Asian Journal of Multidisciplinary Studies

ISSN: 2321-8819 (Online)
2348-7186 (Print)
Impact Factor: 1.498
Vol.4, Issue 5, April 2016

Somatic Cell Count and Antibiotic Residues in raw milk Produced by Smallholder Dairy Farmers in Lusaka Province of Zambia.

B. Kunda[1], G. S. Pandey[2] and J. B. Muma[3]

[1.] PG Scholar, University of Zambia, School of Veterinary Medicine, Department of Disease Control, P.O. Box 32379, Lusaka, Zambia.
[2.] Guide and Professor, University of Zambia, School of Veterinary Medicine, Department of Disease Control, P.O. Box 32379, Lusaka, Zambia,
[3.] Associate Professor, University of Zambia, School of Veterinary Medicine, Department of Disease Control, P.O. Box 32379, Lusaka, Zambia

Abstract: *This study was carried out to evaluate the Somatic Cell Count (SCC) and Antibiotic Residues (ARs) in raw milk produced by smallholder dairy farmers (SHDFs) in Lusaka province of Zambia. The study was done during the months of January and February 2014. The SCC was measured using a DeLaval Cell Counter (DeLaval International AB, Sweden) while Copan Milk Test 100 (Copan Diagnostics Inc., Denmark) was used to test for presence of ARs. A total of 83 farmers participated in the study and results revealed that milk from 51 out of the 83 farms (61.44%) had SCC above the maximum recommended number of 300,000 cells/ml by Zambia bureau of standards. The high SCC ranging from 263 to 22.3 x 10^6 cells/ml was due to subclinical mastitis not noticeable on clinical observation. Milk samples from 25 out of the 83 farms (30.12%) tested positive for ARs. The high SCC and ARs found in the milk indicates the need for further education and training of the SHDFs in aspects of good milk production which should include correct usage of antibiotics and observance of strict withdrawal period after antibiotic treatment from public health point of view. The regulatory authority should establish maximum limits for SCC and ARs in raw milk to be sold to public or processors and monitored regularly.*

Keywords : Raw milk, somatic cell, antibiotic residue, Zambia

INTRODUCTION:

Somatic Cell Count (SCC) of milk directly represents the inflammatory status (mastitis) of the mammary gland (udder) from where milk was collected (Salman and Elnasri, 2011). Somatic cells are primarily leukocytes (white blood cells) and some epithelial cells shed from the lining of the mammary gland (Eberhart *et al*., 1982). The leukocytes are derived from blood and consist of macrophages, lymphocytes and neutrophils (Harmon, 1994). The macrophages are involved in immune recognition and are the predominant cell type present in milk from uninfected mammary glands. Lymphocytes are responsible for immune memory. The neutrophils are involved in defense against an invasion of the mammary gland by microorganisms and are the predominant cell type in milk from infected glands (Karimuribo *et al*., 2005). High SCC is associated with an increased risk of clinical mastitis, decreased milk yield and shorter shelf life of dairy products (Hutton *et al*., 1990). Mastitis in both clinical and subclinical form affects milk production. Apart from lowered productivity, mastitis reduces milk quality as a result of changes in milk composition and also by contamination of milk by drugs used for treatment of the disease (Karimuribo *et al*., 2005) and also affecting public health problem among consumers.

In Zambia, the maximum limit of the number of somatic cells per ml of milk that is acceptable by law is not stated. The Food and Drug Act of 2001 only states that milk sold in Zambia for manufacture into dairy products should not contain any inflammatory product and somatic cells are the main inflammatory product of mastitis. However, the Zambia Bureau of Standards (ZABS) has set 300,000 cells/ml in raw milk as the maximum limit. This maximum limit for somatic cells is low compared with standards set in many other countries. In EU and USA, maximum limit is 400,000 and 750,000 cells/ml of milk respectively (Smith and Hogan, 1999). In South Africa, according to section 15(1) of the Foodstuffs, Cosmetics and Disinfectant Act (Act No. 54 of 1972), the maximum accepted limit for somatic cells is 500,000 cells/ml of milk. The reason for higher maximum accepted limit for somatic cells in these other countries, especially the developed countries, in view of the authors might possibly be due to the fact that high producing exotic breeds of cattle (breeds found in developed countries) are more prone to mastitis and high volume of milk producing cows (Rauberts and Shook, 1982).

