Journal of Advances in Microbiology

20(12): 70-76, 2020; Article no.JAMB.64348
ISSN: 2456-7116

Salmonella Serotypes Isolated from Beef Carcass Samples from Eswatini

B. N. Dlamini[1], C. Mudyanavana[2], T. H. Gadaga[1] and M. T. Masarirambi[1*]

[1]University of Eswatini, P/Bag 4, Kwaluseni, Eswatini.
[2]Ministry of Agriculture, Department of Veterinary and Livestock Services (DVLS), P. O. Box 4192, Manzini, Eswatini.

Authors' contributions

This work was carried out in collaboration among all authors. Author BND initiated the study, literature search, data collection, and coordinated writing and analysis. Authors THG and MTM contributed towards the initiation of the concept note and manuscript revisions. Author CM participated in data collection, analysis and preparation of figures. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMB/2020/v20i1230309
Editor(s):
(1) Dr. Simone Aquino, Instituto de Pesquisas Energéticas e Nucleares (IPEN), Brazil.
Reviewers:
(1) Sherifa Mostafa M. Sabra, Taif University, KSA.
(2) Guido Finazzi, IZSLER, Italy.
Complete Peer review History: http://www.sdiarticle4.com/review-history/64348

Original Research Article

Received 28 October 2020
Accepted 01 January 2021
Published 30 January 2021

ABSTRACT

Aim: The objective of this study was to determine the prevalence of *Salmonella* serovars isolated in beef carcasses slaughtered in Eswatini from 2017 to 2020.
Methods: Data analysed was officially recorded microbiological data from 2017 to 2020. Sterile broth moistened swabs were used aseptically to obtain carcass swab samples by using the non-destructive carcass sampling method. The method required that bacteriological sampling of carcasses be undertaken in the slaughterhouse without obtaining pieces of tissue samples from the sampling sites.
Results: A total of 1095 swab samples were analysed for *Salmonella* spp. A total of 25 *Salmonella* serovar isolates were identified during the study period. The predominant serovar was *Salmonella Shwarzengrund* (N=14; 56%). The second most frequently detected serovar was *Salmonella Heidelberg* (N=5; 20%). In addition, six other serovars were isolated and identified on carcasses. Each with a mean frequency of occurrence on the carcasses of N=1(4%).
Conclusion: This preliminary study has shown that various *Salmonella serovars* are present on beef carcasses slaughtered in Eswatini. About 25 *Salmonella serovars* were detected during the

**Corresponding author: E-mail: mike@uniswa.sz;*

study period with the most prevalent being *S. shwarzengrund* and *S. heidelberg*. It is recommended that further studies should be carried out to determine the antimicrobial resistance of *Salmonella* strains on beef carcasses in Eswatini.

Keywords: Salmonella; serovar; slaughtered cattle; carcass.

1. INTRODUCTION

Eswatini has an allocated annual 3,500 metric tonnes beef export quota to the European Union (EU). Export beef from Eswatini is locally processed at the export beef slaughterhouse. Considerable export earnings have been derived from the sale of beef to the European market. In order to meet the strict hygiene requirements of the European union, the beef slaughterhouse maintains an on-site Meat Hygiene Laboratory (MHL) which monitors microbial contamination standards [1,2,3].

The Norwegian National Food Law has zero tolerance policy on *Salmonella* spp [4]. Salmonellosis is one of several food borne zoonotic diseases [5]. In human salmonellosis is mainly associated with consumption of food contaminated with animal faecal matter [6].

All the developing countries including Eswatini, who are licensed to export beef to the EU, are required to implement and comply with all the regulations and requirements that are mandatory to the EU member states. Some of the EU standards and regulations stipulate the need to implement an effective Hazard Analysis Critical Control Point (HACCP) monitoring system [4,7].

This paper is based on secondary microbiological data of *Salmonella* species commonly isolated from export beef carcasses from Eswatini and discusses the public health and socio-economic importance of Salmonellosis to Eswatini [1,2,3].

According to [4], epidemiological studies on *Salmonella* species are very few. Therefore, detailed information on the prevalence of *Salmonella* spp. in the Southern Africa region is also very limited. On the other hand any research that indicates a low prevalence of *Salmonella* spp. contamination in beef may still point towards a possible risk with the zoonotic pathogen especially following consumption of meat. Additionally, this also implies the possibility of exporting the *Salmonella* serotypes to the export market via beef trade. There is a collective responsibility to address such a risk and as well as to communicate it to the public [8].

