26
		% of farm construction	34.6%	65.4%	100.0%
		% of result	37.5%	32.1%	33.8%
		% of total	11.7%	22.1%	33.8%
	pens of zinc with roof of iron and mats	Count	0	1	1
		% of farm construction	.0%	100.0%	100.0%
		% of result	.0%	1.9%	1.3%
		% of total	.0%	1.3%	1.3%
	pens of fire wood	Count	1	2	3
		% of farm construction	33.3%	66.7%	100.0%
		% of result	4.2%	3.8%	3.9%
		% of total	1.3%	2.6%	3.9%
Total		Count	24	53	77
		% of farm construction	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.15: Result of distribution of prevalence of FMD according to Ventilation

Ventilation * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Ventilation	good	Count	24	53	77
		% of ventilation	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%
Total		Count	24	53	77
		% of ventilation	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.16: Result of distribution of prevalence of FMD according to grazing system

Grazing system * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Grazing system	private	Count	22	51	73
		% of grazing system	30.1%	69.9%	100.0%
		% of result	91.7%	96.2%	94.8%
		% of total	28.6%	66.2%	94.8%
	private and common	Count	2	2	4
		% of grazing system	50.0%	50.0%	100.0%
		% of result	8.3%	3.8%	5.2%
		% of total	2.6%	2.6%	5.2%
Total		Count	24	53	77
		% of grazing system	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.17: Result of distribution of prevalence of FMD according to water resources

Water resources * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Water resources	Private tap water from well in the area (project)	Count	2	6	8
		% of water resources	25.0%	75.0%	100.0%
		% of result	8.3%	11.3%	10.4%
		% of total	2.6%	7.8%	10.4%
	Private tap water	Count	5	8	13
		% of water resources	38.5%	61.5%	100.0%
		% of result	20.8%	15.1%	16.9%
		% of total	6.5%	10.4%	16.9%
	Private	Count	2	6	8
		% of water resources	25.0%	75.0%	100.0%
		% of result	8.3%	11.3%	10.4%
		% of total	2.6%	7.8%	10.4%
	Private tap water and common from Nile canal in the area	Count	2	1	3
		% of water resources	66.7%	33.3%	100.0%
		% of result	8.3%	1.9%	3.9%
		% of total	2.6%	1.3%	3.9%
	Private tap water from well in the area	Count	13	32	45
		% of water resources	28.9%	71.1%	100.0%
		% of result	54.2%	60.4%	58.4%
		% of total	16.9%	41.6%	58.4%
Total		Count	24	53	77
		% of water resources	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.18: Result of distribution of prevalence of FMD according to use of concentrates

Use of concentrates * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Use of concentrates	Yes	Count	24	53	77
		% of use of concentrates	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%
Total		Count	24	53	77
		% of use of concentrates	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.19: Result of distribution of prevalence of FMD according to artificial insemination

Artificial insemination * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Artificial insemination	No	Count	14	40	54
		% of artificial insemination	25.9%	74.1%	100.0%
		% of result	58.3%	75.5%	70.1%
		% of total	18.2%	51.9%	70.1%
	Yes	Count	10	13	23
		% of artificial insemination	43.5%	56.5%	100.0%
		% of result	41.7%	24.5%	29.9%
		% of total	13.0%	16.9%	29.9%
Total		Count	24	53	77
		% of artificial insemination	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.20: Result of distribution of prevalence of FMD according to where stay in dry season

Where stay in dry season * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Where stay in dry season	in the farm	Count	24	53	77
		% of where stay in dry season	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%
Total		Count	24	53	77
		% of where stay in dry season	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.21: Result of distribution of prevalence of FMD according to where stay in rainy season

