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SEWAGE ANALYSES FOR ANTIBIOTIC RESISTANCE WITHIN FECAL *E. COLI* ISOLATES

A THESIS PRESENTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN MICROBIOLOGY AT MASSEY UNIVERSITY, NEW ZEALAND

PAZILAH IBRAHIM

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ABSTRACT

This investigation was undertaken to explore possible surveillance methods which might be applied in surveys of the incidence of acquired antibiotic resistance in fecal bacteria being shed by an urban population; the Palmerston North City sewage system served as a sampling device. Fecal *E. coli* was used as an indicator organism by virtue of its inherent sensitivity to several relevant antibiotics and, further, by virtue of the fact that antibiotic resistance in this microorganism can, in general, be attributed to plasmids coding for the resistance character(s).

In the course of these exploratory studies it was observed that fecal *E. coli* accounted for 6 to 14% of the total coliforms present in sewage samples; the number of fecal *E. coli* in any given sewage sample was affected by the flow rate of the sewage and the rainfall.

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INTRODUCTION

Microbial resistance has been described as the ability of a microbial cell and its progeny to survive and multiply under environmental conditions that would inhibit or destroy other organisms (Tepper, 1969). Recognised to be present in nature long before the introduction of antibiotics in chemotherapy, bacterial resistance to drugs have received increasing attention as the list of resistant organisms has continued to expand ahead of the rapid development of newer antibiotics (Takafuji, 1977).

1.1 DEVELOPMENT OF DRUG RESISTANCE

Resistance to drugs e.g. to arsenicals by trypanosomes, was known for many years, but it was not until 1935 that bacterial resistance was recognised as a growing problem. In the 1930s, *Neisseria gonorrhoea* infections were adequately treated with sulphonamides, but by the late 1940s greater than 80% of the strains were resistant to 100 mg% or more (Takafuji, 1977). Other organisms such as *Strep pyogenes*, *N. meningitidis* and members of the *Enterobacteriaceae* also developed similar resistance to sulphonamides, rendering a once very satisfactory antibacterial drug ineffective in the treatment of serious infections.

Presently, the factors contributing to the emergence and spread of antibiotic resistance include the following: -

a) Antibiotic use in man

The appearance of antibiotic-resistant strains of bacteria is closely linked to antibiotic use in the treatment of human infections (Mouton et al, 1976). Resistance may appear rapidly or slowly, depending on the organism concerned, the volume and type of antibiotic used, and the method of application.

The most clear-cut evidence relating antibiotic use and resistance has come from hospital studies in which outbreaks of resistant nosocomial infections were related to the extensive use of antibiotics, e.g. infections with *Klebsiella* related to the use of ampicillin in a neurosurgical unit (Price and Sleigh, 1970), infections with pseudomonads resistant to carbenicillin in a burns unit (Lowbury et al, 1972), and resistant infections with *Serratia* associated with the use of gentamicin in an intensive care unit.

In community infections, data from Japan showed that a rise in tetracycline resistance in *pneumococci* was closely associated with an increase in tetracycline use. Again, there was a sharp increase in macrolide use in Japan from 1967 onwards, the quantities used had risen from 50,000 kgs/year to almost 200,000 kgs/year by 1973. Macrolide-resistant strains of group A hemolytic *streptococci* were first recognised in Japan during the early 1970s and, by 1974, 75% of the strains isolated were resistant to erythromycin and linconcycin. Resistance to the tetracyclines and that to chloramphenicol were at a level of 90% and 75% respectively (WHO Technical Report, 1978).

b) Antibiotic use in animals

The use of antibiotics as feed additives for growth promotion is widespread (Linton, 1977; Hartley & Richmond, 1975). The commonest drugs currently used for this purpose are the tetracyclines - one of the most potent agents for provoking the emergence and selection of resistance plasmids and at the same time a very useful therapeutic agent.

The rapid emergence and spread of drug-resistant *Salmonellae*, during the 1960s resulted from antibiotic use in animals (Anderson, 1968). Transmission of these *Salmonellae* to man resulted in many human infections, and

the resistances of such strains, plasmid borne and chromosomal, were acquired in the animal host.

c) The role of food in the spread of antibiotic-resistant bacteria

The presence of antibiotic-resistant nonpathogenic enterobacteria in food is of public health significance. Although some food containing resistant bacteria is decontaminated during cooking, before being cooked it may contaminate other cooked or uncooked food in the kitchen and so transmit drug-resistant entero bacteria to man. Multiresistant nonpathogenic bacteria may add to the number of drug-resistant bacteria, with their plasmids, in the human intestine, but when the organisms concerned are pathogenic they may also produce foodborne diseases (WHO Technical Report, 1976).

d) The role of sewage and surface waters

Sewage and surface waters contribute to the distribution and circulation of resistant organisms. They represent a natural medium in which R-plasmid transfer can occur under certain physical, chemical or biological conditions. Sewage and surface waters contain resistant bacteria from human and animal wastes and can be regarded as a source of all plasmid types, which circulate and are selected under appropriate environmental conditions. These resistant bacteria from sewage and surface waters can be transferred under some circumstances to food and drinking water, which leads to a recycling to man and animals (Smith, 1970; Richmond, 1972).