The incidence of sporadic domestically acquired salmonellosis due to *Salmonella typhimurium* is low [9].

1.1 Food Borne Zoonoses

Salmonellosis is described as an infectious disease of both humans and animals [5,6]. The infection in food animals may lead to contamination of meat, eggs, milk and cheese. Salmonellosis is one of the most common and economically important food-borne zoonotic diseases in humans [8-10]. Other *Salmonella* species are known to infect vegetables and subsequently infect humans through vegetable salad consumption.

Contamination of food by *Salmonella* bacteria continues to pose a risk to the health of consumers. Such a health risk becomes more of a reality because our daily routine, in terms of life styles make it a challenge to carry out adequate thermal food preparation. This increases the possibility of undercooking food thus unable to destroy *Salmonella* [11]. Meat is one of the most important foodborne pathogen vehicles [12,13]. The *Salmonella* spp. is listed amongst the most commonly reported bovine zoonotic pathogens [14].

Farm animals, as well as food of animal origin, are well known reservoirs of food borne pathogens. The surveillance and monitoring of such food borne disease causing agents in Europe is controlled according to Regulation (EC) No 854/2004 [15].

As reported by the European Centre for Disease Prevention and Control (ECDC), Annual report of the Director in 2018, indicated Salmonellosis was the second most commonly reported gastrointestinal infection in humans in the EU (91 857 cases reported), after campylobacteriosis (246,571). Similarly, as reported by the by the European Food Safety Authority (EFSA) and the ECDC, nearly one in three foodborne outbreaks in the EU in 2018 were caused by *Salmonella species* [16].

Salmonella has been found to be widespread in minced beef, mutton and pork samples obtained from retail supermarkets in Addis Ababa, Ethiopia. This observation prompted the researchers to suggest that proper cooking of meat before consumption and improving personal and meat hygiene in the line of meat production from farm to fork should be adopted to ensure the safety of meat and meat products for human consumption [17]. Another Ethiopian study revealed that even apparently healthy slaughtered cattle and working personnel at the abattoir maybe a source of *Salmonella* species [18].

1.2 Socioeconomic Importance of Salmonellosis in Eswatini

Salmonellosis is both a zoonotic and a disease of socio-economic importance in Eswatini. The license to export beef to Norway not only provides the necessary foreign exchange income, it also entails provision of technical assistance to local cattle farmers, community service to some rural households and provision of employment to several slaughterhouse workers [1-3].

The Norwegian technical assistance employs the value chain approach from farm to fork. This successful enterprise utilises the livestock industry to impact on poverty reduction in rural farming communities of Eswatini. In the European Union, a study has revealed that livestock does actually contribute towards poverty reduction [19].

Eswatini local subsistence cattle farmers may receive some farming inputs from the slaughterhouse operator. They also get technical and marketing support when the cattle are ready for sale. There is transport that collects and conveys cattle to the slaughterhouse. Livestock generate income for farmers of all categories via sale of animals and livestock products. The income from livestock is a vital source of income for household essentials, such as payment of school fees, medical expenses and enables farmers to afford better diets and to make proper health choices [10,19].

On the negative side, Salmonellosis is viewed as a major public health problem worldwide. It contributes to negative economic impacts associated with the cost of surveillance, treatment and prevention of the disease [5,6]. According to [14], pathogenic agents responsible for causing zoonotic diseases play a significant role as a source of disease to humans, and can have some detrimental effects to public health as well as socio-economically [14]. Zoonotic infections can severely undercut livestock productivity and thereby reduce farm revenues [19,20]. There are few comprehensive studies currently available on the aggregate direct and indirect employment generation and socio-economic impacts of the livestock sector in low and middle-income countries at the country or regional level [10]. This study is aimed at highlighting both the prevalence of *Salmonella* serovars isolated from beef carcasses slaughtered in Eswatini from 2017 to 2018 and the socioeconomic importance of salmonellosis in the country.