Where stay in rainy season * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Where stay in rainy season	in the farm	Count	19	39	58
		% of where stay in rainy season	32.8%	67.2%	100.0%
		% of result	79.2%	73.6%	75.3%
		% of total	24.7%	50.6%	75.3%
	out of the farm	Count	5	14	19
		% of where stay in rainy season	26.3%	73.7%	100.0%
		% of result	20.8%	26.4%	24.7%
		% of total	6.5%	18.2%	24.7%
Total		Count	24	53	77
		% of where stay in rainy season	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.22: Result of distribution of prevalence of FMD according to contact with neighboring herd

Contact with neighboring herd * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Contact with neighboring herd	No	Count	13	25	38
		% of contact with neighboring herd	34.2%	65.8%	100.0%
		% of result	54.2%	47.2%	49.4%
		% of total	16.9%	32.5%	49.4%
	Yes	Count	11	28	39
		% of contact with neighboring herd	28.2%	71.8%	100.0%
		% of result	45.8%	52.8%	50.6%
		% of total	14.3%	36.4%	50.6%
Total		Count	24	53	77
		% of contact with neighboring herd	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.23: Result of distribution of prevalence of FMD according to test of new animals before placement in the herd

Test of new animals before placement in the herd * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Test of new animals before placement in the herd	Yes	Count	6	8	14
		% of test of new animals before placement in the herd	42.9%	57.1%	100.0%
		% of result	25.0%	15.1%	18.2%
		% of total	7.8%	10.4%	18.2%
	No	Count	18	45	63
		% of test of new animals before placement in the herd	28.6%	71.4%	100.0%
		% of result	75.0%	84.9%	81.8%
		% of total	23.4%	58.4%	81.8%
Total		Count	24	53	77
		% of test of new animals before placement in the herd	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.24: Result of distribution of prevalence of FMD according to isolation pen for sick animals

Isolation pen for sick animals * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Isolation pen for sick animals	Yes	Count	10	17	27
		% of isolation pen for sick animals	37.0%	63.0%	100.0%
		% of result	41.7%	32.1%	35.1%
		% of total	13.0%	22.1%	35.1%
	No	Count	14	36	50
		% of isolation pen for sick animals	28.0%	72.0%	100.0%
		% of result	58.3%	67.9%	64.9%
		% of total	18.2%	46.8%	64.9%
Total		Count	24	53	77
		% of isolation pen for sick animals	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.25: Result of distribution of prevalence of FMD according to visit of workers to other farms

Visit of workers to other farms * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
visit of workers to other farms	No	Count	11	19	30
		% of visit of workers to other farms	36.7%	63.3%	100.0%
		% of result	45.8%	35.8%	39.0%
		% of total	14.3%	24.7%	39.0%
	Yes	Count	13	34	47
		% of visit of workers to other farms	27.7%	72.3%	100.0%
		% of result	54.2%	64.2%	61.0%
		% of total	16.9%	44.2%	61.0%
Total		Count	24	53	77
		% of visit of workers to other farms	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.26: Result of distribution of prevalence of FMD according to awareness of farmer with FMD

Awareness of farmer with FMD * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Awareness of farmer with FMD	Yes	Count	24	53	77
		% of awareness of farmer with FMD	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%
Total		Count	24	53	77
		% of awareness of farmer with FMD	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.27: Result of distribution of prevalence of FMD according to disposal of carcasses

Disposal of carcasses * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Disposal of carcasses	burning in incinerator of the project - burning in incinerator	Count	9	15	24
		% of disposal of carcasses	37.5%	62.5%	100.0%
		% of result	37.5%	28.3%	31.2%
		% of total	11.7%	19.5%	31.2%
	Burning out of the farm	Count	8	14	22
		% of disposal of carcasses	36.4%	63.6%	100.0%
		% of result	33.3%	26.4%	28.6%
		% of total	10.4%	18.2%	28.6%
	Throw out farm	Count	7	24	31
		% of disposal of carcasses	22.6%	77.4%	100.0%
		% of result	29.2%	45.3%	40.3%
		% of total	9.1%	31.2%	40.3%
Total		Count	24	53	77
		% of disposal of carcasses	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.28: Result of distribution of prevalence of FMD according to manure disposal

