DETERMINATION OF AMOXICILLIN RESIDUE IN COW MILK

S. K. Singh, Y. P. Sahni, R.P.S. Baghel and S. R. Qureshi

Department of Veterinary Pharmacology and Toxicology

College of Veterinary Science and A.H., Jabalpur (M.P.)

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ABSTRACT

Antibiotics used as therapeutic agents or as feed supplement in milch animal lead to secretion of their residues into milk. Milk samples collected at day-0 (control), day-1, day-3 and day-5 after single IM administration of amoxicillin in 12 lactating cows showed significant level of residual concentration of amoxicillin on day-1. However, similar animals did not show any residual concentration of amoxicillin in milk samples collected on day-3 and day-5. The effect of pasteurization (LTLT) at 65^0C for 30 min on amoxicillin residue in milk was also evaluated. No significant ($p>0.05$) reduction of amoxicillin residue in milk was found on pasteurization.

KEY WORDS: Amoxicillin, Pasteurization (LTLT), Milk, Residue

INTRODUCTION

Antibiotics are widely used to treat numerous bacterial infections both in human beings and animals. Frequent use of antibiotics results in appearance of antibiotic residues in the form of either the parent compound or their metabolite that accumulate within the cells, tissues, organs and also in edible products of animal origin such as milk, egg and meat. Maximum level of residue has been recognized worldwide by various public authorities (Kempe and Verachterf, 2000). Antibiotic contamination of milk may be due to intramammary infusions of drugs for mastitis treatment. Therapeutic agents or as feed supplement in milch animal lead to secretion of their residues into milk (Booth,1982). These residues not only create problems in dairy industry but also have immense public health significance. Hence, the present study was undertaken to evaluate the residual concentration of amoxicillin in cow milk.

MATERIALS AND METHODS

Experiment design: A group of 12 healthy lactating cows (400 kg b.w.) were included in the present study. The animals were treated with a single intramuscular administration of Amoxil (a commercial formulation of amoxicillin, corresponding to a dose of 10 mg/kg b.w. of animal) once in a week for 6 months. The milk samples were collected before (day 0) and after administration of antibiotics on day-1, day-3, and day-5 . Milk samples were stored at -20^0c until HPLC analysis.

Instrument and chromatographic condition: Analysis of Milk samples was performed on a HPLC system (SHIMADZU, SPD-M10A, JAPAN) . The solvent system was: (A) Acetonitrile/ (B) 10 mM Ammonium acetate, PH- 5, the eluting system was 10% (A) / 90% (B) with a flow rate of 1 ml/ min at 220 nm wavelength .

Extraction of amoxicillin from Milk: Milk samples were subjected to liquid phase extraction according to the modified method described in catalogue of Chromatopak (2006-07) with slight modification and final 20 µl volume was injected into the HPLC column. In order to evaluate the recovery of method, blank milk samples (day-0) were spiked with 25, 50 and 100 ppm of amoxicillin. The recoveries were 78.32%, 81.29% and 88.54%.

RESULTS AND DISCUSSION

All animals treated with amoxicillin showed significant level of residual concentration of amoxicillin

on day-1 (post-treatment), however, in similar animals the residual concentration of amoxicillin was below detection limit in milk samples collected on day-0 (pre-treatment), day-3 and day-5 (post treatment). The mean residual concentratrion of amoxicillin on day-1 was 86.9 ppb. Riediker et al. (2001) determined residual concentration of amoxicillin more than MRL (Maximum residual limit) in milk samples which is in close conformity to the findings of the present study. Khaskheli et al. (2008) analyzed 137 unprocessed milk samples for amoxicillin residual concentration. Out of 137 milk samples, 50 samples were found positive for amoxicillin. The mean residual concentration of amoxicillin in milk after pasteurization (LTLT) was 85.85 ppb. No significant difference ($p<0.05$) was found in mean residual concentration of amoxicillin between non-heated (86.9 ppb) and heated (85.85 ppb) milk.

The residual concentration of amoxicillin in all milk samples of day-1 was beyond the maximum residual value (10ppb) as stated by Food and Drug Administration and the Japan Food Chemical Research Foundation. However the residual concentration of amoxicillin in milk samples of day-0, day-3, and day-5 were found below detection limit. The findings substantially indicated that a withdrawal period of at least 24 hrs is needed after administration of amoxicillin and thereby milk from such animals should not be used for human consumption for 24 hours post-treatment. The finding was in agreement with many studies relating with heat stability of other antibiotics which showed that pasteurization (LTLT) did not destroy antibiotic residue.

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