In the dairy industry, especially in the developed countries, SCC has become one of the most reliable indicators for determining milk quality and the price of raw milk (Jayarao *et al*., 2006). In Zambia, however, there is lack of information on quality of raw milk with regards to SCC and antibiotic residues. The only available research based

published literature on SCC of raw milk is by Pandey *et al*. (1996) who attempted to study the sanitary quality of raw milk from dairy farms supplying milk to the now defunct Dairy Produce Board (DPB) in Lusaka, Zambia. The study by Pandey *et al*. (1996) focused on commercial dairy farmers who had high yielding exotic breeds. The current study focused on SHDFs who have average yielding local and cross breeds and are now major source of milk supply to processors due to support to smallholder dairy development in Zambia by many stakeholders in recent time. It was therefore envisaged that in terms of SCC, this study would generate updated information on the quality of raw milk produced by SHDFs in Lusaka province of Zambia.

Antibiotics are usually used for the prevention and treatment of animal diseases and to improve the efficacy of animal production (Sana *et al*., 2005). They have been used in cows for many years to treat infections such as mastitis (Seymour *et al*., 1988). The past several years has seen increased pressure on milk producers to increase milk production from each and every cow. This pressure often results in more infections, and the increased use of antibiotics to counter these infections (Shitandi and Kihumbu, 2004). After treating with antibiotics, if farmers milk their cows without adhering to the withdrawal period recommended by the manufacturer of the antibiotics used, ARs can be found in milk (Khanik, 2007). Antibiotic Residues (ARs) are important for three major reasons. First, microorganisms can develop antibiotic resistance when exposed to milk containing ARs when given underdoses. This renders antibiotic treatment against micro-organisms ineffective (Mitchell *et al*., 1998). Second, some antibiotics have side effects on consumers and therefore consumption of milk containing ARs can cause some complications among milk consumers. Examples of such complications are allergies such as urticaria, dermatitis, asthma and rhinitis (Nero *et al*., 2007). Some antibiotics, including nitrofuranes and chloramphenicol have a carcinogenic effect in laboratory animals. This represents a potential risk to consumers of milk which contain ARs (Movassagh and Karami, 2010). Third, the bactericidal and bacteriostatic activity of ARs can adversely affect the fermentation process of milk into cheese and cultured products, such as mabisi (sour milk) and yoghurt by inhibiting the starter cultures as starter cultures contain selected viable bacteria (Kang'ethe *et al*., 2005).

The Maximum Residue Limits (MRLs) for ARs in milk have been established in many countries including European Union. These MRLs are set at levels which are not likely to be exceeded if the veterinary drugs are used in accordance with approved label instructions (Nouws *et al*., 1998). In Zambia, MRLs have not been established. However, the law only states that milk sold for manufacture into dairy products is required to have no antibiotics or other antimicrobial substances (The Food and Drugs Act, 2001).

The purpose of this study was to assess the SCC and ARs in raw milk produced by SHDFs in Lusaka Province of Zambia. This is the first report, as far as we are aware, of antibiotic residues and somatic cell count in cow's raw milk produced by smallholder dairy farmers in Zambia.

MATERIALS AND METHODS:

The study was done in Lusaka Province of Zambia during the months of January and February 2014. Lusaka province lies in the south central part of Zambia between 28 and 30 degrees east and between 15 and 16 degrees south. It covers a total area of 75,261 square kilometers and has a human population of 2,191,225 people (CSO, 2012).

A cross-sectional study design was used in the study implementation. Records at the milk collection centres (MCCs) and the Dairy Association of Zambia (DAZ) were used to randomly select 83 SHDFs who participated in the study. 26 farmers belonged to Palabana dairy scheme MCC, 20 to Mapepe dairy co-operative society MCC and 27 to Lusaka west farming block who deliver milk directly to Parmalat milk processor. Farmers from these areas were selected to participate in the study because that is where the majority of SHDFs in Lusaka Province are found. Sampling units were individual smallholder dairy farms. 50 ml from the bulk raw milk of each of the 83 selected farms was collected aseptically into separate sterile 50 ml sample bottles as milk was being delivered to MCCs. The collected samples were stored in an ice packed cooler box and transported to University of Zambia (UNZA), School of Veterinary Medicine Public Health laboratory for further analysis.