Manure disposal * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Manure disposal	sell to bricks makers weekly	Count	6	4	10
		% of manure disposal	60.0%	40.0%	100.0%
		% of result	25.0%	7.5%	13.0%
		% of total	7.8%	5.2%	13.0%
	store out farm till selling	Count	0	2	2
		% of manure disposal	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	store in the farm till selling	Count	18	47	65
		% of manure disposal	27.7%	72.3%	100.0%
		% of result	75.0%	88.7%	84.4%
		% of total	23.4%	61.0%	84.4%
Total		Count	24	53	77
		% of manure disposal	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.29: Result of distribution of prevalence of FMD according to veterinary services

Veterinary services * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Veterinary services	daily supervision of a veterinarian	Count	12	21	33
		% of veterinary services	36.4%	63.6%	100.0%
		% of result	50.0%	39.6%	42.9%
		% of total	15.6%	27.3%	42.9%
	Veterinary, public and private hospital or clinic in the area	Count	12	31	43
		% of veterinary services	27.9%	72.1%	100.0%
		% of result	50.0%	58.5%	55.8%
		% of total	15.6%	40.3%	55.8%
	regular veterinary supervision	Count	0	1	1
		% of veterinary services	.0%	100.0%	100.0%
		% of result	.0%	1.9%	1.3%
		% of total	.0%	1.3%	1.3%
Total		Count	24	53	77
		% of veterinary services	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.30: Result of distribution of prevalence of FMD according to using of FMD vaccine to control the disease

Use of FMD vaccine to control the disease * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
using of FMD vaccine to control the disease	Yes	Count	4	5	9
		% of use of FMD vaccine to control the disease	44.4%	55.6%	100.0%
		% of Result	16.7%	9.4%	11.7%
		% of Total	5.2%	6.5%	11.7%
	No	Count	20	48	68
		% of use of FMD vaccine to control the disease	29.4%	70.6%	100.0%
		% of Result	83.3%	90.6%	88.3%
		% of Total	26.0%	62.3%	88.3%
Total		Count	24	53	77
		% of use of FMD vaccine to control the disease	31.2%	68.8%	100.0%
		% of Result	100.0%	100.0%	100.0%
		% of Total	31.2%	68.8%	100.0%

Appendix 2.31: Result of distribution of prevalence of FMD according to use of treatment to control FMD

Use of treatment to control FMD * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Use of treatment to control FMD	Antibiotics	Count	5	14	19
		% of use of treatment to control FMD	26.3%	73.7%	100.0%
		% of result	20.8%	26.4%	24.7%
		% of total	6.5%	18.2%	24.7%
	pure bee honey three days	Count	2	1	3
		% of use of treatment to control FMD	66.7%	33.3%	100.0%
		% of result	8.3%	1.9%	3.9%
		% of total	2.6%	1.3%	3.9%
	cassia nilotica and glycerin	Count	9	22	31
		% of use of treatment to control FMD	29.0%	71.0%	100.0%
		% of result	37.5%	41.5%	40.3%
		% of total	11.7%	28.6%	40.3%
	No	Count	8	16	24
		% of use of treatment to control FMD	33.3%	66.7%	100.0%
		% of result	33.3%	30.2%	31.2%
		% of total	10.4%	20.8%	31.2%
Total		Count	24	53	77
		% of use of treatment to control FMD	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

Appendix 2.32: Result of distribution of prevalence of FMD according to other information

Other information * Result of (VNT)

