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Original Article

DNA Vaccine for Bacterial pathogen *Escherichia coli*

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Abstract

Virulent *E.coli* cause mainly diarrhea and vomiting. It is also main cause for urinary tract infection. Still there is no good vaccine for human infections. In this attempt, DNA vaccine was developed against *E.coli* infection. In this work, killed vaccine, Genomic DNA vaccine, Plasmid DNA vaccine, single digested plasmid DNA and double digested Plasmid DNA were used as vaccine. One control treatment was always maintained. Based on these trails, plasmid DNA vaccine, Hind-III digested DNA and Pst-I +Hind-III double digested DNA shows Maximum cellular immune response. So it is concluded that, above said DNA preparations are highly suitable for *E.coli* DNA vaccine development.
Keywords: *E.coli*, Vaccine, Hind-III, Pst-I, EcoR-I.

Introduction

Escherichia coli causes severe cramps and bloody diarrhea. *E.coli* was discovered by German pediatrician and bacteriologist Theodor Escherich in 1885 (Feng *et al.*, 2002) It is more common during the summer months and in northern states. Symptom starts about 7days after infection with the germ. The first sign is severe abdominal cramps that starts suddenly. After few hours, watery diarrhea starts, then the diarrhea changes to bright red bloody stools. The infection makes sores in intestines. So the stools become bloody diarrhea lasts for 2 to 5 days.

The most common complication is called Hemolytic uremia syndrome, it leads to hemolytic anemia, thrombocytopenia and renal failure. Hemolytic uremic syndrome is more common in children. It can cause acute renal failure in children. This problem starts about 5 to 10 days after the diarrhea starts. The virulent strains can cause serious illness or death in the elderly, the young or the immuno compromised (Hudault *et al.*, 2001). It is also mainly responsible for 90% urinary tract infections. It is normally spread in contaminated food and water.

It also transmitted by flies (Szalanski *et al* 2004., Alam and Zurek,2004) as well as direct contract with farm animals (Rahn *et al.*, 1998; Trevena *et al.*,1999) and air born particles found in animal rearing environments (Varma *et al.*,2003). Still the vaccine research is going on. In this attempt, try to develop advanced DNA vaccine for *E.coli* infections in human beings.

Materials and Methods

The *E.coli* pathogen was collected from patient's sample in local hospital then culture and sub cultured in our Lab. Regular Biochemical and Microbiological tests have been done for confirmation. In this attempt, three experimental trials were conducted. In the first trial, three treatments (Killed pathogen, Genomic DNA and Plasmid DNA) and one control was maintained. In the second trial, three single digested Plasmid DNA treatments (Pst-I, BamH-I and Hind-III digested Plasmid DNA) were maintained. In the third trial, three double digested Plasmid DNA treatment (EcoR-I + Pst-I,EcoR-I + Hind-III and Pst-I + Hind-III digested Plasmid DNA) were maintained. Medox kit was used for all enzyme digestion and isolation of Genomic DNA, Plasmid DNA from *E.coli*. Albino rat was used as experimental animal, after the preparation of all the treatments, they were provided through intramuscular injection, After one week of first injection, booster dose (second dose) were given, then one week later, blood samples were collected and cellular immunity was analyzed.

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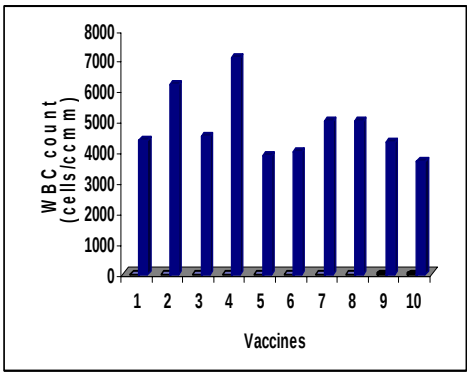
Results and Discussion

Escherichia coli mainly cause Vomiting and Bloody diarrhea and it also produce various illnesses to human beings and it is also mainly responsible for 90% of urinary tract infections. Researchers have actively been working to develop safe effective vaccines to lower the world wide incidence of *E. coli* infection (Girard *et al*.,2006). Ahemed *et al*., (2006) reported polysaccharide based vaccine for *E. coli* infection. In the same year Fort Dodge Animal Health introduce an effective live attenuated vaccine for chicken (Wattpoutry.com).