Somatic Cell Count (SCC)

A DeLaval Cell Counter (DeLaval International AB, Sweden) was used to count somatic cells in the milk. To measure somatic cells, a cassette of the DeLaval Cell Counter, which contains a reagent (a DNA specific fluorescent probe), was used to collect 1.0 µl of milk sample. Once inside the cassette, milk mixes with the reagent. The reagent then reacts with the nuclei of the somatic cells and when the cassette is inserted in the DeLaval Cell Counter, it is exposed to light emitted by the DeLaval Cell Counter. This gives rise to fluorescence signals which are recorded in an image. The image is used to determine the number of somatic cells in the milk which appears on the screen of the DeLaval Cell Counter (DeLaval, 2005) and the value was recorded for each milk sample on the day of collection.

Antibiotic Residues (ARs)

To test milk samples for presence of ARs, the Copan Milk Test 100 (Copan Diagnostics Inc.,

USA) kit was used. This is a qualitative test for detecting the presence of ARs in milk. In this test, *Bacillus stearothermophilus var. calidolactis* spores are enclosed within an agar based gel matrix containing nutritive substances and a pH indicator within a tube. When milk sample which is free from ARs is added and incubated at 64°C for 3 hours, the bacterial spores within the test kit media germinate and produce acid which contributes to a pH drop. The pH drop causes a colour change from purple to yellow. However, if ARs are present, the spores will not germinate and no acid produced and hence the colour of the media remains unchanged, that is purple.

Statistical analysis

Data which was generated from laboratory tests on the 83 samples of raw milk was first entered into Microsoft excel then transferred to SPSS version 20 for analysis. Independent samples t test to establish whether there was any statistical significant difference between milk which tested positive and that which tested negative for ARs regarding SCC at a significance level of 0.05 was conducted.

RESULTS AND DISCUSSION:

In this study, the somatic cell count and antibiotic residue in raw milk produced by SHDFs in Lusaka Province of Zambia was established by evaluating its SCC and ARs, Somatic Cell Count (SCC) ranged from 263 to 2.312 x 10^6 cells/ml of raw milk. Milk from 51 out of the 83 farms (61.44%) had SCC above the recommended maximum limit of 300,000 cells/ml of raw milk. Milk from 25 out of 83 farmers (30.12%) tested positive for presence of ARs. Independent samples t - test results showed that there was no statistical significant difference in SCC for milk that was positive for ARs and that which was negative for ARs. Production of raw milk of good sanitary quality by farmers is important to milk processing companies, milk consumers and the farmers themselves. This is so for milk processing companies because raw milk containing high somatic count and antibiotic residue has reduced processing properties and processed milk and milk products made from such raw milk have a reduced shelf life (Oliver *et al*., 2005). For consumers, consumption of milk contaminated with bacterial toxins and ARs, can lead to diseases, allergic reactions, toxication and a risk of microorganisms developing resistance. For SHDFs, producing milk of good quality is important because milk processing companies pay farmers in accordance with the hygienic and compositional quality of raw milk delivered to them (Yambayamba and Zulu, 2011) and even rejection of milk by processors if milk contained antibiotic residue or milk was obtained from cows with mastitis.

In our study, SCC of raw milk from 51 out of 83 farmers (61.44%), did not conform to recommended standards (300,000 cells / ml of raw milk). This was an indication that raw milk came from cows with inflamed udders (mastitis) and that the farmers were making losses in milk production because cows with high SCC (mastitis) have decreased levels of milk production. In addition, when cows are put on mastitis treatment, milk is discarded and not sold during treatment and withdrawal periods thus a loss to farmers. In similar studies done in other countries, Salman and Elnasri (2011) found high SCC in a study conducted in Khartoum State of Sudan. Authors found that SCC of raw milk produced by 56.70% SHDFs did not conform to that country's recommended standards of 5.0 x 10^5 cells/ml of raw milk. In a study done in Mbarara and Kiruhura districts, the major cattle corridor in Uganda, Rutaro (2004) found an average SCC of 507,000 cells/ml of raw milk and milk samples from 66% of the farms had SCC which did not conform to recommended standards.

This study has produced the first ever report on ARs in milk in Zambia where 25 out of 83 farms (30.12%), tested positive for ARs. Farmers were probably not using antibiotics correctly and were not observing recommended withdrawal periods of antibiotics before supply of milk to processors. Findings in this study were close to those reported in Pakistan, Iran and Brazil. In Pakistan, Khaskheli *et al*. (2006) found ARs level of 36.5%, while in Iran 32.9% of raw milk tested was found positive for ARs (Mokhtari *et al*., 2013). In Triangle Region of the State of Minas Gerais, Brazil Tetzer *et al*. (2005) found 33.3% of raw milk samples positive for ARs. In Zimbabwe, Mhone *et al*. (2012) found 2.5% of raw milk from smallholder dairy farms positive for ARs and in Kenya 21% of raw milk was found positive for ARs (Shitandi and Kihumbu, 2004). In Nepal Sharma et al (2011) found 27% of the milk sample were positive for antibiotic residue above the recommended level.