			Result of VNT		Total
			Negative	positive	
Other information	CBPP, Tick infestation and John's disease suspicious.	Count	0	2	2
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	Brucellla,Tick infestation and calves diarrhea.	Count	2	1	3
		% of other information	66.7%	33.3%	100.0%
		% of result	8.3%	1.9%	3.9%
		% of total	2.6%	1.3%	3.9%
	Brucellla, Tick infestation and mastitis	Count	4	3	7
		% of other information	57.1%	42.9%	100.0%
		% of result	16.7%	5.7%	9.1%
		% of total	5.2%	3.9%	9.1%
	Theileriasis,Tick infestation and House flies	Count	4	7	11
		% of Other information	36.4%	63.6%	100.0%
		% of result	16.7%	13.2%	14.3%
		% of total	5.2%	9.1%	14.3%
	Theileriasis and mastitis	Count	0	4	4
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	7.5%	5.2%
		% of total	.0%	5.2%	5.2%
	CBPP, Tick infestation and Theileriasis	Count	1	2	3
		% of other information	33.3%	66.7%	100.0%
		% of result	4.2%	3.8%	3.9%
		% of total	1.3%	2.6%	3.9%
	calves diarrhea, Tick infestation and theileriasts	Count	0	4	4
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	7.5%	5.2%
		% of total	.0%	5.2%	5.2%
	Pneumonia, calves diarrhea and Brucella suspicious	Count	3	4	7
		% of other information	42.9%	57.1%	100.0%
		% of result	12.5%	7.5%	9.1%
		% of total	3.9%	5.2%	9.1%

Appendix 2.32: continued

			Result of VNT		Total
			Negative	positive	
Other information					
	CBPP, calves diarrhea, Brucella suspicious, Tick infestation	Count	1	3	4
		% of other information	25.0%	75.0%	100.0%
		% of result	4.2%	5.7%	5.2%
		% of total	1.3%	3.9%	5.2%
	mastitis and calves diarrhea	Count	0	2	2
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	calves diarrhea and Tick infestation	Count	0	2	2
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	3.8%	2.6%
		% of total	.0%	2.6%	2.6%
	Tick infestation and Theileriasis.	Count	7	12	19
		% of other information	36.8%	63.2%	100.0%
		% of result	29.2%	22.6%	24.7%
		% of total	9.1%	15.6%	24.7%
	mastitis and lumpy skin disease	Count	1	2	3
		% of other information	33.3%	66.7%	100.0%
		% of result	4.2%	3.8%	3.9%
		% of total	1.3%	2.6%	3.9%
	mastitis	Count	0	1	1
		% of other information	.0%	100.0%	100.0%
		% of result	.0%	1.9%	1.3%
		% of total	.0%	1.3%	1.3%
	Theileriasis Tick infestation and mastitis	Count	1	4	5
		% of other information	20.0%	80.0%	100.0%
		% of result	4.2%	7.5%	6.5%
		% of total	1.3%	5.2%	6.5%
Total		Count	24	53	77
		% of other information	31.2%	68.8%	100.0%
		% of result	100.0%	100.0%	100.0%
		% of total	31.2%	68.8%	100.0%

APPENDIX 3

Hypothesis testing for association between FMD and potential risk factors in univariate analysis using Chi-Square Tests (χ^2).

Appendix 3.1: Locality

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	7.040(a)	5	.218
Likelihood Ratio	7.073	5	.215
Linear-by-Linear Association	.280	1	.596
N of Valid Cases	77		

a 5 cells (41.7%) expf < 5. Min exp = .31...

Appendix 3.2: Location

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	10.285(a)	11	.505
Likelihood Ratio	12.667	11	.316
Linear-by-Linear Association	.568	1	.451
N of Valid Cases	77		

a 20 cells (83.3%) expf < 5. Min exp = .31...

Appendix 3.3: Sex

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.194(b)	1	.659		
Continuity Correction(a)	.000	1	1.000		
Likelihood Ratio	.187	1	.666		
Fisher's Exact Test				.644	.499
Linear-by-Linear Association	.192	1	.661		
N of Valid Cases	77				

a Computed only for a 2x2 table

b 2 cells (50.0%) expf < 5. Min exp = 1.56...

Appendix 3.4: Age

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	8.190(a)	2	.017
Likelihood Ratio	7.723	2	.021
Linear-by-Linear Association	8.082	1	.004
N of Valid Cases	77		

a 2 cells (33.3%) expf < 5. Min exp = 2.81...