1. Control, 2.killed vaccine 3.Genomic DNA, 4.Plasmid DNA, 5.Pst-I digested plasmid DNA, 6.BamH-I digested plasmid DNA,7.Hind-III digested plasmid DNA, 8.EcoR-I+Pst-I digested plasmid DNA, 9.EcoR-I+Hind-III digested Plasmid DNA,10.Pst-I+Hind-III digested Plasmid DNA.

Fig.1: Various DNA vaccine influence on WBC count of Albino rats

In January 2007, the Canadian Biopharmaceutical company Bioniche announced it has developed a cattle vaccine (pearson, 2007. www.ctv.ca., www.cnxmarketlink.com). Still there is no advanced vaccine for *E.coli* infection in human beings. In this attempt, DNA based vaccines was developed for *E.coli* infection in humans. In this work, three trials have been done. In the first trial, maximum leukocyte response was observed in Plasmid DNA vaccine.

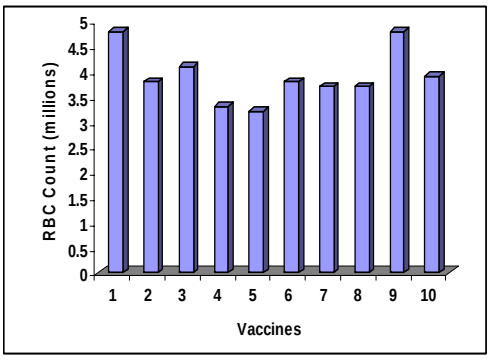


Fig.2:Various DNA vaccine influence on RBC count of Albino rats

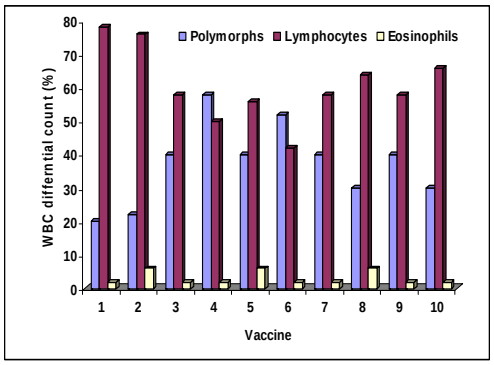
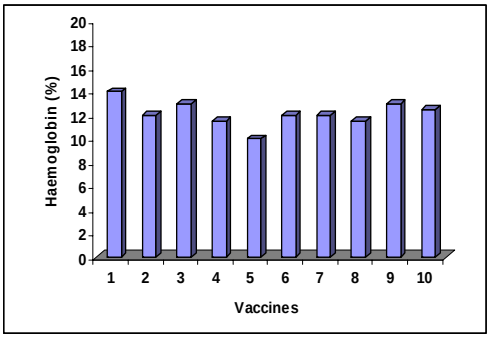


Fig.3: Various DNA vaccine influence on WBC differential count of Albino rats



1.Control, 2.killed vaccine 3.Genomic DNA, 4.Plasmid DNA, 5.Pst-I digested plasmid DNA, 6.BamH-I digested plasmid DNA,7.Hind-III digested plasmid DNA, 8.EcoR-I+Pst-I digested plasmid DNA, 9.EcoR-I+Hind-III digested Plasmid DNA,10.Pst-I+Hind-III digested Plasmid DNA.

Fig.4: Various DNA vaccine influence on haemoglobin levels of Albino rats

In the second and third trials, maximum cellular immune response were observed in Hind-III digested Plasmid DNA and double digested (EcoR-I + Pst-I) Plasmid DNA compare to all other treatments. Based on these trials, it is concluded that, whole Plasmid DNA, Hind-III digested Plasmid DNA and double digested DNA (EcoR-I + Pst-I) are highly suitable for develop a best vaccine for *E.coli*.

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