2. MATERIALS AND METHODS

The recorded microbiological data of the results of regular *Salmonella* bacteria checks conducted on slaughter carcasses from 2017 to 2020 were analysed. In addition, this study investigated and briefly outlined the socio-economic role of *Salmonella* spp. and the technical assistance offered to subsistence cattle farmers in the Eswatini livestock industry [1-3].

The sampling by the slaughterhouse operator was carried out according to Commission Decision 2001/471/EC. Carcass swab samples were taken from sites where the risk of microbiological contamination was most likely to occur. From each carcass 4 sites were sampled and pooled to make one sample from that carcass. This was done on the day of slaughter, before carcasses were chilled. Carcass swab samples were then stored at 4^0 C before transportation to the veterinary laboratory. The non–destructive carcass sampling method was used, whereby swabs were moistened prior to sampling using 0.1% sterile peptone + 0.85% sodium chloride diluent. The moistened swab was rubbed to cover a sampling area of at least 10 cm^2 per sampling site (Fig. 1) [21,22].

2.1 Microbial Analyses

The laboratory testing of export beef carcass meat samples for *Salmonella* species and total bacteria counts are subcontracted to the Deltamune veterinary laboratory, South Africa. For the detection of *Salmonella* in clinical cases, egg and egg products, environmental samples, feed and feed raw materials test method SER-

TMD-ME-0008 is used routinely. The said laboratory is known to be in full compliance with Commission Regulation (EC) No. 2073/2005 [8,21-23].

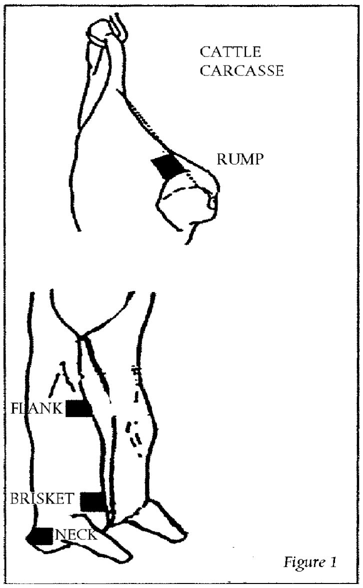

Fig. 1. Sampling sites for Salmonella testing of carcasses (neck, brisket, flank and rump)
Source: [21]

3. RESULTS

From 2010 to 2016 a steady increase in the total number of cattle slaughter was realised. However, there was a drastic decline in cattle slaughter experienced in the years 2017 and 2018 because of the suspension of beef exports from Eswatini to the EU (Fig. 2). The country thereafter resumed beef exports to the EU in November 2018. Fig. 2 indicates the relationship between total slaughtered cattle and the primal beef cuts exported to the global market.

The frequency of *Salmonella serovars* isolated and identified from all the carcass swabs samples collected during the study period are shown in Fig. 3.

During the period from 2017 to 2020, a total of 1095 carcass samples were collected and sent for testing at the Deltamune Laboratory in South Africa. There were a total of 25 *Salmonella* positive samples. *Salmonella Schwarzengrund* and *S. heidelberg* were the most commonly isolated serovars. Fig. 3. shows the histogram of distribution of *Salmonella serovars* that were isolated and serotyped.

4. DISCUSSION

The results of this study indicated carcass contamination with various *Salmonella serovars* that were identified on carcasses. However, the source of the carcass contamination and susceptibility towards local available antimicrobials is not yet determined [1-3].

