recombinant parasites were generated. The intensity level of fluorescent was monitored in different phases of parasites and *ex vivo* system by both fluorescence microscopy examination and Flow-cytometry analysis. Western blot and RT-PCR analysis were also performed to show specifically the expression of cpa/*cpb/egfp* fused gens in both lines of transgenic parasites. Results:The presence and correct location of genes inside the transgenic parasite genome was done at molecular level (both by DNA and RNA). The expression of triple-fusion(CPA/CPB/EGFP) was confirmed at protein level using western blot and EGFP intensity by flow cytometry in both recombinant parasites. In logarithmic phase, the intensity of EGFP was proportional to amplification of parasites and was 20 fold higher in compare to stationary phase. After 5 days when the culture is near to stationary phase, the level of intensity decreased to 2.56-11.04% in different concentration of G418. One of the hallmarks of episomally expression is their instability without any drug pressure *in vivo* and copy number of plasmids decreases rapidly. In addition, the copy number of plasmids between transfectedparasites are unequal and expression rate within parasites are severely different. Conclusion:Recombinant *L. tarentolae* expressing *cpa/cpb/egfp* fused genes with two different approaches were established. Our data suggest that it is more secure to use the stably transfected*L. tarentolae* for further vaccine studies.
Keywords: *Leishmaniatarentolae,*Episomal, Stable Transfection,Vaccine Candidate Antigens

923. *In silico* Analysis of Chimeric Recombinant Vaccine Against Causing Diarrhea Agents
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Background: Entericinfectionresultingindiarrheal diseases remains health problem worldwide. *Shigella* and *Escherichia* causes the most diarrheas in the world. *Shigella* can cause bacterial dysenteries and shigellosis through invasion. One of the most effective proteins for pathogenesis is invasion plasmid antigen C (IpaC). Other bacteria like ETEC and EHEC are also causing diarrhea. Most of the travelers' diarrheas are resulted from ETEC infection. Colonization factor antigen I (CFA/I) is critical virulence protein for these infections with two subunits called CfaB and CfaE. Another major pathogen is EHEC which produces intestinal disorders. Attachment of bacteria is the main step of infection andthe protein intimin plays the key role in this function. Protection against the vast majority of responsible pathogens of diarrheas, requires the developed the polyvalent vaccine against *Shigella*, ETEC and EHEC. *In silico* techniques are best tools to study, design and evaluate of new vaccines. In the present study, we designed a multisubunit protein that could be a suitable vaccine candidate against thesepathogens. Materials and Methods: A synthetic chimeric gene (CII) containing immunologically significant parts of CfaB, IpaC and Intimin proteins was designed. After codon optimization for *Ecoli*, modeling was done to study the 3D structure of the protein. To assess immunological points, the humoral and cellular immunity was analyzed. Prediction of allergens and mapping of IgE epitopes was obtained. Results: The bioinformatic analysis showed that each domainfoldedseparately in protein structure. CII had many T and B cell epitopes in both linear and 3D structure. This prediction chimeric construct had the potential to induce CD^{4+} and CD^{8+} immune responses against these pathogens. In addition CII could be accessible to surveillance by the immune system in mouse and human. Conclusion: *In-silico* analysis showed that this chimeric protein can be used as a candidate vaccine against *Shigella*, ETEC and EHEC.
Keywords: *In silico,* Chimeric Recombinant Vaccine, Diarrhea Agents

924. Immunological Evaluation of a Cocktail Vaccine againstEnterotoxigenic *Escherichiacoli*
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Background:Enterotoxigenic*Escherichia coli* (ETEC) is the most significant agent leading to childhood diarrhea and death in developing countries. Due to its prevalence as well as difficulties in its treatment, designing effective vaccines against ETEC is a goal of World Health Organization. Colonization factors such as CfaB and CfaE as major and minor subunit of fimberiae, respectively; have a vital function in bacterial binding to epithelium cells.Heat labile enterotoxin (LT) B subunit is nontoxic subunit of LT molecule that plays an important role in ETEC pathogenesis. Hence, these molecules alone or together with other candidate molecules have been considered in vaccine design.In this investigation, we produced recombinant LTB, CfaB and CfaE in *E. coli* with the mean of examine its immunogenicity as a cocktail vaccine.Materials andMethods:The cfaB, cfaE and ltb genes were isolated from a local isolated ETEC, separately cloned and expressed using pET28a expression vector. The recombinant proteins was purified and used as antigens for mice immunization and in immunological tests. Results:The immunological analyses showed production of high titer of specific antibody in immunized mice. Anti LTB Antibody could bind to whole toxin and neutralize the toxin through inhibition of its binding to the Ganglioside M1 receptor. Relying on agglutination inhibition experiment, anti-CfaB and anti-CfaE serum was able to block the binding of CFA/I fimbriated ETEC to erythrocytes.Conclusion:Considering the LTB, CfaB and CfaE roles in ETEC pathogenesis, they can be use ascocktail vaccine against ETEC.
Keywords: Cocktail Vaccine, Enterotoxigenic *Escherichiacoli*

VETERINARY IMMUNOLOGY

Oral Presentation

925. MicroRNAs; Novel Interferon-Induced Gene Regulators
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Background: MicroRNAs (miRNAs) are a new class of 18 23 nucleotide long noncoding RNAs that play essential roles in a wide spectrumofbiological processes. Recent reports also clarify the role of miRNAs ascritical effect or sin the complicated host pathogen interaction networks. Emerging evidence suggests that miRNAs play a key role in the regulation of immunological functions, including innate and adaptive immune responses. Materials and Methods: In the present study, we use microarray technology to identify up regulation of miRNAs in Rainbow Trout after exposure to the fish pathogenic rhabdovirus causing Viral Haemorrhagic Septicaemia (VHS) which is an economically important disease in freshwater aquaculture.
Results & Conclusions: we discuss the mechanism of interferon induced miRNAs in Rainbow Trout infected by thisvirus.
Keywords: micrornas, Interferon, Rainbow Trout, Viral haemorrhagicsepticaemia Virus.

926. Molecular cloning and expression of the constant region of chicken μ immunoglobulin chain in *Escherichia coli*
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