The current search on the carrier potential of *P. americana* of bacterial pathogens in the intestine tempted to revise the conventional concept of this insect as a nuisance pest to a harmful vehicle of human bacterial pathogens. The bacteriological examination of the intestinal content of these cockroach species revealed the presence of various potential bacterial pathogens including *Listeria* species. On analyzing the bacterial isolates, the major groups of bacteria with epidemiological significance in nosocomial infection were noticed to be *Enterococcus* spp., *Klebsiella* spp., *Proteus* spp., and *P. aeruginosa*. Enterococci were the predominant bacteria isolated (95.6%) with *E. faecium* forming the major species. 51.2% of cockroach samples were found to be harboured with *Listeria* species with *L. grayi* being the dominant species. *L. innocua* and *L. monocytogenes* were the other *Listeria* species isolated. When the incidence of isolates of various bacterial species from *P. americana* in relation to source of collection was assessed, *E. faecium*, *E. faecalis*, *S. epidermidis*, *K. pneumoniae* and *P. aeruginosa* were found to be more in samples captured from hospitals indicating the ability of this insect to disseminate potential pathogens in hospital environment. The hospital samples also formed the source for the only two isolates of *L. monocytogenes* which is to be viewed with caution as infection with this bacterial species is reported to be more common in immunocompromised individuals.

The virulence studies conducted in *Listeria* isolates include its haemolytic activity, phosphatidylinositol phospholipase production,
resistance to lysozyme, serum, bile and biofilm formation potential. The haemolytic and phosphatidylinositol phospholipase activity observed in *L. monocytogenes* phenotypically were further confirmed by genotypic methods by detecting *hlyA* gene encoding the haemolysin and *plcA* gene encoding phosphatidylinositol phospholipase C indicating the ability of this bacterial isolate for intracellular survival and cell to cell spread in human beings. These virulence markers were, however, absent in *L. innocua* and *L. grayi*. Though lysozyme resistance was not noticed in *L. innocua* or *L. grayi*, the resistance was obvious in *L. monocytogenes* showing its ineffectiveness as a food preservative against this bacterial species. The ability to resist antibacterial activity of human serum shown by all *Listeria* isolates suggests their resistance to the bactericidal effect of the serum and thereby its pathogenic potential. All the *Listeria* isolates under study irrespective of the species presented tolerance to bile indicating their potential to survive the conditions prevailing in the intestinal tract.

On evaluating the biofilm formation potential, all the *Listeria* strains irrespective of the species has been observed to be forming biofilm under natural conditions. The optimum biofilm formation at 37°C of *L. monocytogenes* noticed in the study shows the possibility of this bacterial species forming biofilm on indwelling medical devices allowing their survival in hostile environments. The ability of *L. monocytogenes* for forming biofilm, though weak, at lower temperatures of 4°C and 22°C demonstrates the ability of this emerging pathogen to adapt and establish in a wide range of environmental conditions. In the current study all the three species of *Listeria* were capable of producing biofilm at pH 5.5, 7.5 and pH 8.5. Though biofilm formation was observed to be formed in alkaline pH conditions (pH 8.5), an increase in biofilm production was noticed when the
Listeria strains were exposed to acidic pH of 5.5. The biofilm formation was noticed to be increased with moderate levels of glucose and sodium chloride indicating the possibility of biofilm formation on equipments associated with food industry and patient care.

The Listeria species in the current study appears to be relatively resistant to various food processing practices such as growth at low temperature, tolerance to sodium chloride and survival at mild acidic pH. The study noticed the ability of all Listeria strains to grow at 4°C. Its ability to grow at low temperature may be a health concern as far as chilled foods particularly which are consumed without further processing. The observation of L. innocua and L. grayi to survive better at 4°C will be relevant if these strains were proved to have food spoilage potential. Hence further studies on their food spoilage potential would be of public health importance. When the tolerance of Listeria isolates to sodium chloride was assessed, their survival at high concentrations was observed. Previous studies noticing increased invasive ability of L. monocytogenes on short term NaCl adaptation and the protective effect of sodium chloride on this bacterial species during thermal treatment of food products enabling its survival is a cause of concern. Therefore the use of sodium chloride as a food preservative may not be dependable as far as Listeria contamination is concerned. On analyzing the survival of Listeria at different pH, the survival of these bacterial species at pH 5 was noticed suggesting their ability to sustain in conditions associated with low acid foods where organic acid is used as food preservative. The survival of Listeria isolates at low pH may also enable them to endure the acidic pH of the stomach during its passage along with food. The ability to survive at pH 8.5 as noticed in the study suggests their persistence in hospital or food processing environments where the decontamination procedures
using alkaline detergents are followed. The alkali tolerance may also enable them to survive pancreatic secretions and the alkaline phase of phagocytosis so as to establish human infection. The survival of *Listeria* in dry conditions noticed in the study indicates the likelihood of these bacteria to act as a possible source of infection or contamination in health care facilities or catering establishments. Moreover, the chances of surviving *Listeria* to form multispecies biofilm with other bacteria in these environments add significance to this finding.