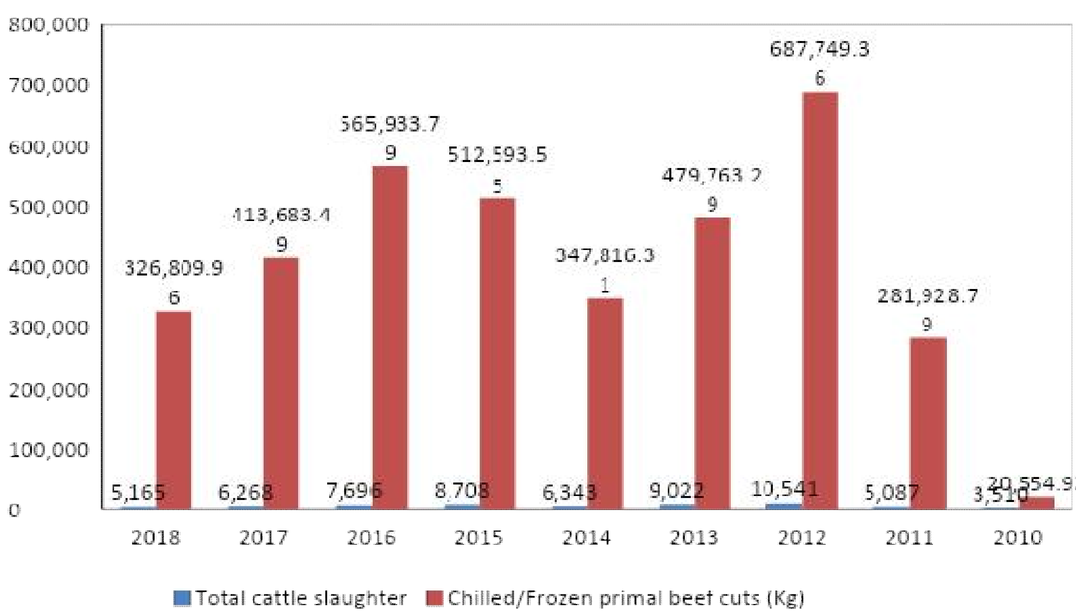

Fig. 2. Total amount of beef carcass meat certified for export from 2010 – 2018

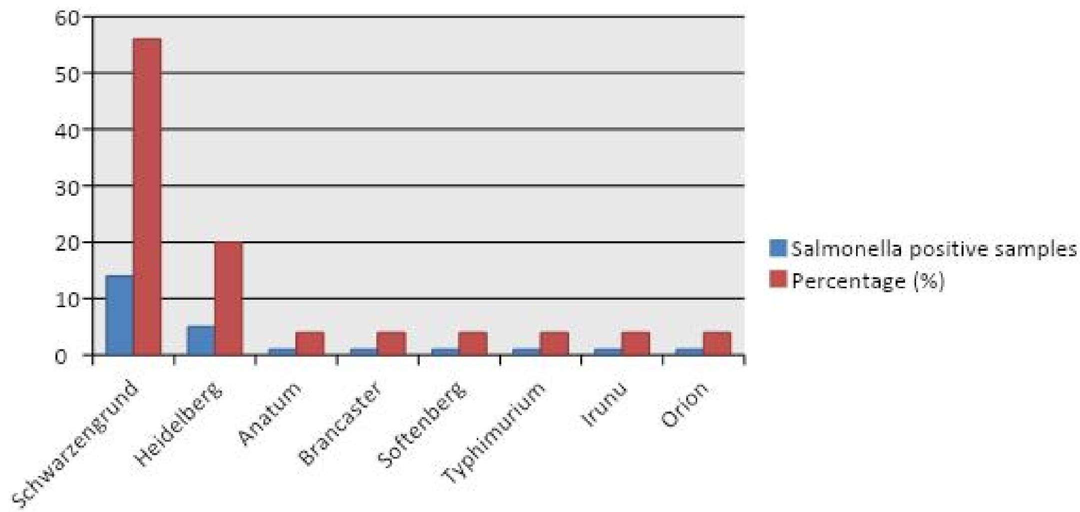

Fig. 3. Frequency of Salmonella serovars isolated from carcass swabs analysed from 2017 – 2020

It is important to note that beef from Eswatini, had *Salmonella enterica serovar Schwarzengrund* as the most prevalent serovar during the 4 year period of study of 2017 to 2020. This serovar was the most predominant and was detected in 56% of the carcasses. *Salmonella Schwarzengrund* was previously reported to be a rare cause of human salmonellosis. Some recent reports showed that the prevalence of this serovar was increasing [24]. Unlike the study on *Salmonella* species from beef originating from another African country, the most prevalent *Salmonella serovars* isolated from beef in Namibia was *Salmonella Chester* (18.54%) followed by *Salmonella Schwarzengrund* (7.30%) [5,13]. *Salmonella Chester* was not isolated from beef carcasses from Eswatini during the study period from 2017 to 2018.

The second highest reported *Salmonella enterica serovar* from Eswatini for the fresh bovine carcass meat was *S. enterica serovar Heidelberg* (20%). This strain is among the most commonly detected serovars from retail meats and food animals and ranks fourth among serovars associated with human infections [25,26].