Appendix 3.5: Breed

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	1.911(b)	1	.167		
Continuity Correction(a)	.685	1	.408		
Likelihood Ratio	3.086	1	.079		
Fisher's Exact Test				.304	.216
Linear-by-Linear Association	1.886	1	.170		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 2 cells (50.0%) expf < 5. Min exp = 1.25...

Appendix 3.6: Body condition

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.014(b)	1	.905		
Continuity Correction(a)	.000	1	1.000		
Likelihood Ratio	.014	1	.906		
Fisher's Exact Test				1.000	.613
Linear-by-Linear Association	.014	1	.906		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 2 cells (50.0%) expf < 5. Min exp = 1.87...

Appendix 3.7: Herd size

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	3.294(a)	2	.193
Likelihood Ratio	3.557	2	.169
Linear-by-Linear Association	.033	1	.855
N of Valid Cases	77		

a 1 cells (16.7%) expf < 5. Min exp = 2.49...

Appendix 3.8: Production purpose

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.533(b)	1	.465		
Continuity Correction(a)	.223	1	.637		
Likelihood Ratio	.542	1	.462		
Fisher's Exact Test				.608	.322
Linear-by-Linear Association	.526	1	.468		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 8.42...

Appendix 3.9: Herd type

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.533(b)	1	.465		
Continuity Correction(a)	.223	1	.637		
Likelihood Ratio	.542	1	.462		
Fisher's Exact Test				.608	.322
Linear-by-Linear Association	.526	1	.468		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 8.42...

Appendix 3.10: Mixed species

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	2.579(a)	3	.461
Likelihood Ratio	2.396	3	.494
Linear-by-Linear Association	.009	1	.924
N of Valid Cases	77		

a 4 cells (50.0%) expf < 5. Min exp = .94...

Appendix 3.11: Previous history of FMD in the herd

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.982(b)	1	.322		
Continuity Correction(a)	.505	1	.477		
Likelihood Ratio	.956	1	.328		
Fisher's Exact Test				.402	.236
Linear-by-Linear Association	.969	1	.325		
N of Valid Cases	77				

a Computed only for a 2x2 table

b 0 cells (.0%) expf < 5. Min exp = 6.23...

Appendix 3.12: Other diseases

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	1.546(a)	3	.672
Likelihood Ratio	2.108	3	.550
Linear-by-Linear Association	.017	1	.898
N of Valid Cases	77		

a 5 cells (62.5%) expf < 5. Min exp = .62...

Appendix 3.13: Production System

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.126(b)	1	.723		
Continuity Correction(a)	.004	1	.949		
Likelihood Ratio	.128	1	.721		
Fisher's Exact Test				.780	.483
Linear-by-Linear Association	.124	1	.724		
N of Valid Cases	77				

A Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 5.61...

Appendix 3.14: Farm construction

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	.745(a)	4	.946
Likelihood Ratio	1.041	4	.904
Linear-by-Linear Association	.002	1	.969
N of Valid Cases	77		

a 5 cells (50.0%) expf < 5. Min exp = .31...

Appendix 3.15: Ventilation

	Value
Pearson Chi-Square	.(a)
N of Valid Cases	77

a No stats coz Ventilation constant

Appendix 3.16: Grazing system

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.697(b)	1	.404		
Continuity Correction(a)	.079	1	.779		
Likelihood Ratio	.648	1	.421		
Fisher's Exact Test				.585	.368
Linear-by-Linear Association	.688	1	.407		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 2 cells (50.0%) expf < 5. Min exp = 1.25...

Appendix 3.17: Water resources

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	2.477(a)	4	.649
Likelihood Ratio	2.308	4	.679
Linear-by-Linear Association	.024	1	.878
N of Valid Cases	77		

a 5 cells (50.0%) expf < 5. Min exp = .94...

