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A REVIEW ON FOOD BORNE ILLNESS CAUSED BY STAPHYLOCOCCAL AUREUS

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Abstract:

Staphylococcus aureus is a major human pathogen that may produce a variety of toxins causing food poisoning and various infections in animals and humans. Food poisoning due to bacterial toxins can caused by the ingestion of exotoxins, which are preformed in the food, or by the ingestion of food contain large numbers of bacterial cells which then release endotoxins in the gastrointestinal tract. The pathogenic microorganism of public health importance that may be transmitted through contaminated food. Due to its multidrug resistance and virulence, staphylococcus aureus has become a major concern to public health and food safety in many countries. Staphylococcus aureus food poisoning is caused by the ingestion of food containing enterotoxins produced by some strains of staphylococcus aureus. Hemolysis

is one of the virulence factors of Staphylococcus aureus. It is said that staphylococcus aureus may produce three hemolysin designated as Alpha, Beta, Gamma. Staphylococcus is a very common organism capable of producing several enterotoxins (SEs) that cause intoxication symptoms of varying intensity in humans when ingested through contaminated food. Genes encoding hemolysin were amplified with specific primers by using polymerase chain reaction (PCR) technique. The aim of the present review is to obtain the comprehensive understanding of the causes of toxins, a long list of toxin effects and role of hemolysin in the pathogenesis and different biochemical and immunological methods used for detection.

Keywords: Food Poisoning, Bacterial agents, Toxins.

Introduction:

Safe food and water is a public health requirement. safety refers to all the hazards that make food injurious to health. These hazards arise from improper agricultural practices, poor sanitary and hygiene conditions at all stages of the food, lack of preventive controls in food processing operation, misuse of food additives and chemicals colored with inappropriate storage and handling may lead to food poisoning. Specific concerns about food hazards are mainly chemical microbiological, pesticide residues, veterinary drug residues and allergic compounds[1,2].Bacterial food borne pathogens are considered the most frequently implicated biological agents in food poisoning syndrome in humans,often called as food born illness.Food handlers during production and (storage),the obtaining of safe and nutritious food products for the consumers is considered to be a great challenge for food industry,worldwide .

The globalization of food trade increases the potential to spread food borne hazards around the world. Concerns regarding the contamination of food with chemicals and microbial hazards in recent years resulted in banning of imported food products by nation[3].Staphylococcus aureus is an important food-borne pathogen because of its ability to produce a wide range of extra cellular protein toxins and virulence factors that contribute to pathogenecity of the organism4.The pathogenicity of Staphylococcus aurues,is related to the production of a wide variety of exoproteins,including alpha,beta heamolysins which contributes to its ability to cause diseases in many mammalian species5.Eary methods for the assay of bacteria toxins were based on in vivo(i.e.,animal challenge tests) or in vitro tests (I.e.,tissue culture).Later,immunological tests were developed.Molecular biology techniques such as polymerase chain reaction (PCR),real_time PCR and DNA hybridization have been developed and become popular for pathogen identification6.Although these technologies can be detect low number of bacterial cells,they usually need several hours.Moreover,such techniques required prior cultural enrichment and bacterial DNA isolation,preparation of enzyme reaction mixtures and expensive equipment.(7)

The pathogenic microorganisms of public health importance that may be transmitted through contaminated food are Bacteria, Virus, Protozoa, Trematodes, Cestodes and Nematodes. Among these the most important organism which causes food borne poisoning include bacteria Staphylococcus aureus. Staphylococcus aureus is an extremely common pathogenic bacterium that can be found in a number of different food varieties, including mixed food (Pasta dishes, Salads) meat and meat products, eggs products, vegetables, baked goods and cheeses[4].