Presence of antibiotic residue in food of animal origin such as milk, meat, eggs poses significant health hazards in humans. It is the duty of the state to ensure that these foods need to be screened out routinely for human safety (Sharma *et al* 2011).

In Zambia, antibiotics for pharmacological and agricultural use are sold without prescription and used indiscriminately by farmers. This could have contributed to higher number of milk samples being positive for ARs. Heat processing of milk does not eliminate or degrade ARs contained in milk (Moats 1988, Loksuwan 2002, Sharma *et al* 2011, de Oliveira *et al* 2012). There is therefore need for enforcing the withdrawal period of milk after antibiotic treatment and the enforcing by the regulatory authority the sale of antibiotics on prescription by veterinary personnel.

CONCLUSION AND RECOMMENDATIONS:

- This study found that raw milk produced by SHDFs (61.44%) in Lusaka Province of Zambia did not conform to recommended standards of somatic cell count and 30.12 % of the farms had antibiotic residues, in their milk,
- The study recommended that training in good dairy farm management practices should be conducted regularly in order to assist SHDFs adhere to correct usage of antibiotics, improve the udder health of their cows.
- It was also recommended that the regulatory authority in Zambia should establish maximum limits for SCC and ARs in raw milk to be sold to public or processors.

REFERENCES:

Central Statistical Office of Zambia (CSO) (2012) Census of population and housing of 2010.

de Oliveira L. P., Barros L. S. S., Silva V. C. and Cirqueira M. G. (2012) Microbiological quality and detection of antibiotic residue in raw and pasteurized milk consumed in the Reconcavo area of the state of Bahia, Brazil. *Journal of Food Processing Technology,* 3, 1 doi.org/10.4172/2157-7110.1000137.

DeLaval cell counter DCC instruction manual. 2005.

Eberhart, R.J., Hutchinson, L.J. and Spencer, S.B. (1982) Relationships of bulk tank somatic cell counts to prevalence of intramammary infection and indices of herd production. *Journal of Food Protection,* 45, 1125-1128.

Harmon, R.J. (1994). Physiology of mastitis and factors affecting somatic cell counts. *Journal of Dairy Science,* 77, 2103-2113.

Hutton, C.T., Fox, L.K. and Hancock, D.D. (1990) Mastitis control practices: differences between herds with high and low milk somatic cell counts. *Journal of Dairy Science,* 73, 1135-1143.

Jayarao, B.M., Donaldson, S.C., Straley, B.A., Sawant, A.A., Hegde, N.V. and Brown, J. L. (2006) A survey of foodborne pathogens in bulk tank milk and raw milk consumption among farm families in Pennsylvania. *Journal of Dairy Science,* 89, 2451-2458.

Kang'ethe, E.K., Aboge, G.O., Arimi, S.M., Kanja, L.W., Omore, A.O. and McDermott, J. J. (2005). Investigation of the risk of consuming marketed milk with antimicrobial residues in Kenya. *Journal of Food Control,* 16 (4), 349-355.

Karimuribo, E.D., Mdegela, R.H., Kusiluka, L.J.M. and Kambarage, D.M. (2005) Assessment of drug usage and antimicrobial residues in milk on smallholder farms in Morogoro, Tanzania. *Bulletin of Animal Health and Production in Africa*, 53, 234-241.

Khanik, G.R.J. (2007) Chemical contaminants in milk and public health concerns: A review. *International Journal of Dairy Science,* 2(2), 104-115.

Khaskheli, M., Malik, R.S., Arain, M.A., Soomro, A.H. and H.H. Arain. (2006). Detection of ß - Lactam Antibiotic Residues in Market Milk. *Pakistan Journal of Nutrition*,7(5), 682-685.

Loksuwan J. (2002) The effect of heating on multiple residue of tetracycline in milk. Thammaset. *International Journal of Science and Technology,* 7 (3), 17-21.

Mhone, T.A., Matope, G. and Saidi, P.T. (2012) Aerobic bacterial, coliform, Escherichia coli and *Staphylococcus aureus* counts of raw and processed milk from selected smallholder dairy farms of Zimbabwe. *International Journal of Food Microbiology,* 151 (2), 223-228.