Appendix 3.18: Use of concentrates

	Value
Pearson Chi-Square	.(a)
N of Valid Cases	77

a No stats coz Using of concentrates constant

Appendix 3.19: Artificial insemination

	Value	D.f.f	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	2.316(b)	1	.128		
Continuity Correction(a)	1.570	1	.210		
Likelihood Ratio	2.250	1	.134		
Fisher's Exact Test				.179	.106
Linear-by-Linear Association	2.286	1	.131		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 7.17...

Appendix 3.20: Where stay in dry season

	Value
Pearson Chi-Square	.(a)
N of Valid Cases	77

a No stats coz Where stay in dry season constant

Appendix 3.21: Where stay in rainy season

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.277(b)	1	.599		
Continuity Correction(a)	.058	1	.810		
Likelihood Ratio	.283	1	.595		
Fisher's Exact Test				.777	.412
Linear-by-Linear Association	.273	1	.601		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 5.92...

Appendix 3.22: Contact with neighboring herd

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.324(b)	1	.569		
Continuity Correction(a)	.104	1	.747		
Likelihood Ratio	.324	1	.569		
Fisher's Exact Test				.628	.374
Linear-by-Linear Association	.319	1	.572		
N of Valid Cases	77				

a Computed only for a 2x2 table

b 0 cells (.0%) expf < 5. Min exp = 11.84...

Appendix 3.23: Test of new animals before placement in the herd

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	1.090(b)	1	.297		
Continuity Correction(a)	.525	1	.469		
Likelihood Ratio	1.045	1	.307		
Fisher's Exact Test				.345	.231
Linear-by-Linear Association	1.075	1	.300		
N of Valid Cases	77				

a Computed only for a 2x2 table

b 1 cells (25.0%) expf < 5. Min exp = 4.36...

Appendix 3.24: Isolation pen for sick animals

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.667(b)	1	.414		
Continuity Correction(a)	.313	1	.576		
Likelihood Ratio	.659	1	.417		
Fisher's Exact Test				.448	.286
Linear-by-Linear Association	.659	1	.417		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 8.42...

Appendix 3.25: Visit of workers to other farms

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.692(b)	1	.405		
Continuity Correction(a)	.336	1	.562		
Likelihood Ratio	.686	1	.407		
Fisher's Exact Test				.455	.280
Linear-by-Linear Association	.683	1	.408		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 0 cells (.0%) expf < 5. Min exp = 9.35...

Appendix 3.26: Awareness of farmer with FMD

	Value
Pearson Chi-Square	.(a)
N of Valid Cases	77

a No stats coz Awereness of farmer with FMD constant

Appendix 3.27: Disposal of carcasses

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	1.791(a)	2	.408
Likelihood Ratio	1.834	2	.400
Linear-by-Linear Association	1.480	1	.224
N of Valid Cases	77		

a 0 cells (.0%) expf < 5. Min exp = 6.86...

Appendix 3.28: Manure disposal

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	5.146(a)	2	.076
Likelihood Ratio	5.385	2	.068
Linear-by-Linear Association	3.407	1	.065
N of Valid Cases	77		

a 3 cells (50.0%) expf < 5. Min exp = .62...

Appendix 3.29: Veterinary services

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	1.081(a)	2	.582
Likelihood Ratio	1.368	2	.504
Linear-by-Linear Association	.912	1	.340
N of Valid Cases	77		

a 2 cells (33.3%) expf < 5. Min exp = .31...

Appendix 3.30: Use of FMD vaccine to control the disease

	Value	D.f.	Asymptotic Significance	Exact Significance (2-sided)	Exact Significance (1-sided)
Pearson Chi-Square	.837(b)	1	.360		
Continuity Correction(a)	.283	1	.595		
Likelihood Ratio	.795	1	.373		
Fisher's Exact Test				.448	.289
Linear-by-Linear Association	.826	1	.363		
N of Valid Cases	77				

a Computed only for a 2x2 table
b 1 cells (25.0%) expf < 5. Min exp = 2.81...