Due to its multidrug resistance and virulence,staphylococcus aurues has become a major concern to public health and food safety in many countries.(5) Staphylococcus aureus is a factitive aerobic gram + positive coccus ,it is non motile and catalyse and coagulase positive .Cells are spherical single or paired cocci,or form grape like clusters (staphylo means grape in Greek).The staphylococcus cell wall is resistant tp lysozyme and sensitive to lysostaphin,which specifically cleaves the pentaglycin bridge of staphylococcus species some staphylococcus aureus strains are able to produce staphylococcus enterotoxins and are causative agents of staphylococcus food poisoning.(6)

Staphylococcus aureus is considered one of the main causes of food borne intoxication,being widespread bacteria throughout nature.It's food poisoning effect is related to the ingestion of the preform enterotoxin.

TYPES OF HEMOLYSIN:

Staphylococcus aureus is able to produce nosocomial and invasive infection in human.Bacteriologist have sought to describe roles to the many toxins produced by Staphylococcus aurues particularly in relation to pathogenesis and threat to life.It is said that Staphylococcus aurues may produced three hemolysin designated as Alpha,Beta,Gamma.(7)
Causative agents of food-borne outbreaks are recorded:

Table:1

causative agents	Outbreaks [N=530]	Cases [N=6451]	Hospitalization [N=872]	Death [N=7]
Salmonella Sp (Enteritis, Typhimurium, Heidelberg & other serotypes)	63.8	47.7	16.8	100
Staphylococcus aureus	16	25.6	17.1	0
Clostridium perfringens	5.1	12.3	0.5	0
Bacillus cereus	2.8	3.7	10.0	0
Histamine	3.8	1.4	30.4	0
Other pathogens (Campylobacter, Sp, Dirophysis, C.botulinum, Shillelagh Sp, Calci virus, HAV, Vibrio Sp, E.coli Sp)	8.5	9.2	7.6	0

(8)

The types of food incidences and number of people effected are described in table:2.

TABLE 2:

Place	Incidences	No of people affected	Microorganisms	Food
Party	3	98	Salmonella	Veg-food (Coconut balls, Fish, Sand

			Paratyphoid A Var durazoo S.aureus	witch)
School	2	135	Staphylococcus aureus	Soybean milk & bhalla

(10)

The percentage of contamination seen in some of the few food materials are seen in frequent food materials such as meat, Khoa are seen in table 3.

Table:3

Type of food	Bacteria	% of contamination
Meat	Staphylococcus spp	21
Khoa	Staphylococcus spp	20-30

(11)

3.DETECTION METHOD

Immunological assays are are simpler and cheaper than biological assays and have therefore been widely adopted. The bacterial toxins in the literature and commercial sources of kits are depleted in the table.

Table:4

Organism	Toxin	Immunoassay method	REFERANCE
Staphylococcus aureus	SEB++	ELISA	12,13
	All enterotoxins	ELFA	14,15
	SEA, SEB	EIA	16,17
	SEA, SEB, SEE	ELISA	12,13
	SEA	LAMP	18

Staphylococcal food poisoning is caused by the ingestion of food containing enterotoxins produced by some strains of Staphylococcal aureus To date, seven serologically distinct enterotoxins haven been identified and characterized (A,B,C1,C2,C3,D&E).

The SET-RPLA kit (Oxide) is a latex-based immunological test for the simultaneous detection of staphylococcal enterotoxins A,B,C&D in food extracts and culture filtrates.

The TECRA Staphylococcal Enterotoxin Visual Immuno assay(19) (Bio Enterprises Pty Ltd,Rosevilla,Australia) is a rapid microtitre plate based screening test which uses polyvalent anti-sera to detect staphylococcal enterotoxins A-E at concentrations down to 1ng ml-1 in foods and culture filtrates. Results can be read visually with a colour card or by measuring the absorbance at 414nm in a

plate reader.

The VIDAS staphylococcal enterotoxin (SET) assay (20) (bioMerieux, Marcy-l'l Etoile, France) is an automated `enzyme-linked fluorescent immunoassay(ELFA) for the simultaneous detection of staphylococcal enterotoxins A,B,C1,C2,C3,D&E in foods.In this assay, an aliquot of food extract is placed into a reagent strip which is then loaded into the VIDAS instruments.