From the Eswatini results, the following remaining *Salmonella enterica serovars* namely; *Irunu, Anatum, Heildelberg, Brancaster, Softenberg, Typhimurium and Orion*, were all detected at 4% of the carcass samples. The results of this study indicate that 25 carcasses were positive for *Salmonella* species out of a total of 219 carcasses (11.4%) tested during the study period. Considering the Norwegian policy of zero *Salmonella* tolerance, this prevalence of *Salmonella spp.* could be viewed to suggest that even more stricter biosecurity measures may be necessary to eliminate any remaining possible sources of faecal carcass contamination with *Salmonella* species [27]).

Also, the analysed recorded microbiological data did not include environmental samples neither did it cover antimicrobial resistance. Only recorded microbiological data from carcass swab samples (N= 1095) from the abattoir was available for our analysis. Therefore, this study may indicate the need to further research [1-3].

5. CONCLUSION

A total of 25 carcasses were positive for *Salmonella serovars* from a total of 219 carcasses sampled from slaughtered cattle during the study period (11.4%). Also, this work has briefly outlined the socioeconomic importance of the *Salmonella* spp. monitoring and surveillance program to the subsistence cattle farmers of Eswatini. Further detailed studies are recommended to determine the exact source of faecal *Salmonella* spp. carcass contamination, the potential risk factors that influence the carcass contamination from farm to slaughter. Such a study, on the susceptibility of the identified *Salmonella serovars* to locally available antimicrobials may contribute towards making informed decisions on the antimicrobial resistance pandemic.

ACKNOWLEDGEMENTS

The authors of this paper would like to acknowledge D.C. Mabuza for his kind assistance during the study. Likewise, we wish to recognise the Deltamune laboratory, South Africa for conducting the necessary microbiological analyses on all the carcass swab samples submitted by the responsible government officials.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Ministry of Agriculture. Annual performance reports. Mbabane, Swaziland; 2014.
2. Ministry of agriculture and cooperatives. Master plan for veterinary and livestock research centre. Agricultural Res. Foundation, Nairobi, Kenya. 1993;1:1-8.
3. Ministry Of Agriculture. Annual Performance reports 2018, Mbabane, Eswatini; 87-90.
4. Shilangale P, Kaaya P, Chimwamurombe M. Prevalence and characterization of *Salmonella* isolated from beef in Namibia. Euro. J. Nutrition and Food Safety. 2015;5(4):267-274.
5. Pui F, Wong C, Chai C, Tunung R, Jeyaletchumi P, Noor Hidayah S, Ubong A, Farinazleen G, Cheah K, Son R. Review article *Salmonella*: A foodborne pathogen. Int. Food Res. J. 2011;18(1):465-473 1.
6. European food safety authority and european centre for disease prevention and control. The European Union One Health 2018 Zoonoses Report. EFSA J. 2019;17(12):5926,276. Available:https://doi.org/10.2903/j.efsa.2019.5926 ISSN: 1831-4732 Accessed on 13/5/2020.
7. Bhunia A. Biosensors and bio-based methods for the separation and detection of foodborne pathogens. Advances in Food and Nutrition Res. 2008;54:1-44.
8. World Organisation for Animal Health (OIE) Terrestrial Manual. Paris, France, Chapter 3. 9.8 – Salmonellosis. 2018;1735-1746.
9. Heier T, Lange H, Hauge K, Hofshagen M. Norway–trends and source of zoonoses and zoonotic agents in humans, foodstuffs, animals and feeding stuffs, 2013;7-19.
10. Varijakshapanicker P, Mckune S, Miller L, Hendrickx S, Balehegn M, Dahl E, Adesogan T. Sustainable livestock systems to improve human health, nutrition, and economic status, Animal Frontiers. 2019;9(4):39–50.
11. Cabral C, Conte-Junior A, Silva T, Paschoalin F. *Salmonella* spp. contamination in fresh pork and chicken sausages marketed in Nitero´i, Rio de Janeiro, Brazil J. of Consumer Protection and Food Safety. 2014;1 – 7.
12. Barbau-Piednoir E, Botteldoorn N, Mahillon J, Dierick K, Roosens H. Fast and discriminative CoSYPS detection system of viable *Salmonella* spp and *Listeria* spp. In carcass swab samples. Int. J. of Food Micro. 2015;2:103-110.
13. Shilangale R, Kaaya G and Chimwamurombe P. Antimicrobial resistance patterns of *Salmonella* Strains isolated from beef in Namibia. British Microbiology Research Journal. 2016;12(1):1-6.
14. McDaniel J, Cardwell M, Moeller Jr. B and Gray C. Humans and cattle: A review of Bovine Zoonoses. Vector Borne and Zoonotic Dis. 2014;14(1):1–19.
15. Giombelli A, Cavani R, Gloria MBA. Evaluation of three sampling methods for the microbiological analysis of broiler carcasses after immersion chilling J. of Food Prot. 2013;76(8):1330-1335. Available:doi:10.4315/0362-028X.JFP-13004
16. European food safety agency and european centre for disease prevention and control. The European union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2013. European food Safety authority (EFSA), Parma, Italy. European centre for disease prevention and control (ECDC), stockholm, sweden. EFSA Journal. 2016;13(1):3991.
17. Ejeta G, Molla B, Alemayehu D and Muckle A. *Salmonella* serotypes isolated from minced meat beef, mutton and pork in Addis Ababa, Ethiopia. Website for the Revue de Méd. Vét. 2004;11(155):547-551.
18. Takele S, Woldemichael K, Abdissa A. Prevalence and drug susceptibility pattern of *Salmonella* isolates from apparently healthy slaughter cattle and personnel