Appendix 3.31: Use of treatment to control FMD

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	2.089(a)	3	.554
Likelihood Ratio	1.925	3	.588
Linear-by-Linear Association	.078	1	.780
N of Valid Cases	77		

a 2 cells (25.0%) expf < 5. Min exp = .94...

Appendix 3.32: Other information

	Value	D.f.	Asymptotic Significance
Pearson Chi-Square	12.000(a)	14	.606
Likelihood Ratio	16.038	14	.311
Linear-by-Linear Association	.604	1	.437
N of Valid Cases	77		

a 27 cells (90.0%) expf < 5. Min exp = .31...

APPENDIX 4

Questionnaire

Sudan University of science and Technology

College of Graduate Studies and Scientific research

College of Veterinary medicine

Master of Preventive Veterinary Medicine (MPVM)

Sero-Prevalence and Risk Factors of Foot and mouth disease Virus (sero-type A) in Cattle in Khartoum State, Sudan

a) General information:

State: …… locality: ……. Location: ……..

longitude: …….. Latitude: …….

Owner Name: ……

Tel. No.: ……

b) The individual risk factors:

1. Sex: Male () Female ()
2. Age: <2 year (younge) () 3- 4 year (median) () >4year Old ()
3. Breed : Local () Cross ()
4. Body condition: Good () Emaciated ()
5. Herd size: <50 cattle small herd() 50-150cattle medium herd () >150 cattle big herd()
6. Production purpose: Milk() Meat and milk()
7. Herd type mixed with other species: Yes() No()
8. Mixed species: Not mixed() Goa() Sheep() Sheep and Goat()
9. Previous history of FMD in the herd: Yes() No()
10. Other diseases: (mention the disease)……

c) The environmental risk factors (management):

1. Production system: Resident () Transhuman ()
2. Farm construction: Closed sheds of mud with roof of mats() Brick stables and zinc roof() pens of iron with roof of iron and mats() pens of zinc with roof of iron and mats() pens of firewood()
3. Ventilation: Good () Bbad ()
4. Grazing system: Private() Private and common()
5. Water resources: Private tap water from well in the area (project) () Private tap water () Private() Private tap water and common from Nile canal in the area() Private tap water from well in the area()s
6. Use of concentrates: Yes () No ()
7. Artificial incimination: No() Yes()
8. Where stay in dry season: In the farm () Out of the farm ()
9. Where stay in rain season: In the farm () Out of the farm()
10. Contact with neighboring herd : No() Yes()
11. Test of new animals before placeme: Yes () No ()
12. Isolation pen for sick animals: Yes () No ()
13. Visit of workers to other farms: No () Yes()
14. Awereness of farmer with FMD: Yes () No ()
15. Disposal of carcasses: Burning in incenerator of the project - burning in incenera () Burning out of the farm () Throw out farm()
16. Manure disposal : Sell to brecks makers weekly () Store out farm till selling () Store in the farm till selling()
17. Veterinary services: Daily supervision of a veterinarian () Veterinary, puplicand private hospital or clicinc in the are () Reguler veterinary supervision() not under veterinarian supervision() (mention it): …….
18. Use of FMD vaccine to control the disease : Yes () No ()

19. Other treatment to control FMD: Antibiotics() Pure bee haney 3 days() *Acassia nilotica* and glysren() No treatment()
20. Other informations:CBPP,Tick infestation and suspected with John's disease ()

Brucellla,Tick infestation and calves diarrhea ()

Brucellla,Tick infestation and mastitis ()

TheileriasisTick infestation and House flies ()

Theileriasis and mastitis ()

CBPP,Tick infestation and Theileriasis ()

calves diarrhea, Tick infestation and Theileriasts ()

Pneumonia, calves diarrhea and Brucella suspicious()

CBPP, calves diarrhea, suspected with Brucella,Tick infestation()

mastitis and calves diarrhea ()

calves diarrhea and Tick infestation ()

Tick infestation and Theileriasis ()

mastitis and Lumby skin disease ()

mastitis ()

TheileriasisTick infestation and mastitis ()