The resistance to heavy metal noticed in *Listeria* isolates contribute to their persistence in the environment. The detection of plasmid borne *cadA* gene in *L. monocytogenes* isolate and the loss of cadmium resistance noticed on plasmid curing in *L. monocytogenes* isolates suggests the role of plasmid in conferring cadmium resistance. Since benzalkonium resistance was also noticed in all *Listeria* strains under study the possibilities of their plasmid mediated co-dissemination to the co-existing bacterial flora with pathogenic potential cannot be underestimated.

Regarding the susceptibility/resistance of various bacterial isolates towards different disinfectants, varying degrees of susceptibility among the isolates was noticed. *P. aeruginosa* isolates presented more resistance than other bacterial isolates. All the isolates were noticed to be susceptible to low concentrations of glutaraldehyde. The current study reveals that the bacterial isolates from *P. americana* showed no considerable difference in susceptibility to various disinfectants compared to the observations made earlier studies on various nosocomial pathogens. When the comparison of planktonic cells and biofilm of *Listeria* has been made in terms of their susceptibility to different disinfectants, biofilm cells showed more resistance to all the disinfectants than their planktonic counter parts under study. The
overall analysis of susceptibility of bacterial isolates towards various disinfectants indicates that there is no substantial difference in the disinfectant susceptibility profile of *Listeria* isolates in comparison with other bacterial isolates with the exception of benzalkonium chloride which showed poor efficacy on *Listeria* species in lower concentration.

On considering the antibiotic resistance among the predominant nosocomial pathogens isolated from *P. americana*, resistance appeared to be more pronounced in *E. faecium, K. pneumoniae, P. mirabilis, P. aeruginosa* and *L. grayi*. A general feature observed with all pathogens under study was their tendency to resist the cephalosporins. 16% of *E. faecium* were noticed to be resisting vancomycin posing a risk of transferring the same to other bacterial flora inhabiting the intestine through horizontal gene transfer. The carrier potential of cockroaches of multi resistant *Listeria* species including *L. monocytogenes* in the intestinal tract as noticed in this study contribute to the dissemination of this food-borne pathogen in hospitals, food processing establishments and in domestic environments. Although multi resistant strains of *Listeria* spp. are rare in nature as reported in earlier studies, the results obtained in the current study provide an evidence of the appearance of multiple resistance in this bacterial species. This is a concern particularly when the pathogenic *L. monocytogenes* is taken into consideration. Multiple resistance was found to be more predominant in *L. grayi* isolates with 83.8% showing resistance to more than five antibiotics. Results obtained in the conjugation study shows the transfer of plasmids with ampicillin resistant genes from *L. grayi* to *E. faecalis* with the conjugation frequency of $10^{-6}$ to $10^{-7}$. This signifies the potential of *L. grayi* inhabiting in the intestine of the insect to transfer resistance through plasmid to other co-existing bacterial species.
This study demonstrates the possible role of cockroaches in the dissemination of bacterial pathogens including *Listeria* species in domestic and peridomestic environments warranting provision for an effective and integrated pest management system. The ability of *Listeria* isolates resisting various physicochemical conditions opens a new area of study of this bacterium giving emphasis on its food spoilage potential. The multidrug resistant bacteria carried by the omnipresent insect cockroach as noticed in this study urges the necessity of further epidemiological studies for revealing the role of this insect in nosocomial infection and food spoilage.

The observations made in this study establish the possible role of the insect *P. americana* in carrying and transmitting human pathogens especially in nosocomial and food borne infections and goes to suggest that the pest cannot be ignored as a casual harmless inhabitant of the human environments.