working at the Jimma municipal abattoir, South-West Ethiopia. Tropical Diseases; 2018.

19. Perry B, Grace D. The impacts of livestock diseases and their control on growth and development processes that are pro-poor. Philosophical Transactions of The Royal Society, series B, Bio. Scie. 2009; 364(1530):2643-55.
 DOI: 10.1098/rstb.2009.0097
20. Grout L, Baker MG, French N, Hales S. A review of potential public health impacts associated with the global dairy sector. Geo Health. 2020;4:2019GH000213.
21. Commission Decision 2001/471/EC. Laying down rules for the regular checks on the general hygiene carried out by the operators in establishments according to Directive 64/433/EEC on health conditions for the production and marketing of fresh meat and Directive 71/118/EEC on health problems affecting the production and placing on the market of fresh poultry meat. Official Journal of the European Communities. 2001;L165/48–L165/53.
22. Council Directive 96/23/EC. Consolidated text on measures to monitor certain substances and residues thereof in live animals and animal products and repealing Directives 85/358/EEC and 86/469/EEC and Decisions 89/187/EEC and 91/664/EEC.
 Available:http://data.europa.eu/eli/dir/1996/23/2019-12-14.
 Accessed 27/4/2020
23. Commission Regulation (EC) No 2073/2005. On microbiological criteria for foodstuffs (Text with EEA relevance). OJ L. 2005;338:1.
 Accessed on 4/5/2020.
24. Nueesch-Inderbinen M, Cernela N, Hächler H. Characteristics of *Salmonella enterica* serotype *Schwarzengrund* associated with human infections in Switzerland: 2006-2010. J. of Food Safety and Food Quality. 2015;66:56-60.
 Accessed on 2/7/2020.
25. Han J, Donna E, David JD, Deck J, Lynne AM, Kaldhone P, Nayak R, Stefanova R, Foley SL. Comparison of *Salmonella enterica Serovar Heidelberg* isolates from human patients with those from animal and food sources. J. of Clini. Micro. 2010;49(3):1130-1133.
 DOI:10.1128/JCM.01931-10 Accessed on 3/7/2020
26. MacDonald E, White R, Mexia R, Bruun T, Kapperud G, Brandal LT, Lange H, Nygård K, Vold L. The role of domestic reservoirs in domestically acquired *Salmonella* infections in Norway: Epidemiology of salmonellosis, 2000–2015, and results of a national prospective case–control study, 2010–2012, Epidemiology and Infection. 2019;147-151.
27. Sasaki Y, Murarami M, Maruyama N, Tsujiyama Y, Kusukawa M, Asai T, Yamada Y. Risk factors for *Salmonella* prevalence in laying-hen farms in Japan, Epidemiology and Inf. 2012;140(6):982990.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/64348