Mitchell, J.M., Griffiths, M.W., McEwen, S.A., McNab, W.B. and Yee, A.J. 1998. Antimicrobial drug residues in milk and meat: Causes, concerns, prevalence, regulations, tests and test performance. *Journal of Food Protection,* 61, 742-756.

Moats W A (1988) Inactivation of antibiotics by heating in foods and other substrates; A review. *Journal of Food Protection,* 51 (6), 491-497.

Mokhtari, A., Hosseini, B. and Panahi, P. (2013) B-Lactam and tetracycline antibiotic residues detected in bulk tank milk in Iran. *Iranian Journal of Public Health,* 19, 447-448.

Movassagh, M.H. and Karami, A.R. (2010) Determination of Antibiotic Residues in Bovine Milk in Tabriz, Iran. *Global Veterinary Journal,* 5(3), 195-97.

Nero, L.A., Mattos, M.R., Beloti, V., Barros, M.A.F. and Franco, B.D.G.M. (2007) Residues of antibiótics in raw milk from four dairy regions in Brazil. *Ciênc Tecnol Aliment Campinas,* 27, 391-393.

Nouws, J.F.M., Loeffen, G., Schouten, J., Van Egmond, H., Keukens, H. and Stegeman. H. (1998) Testing of raw milk for tetracycline residues. *Journal of Dairy Science,* 81(9), 2341-2345.

Oliver, S.P., Jayarao, B.M and Almeida, R.A. (2005) Foodborne pathogens in milk and the dairy farm environment: food safety and public health implications. *Journal of Foodborne Pathogen and Diseases,* 2(2),115-129.

Pandey, G.S., Misra, D.S., Mule, D. and Mubita, C. (1996) Studies on sanitary quality and cell count of raw milk from dairy farms supplying milk to Dairy Produce Board in Lusaka, Zambia. *Bulletin of Animal Health Production in Africa,* 44, 9-13.

Raubertas, R.F. and Shook, G.E. (1982) Relationship between lactation measures of somatic cell concentration and milk yield. *Journal of Dairy Science,* 65, 419-425.

Rutaro, H. (2004) Milk quality analysis in southwestern Uganda. MSc Thesis submitted to Kansas State University.

Salman, A. and Elnasri, H.A. (2011) Somatic Cell Count, Total Bacterial Count and acidity properties of milk in Khartoum State, Sudan. *Journal of Cell and Animal Biology, 13 (4).* 223-230.

Sana, O., Yagoub, N., Awadalla, E. and Ibtism, E.M.E. 2005. Incidence of some potential pathogens in raw milk in Khartoum North- Sudan-and their susceptibility to antimicrobial agents. *Journal of Animal Veterinary Advances,* 4(3), 341-344.

Seymour, E.H., Jones, G.M. and McGilliard, M.L. 1988. Persistence of residues in milk following antibiotic treatment of dairy cattle. *Journal of Dairy Science,* 71 (8), 2292-2296.

Sharma, B. K., Bhandari, M. R., Prasai, T. P. and Anal, A. K. (2011) Study on the prevalence of antibiotic residues in milk samples collected from Kathmandu Valley in Nepal. *Nepalese Veterinary Journal,* 30 (1), 137-142

Shitandi, A. and Kihumbu, G. (2004) Assessment of the California mastitis test usage in smallholder dairy herds and risk of volatile antimicrobial residues. *Journal of Veterinary Science,* 5(1), 5-9.

Smith, K.L. and Hogan, J.S. (1999) Quality and safety of raw milk and its impact on milk and milk products. *Bulletin – International Dairy Federation, pp 16-18,* ISSN: 0259-8434.

Tetzer, T.A.D., Benedetti, E., Guimarães, E.C. and Peres, R.F.G. (2005) Prevalence of antibiotic residues in raw milk samples in the region of Minas, Brazil. *Hig. Aliment,* 19,69-72**.**

The Food and Drugs Act. Government of Zambia Statutory Instrument No. 90 of 2001.

The Foodstuffs, Cosmetics and Disinfectant Act. Government of South Africa Act No. 54 of 1972.

Yambayamba, K.E.S. and Zulu, M.P. (2011). Influence of the milking environment on the microbial quality of raw milk produced by smallholder farmers in Magoye. *UNZA Journal of Science and Technology,* 15(1), 37-43.