VIDAS had good sensitivity for enterotoxins A,B,D&E, but was less sensitive for the the C enterotoxins. With artificially contaminated foods, the detection sensitivity ranged from <1ng ml-1 - 1.5ng ml-1, depending on the toxin and food (21).

4.BIOCHEMICAL TEST:

The following biochemical tests and characteristics of bacterial strains were taken into consideration Gram staining,Catalase, Oxidase tests, Colony pigmentation, hemolysin ,tube coagulase test(Rabbit plasma supplied by (22) NCIPD,SOFIA BULGARIA),VP TestONGP(Beta-galactosidase,4mg disk,Hi media,India),test with polymyxin B(300units disk oxoid UK),acid from mannitol determined on manittol salt agar (22) NCIP,Bulgaria),utilization of trehalose (22) NCIPD,SOFIA,BULGARIA) and maltose (23) MKB test, Rosina, Sbvak republic).All test were carried out according to the manufacturers instructions and in compliance with the general bacteriology procedures.

4.1 LATEX AGGLUTINATION: Staphylococcus aurues rapid latex kit (24) (ATLAS MEDICAL,CAMBRIDGE,UK)for the detection of Staphylococcus aurues in culture was used following precisely the instruction of the company.

4.2 ELISA:

ELISA was first proposed by Swedish scholars Engvall and Perimann.The way of ELISA has become a new method for the determination of the title substances (25,26) provided a new idea for microbial detection by the method of ELISA.Nagraj(27) and Zoha et.al (28) established double antibody sandwich method to detect SA enterotoxin G and enterotoxin M.li.et.al(29) used cell fusion technology to prepare anti-SEA monoclonal antibodies, its sensitivity of SEA can be as high as 1.56 mg.the ELISA method is practical ,specific and sensitive ,and its detection range is at mg and pg levels. It can directly detect enterotoxin in solution and is suitable for qualitative test {30,31} .However, the external environment can influence the ELISA detection,and this method has a degree of cross reaction to similar compounds (32,33).

4.3 ELFA:

The results of ELFA are expressed by fluorescence intensity which is more intuitive than ELISA.Hennekinne et al.(34) used this technique to detection limit enterotoxin A to 0.09ng /ml VIDAS system detect SA entertoxins and the minimum detection value is less than 0.25ng/ml (32).ELFA is a highly sensitive and reduces the detection limit and improves the detection sensitivity(30).However,SA and its enterotoxins are inactivated during food processing,which may lead to incorrect of results (35).

4.4 Nucleic acid probe:

Nucleic acid probe technology detects microbes based on the principle of complementary base pairing.Firstly applied to detect E.coil (36) .Now the technology has been successfully applied to detect SA DNA probe technique amplifies target bacteria by designing specific primers based on highly conserved nucleic acid sequence ces (33).Its greatest feature is strong specificity,accuracy and sensitivity.Gao et al.(37)used nucleic gene as probe to detect SA in laboratory animals.The sensitivity of this method can reach 1pg genomic DNA,and the results are consistent with bacteriological tests. Xue

(38) and Chen et al.{39} used nucleic acid probes to detect bacterial colonies respectively minimum of 10^6CFU/ml and 10^7 CFU/ml.The initial level of 10 CFU of SA is detected in the overnight cultures pr-concentrated food samples.Bacteria need to be enriched before detection,which increases the detection time (3).

4.5PCR based on nuclei gene:

DNA was extracted without using a commercial kit.Bacterial suspension were prepared in sterile distilled water, boiled and centrifuged for 5 min at 1200g(22).The concentration and purity of DNA extracts were determined by DNA/RNA spectro photometer Gene Qaunt1300 at A 260/A280.The DNA extracts were stored at -20c until the beginning of the trials .PCR was run as described by Braksetad et al.(22),(43) with an expectorated amplicon size of 270bp.

CONCLUSION

In conclusion the methods developed in the study could be utilize as an effective detection tool for the early on site detection of Staphylococcal aureus in food samples.

This paper reviewed types of toxins produced by staphylococcus aureus and the immunological techniques such as ELISA,EIA,ELFA, LAMP to detect the percentage of toxin contamination in particular foods.

Example:

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