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ENTEROBACTERIACEAE ON SELECTIVE MEDIA

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Abstract

The Enterobacteriaceae family represents the most extensive taxonomic unit that includes 44 types, among which 25 were involved in human pathology. They are gram-negative bacilli of average dimensions, with round heads, with non characteristic disposition generally. In Klebsiella the bacilli are disposed in diplo, in the sense of the length. The species of Yersinia are more frequently coccobacillary, bipolar colored, and the species of Proteus are sometimes extremely polymorphous. They can be mobile or immovable. They don't sporulate. The majority of the enterobacteriaceae are not capsulated. Some can have a prominent capsule, Klebsiella, and other Salmonella, E. coli, can be wrapped by a capsular material. The Enterobacteriaceae are aerobe germs, facultative anaerobe, non demanding from the nutrition point of view. They represent 80% of the total isolated gram-negative bacilli and over 50% of the total of isolated germs. Also, they are involved in the etiology of 40% of the septicemias, in over 75% of the urinary infections and in majority of the food poisoning.

Key words: bacilli, nutritive, coccobacilli

INTRODUCTION

The *Enterobacteriaceae* are catalase positive and oxidase negative. They are chemoautotroph bacteria having both types of metabolism: respiratory and fermentative. Following the fermentation of the glucose is forming an acid with or without gas. Some enterobacteriaceae use fermentation lactose.

The members of the *Enterobacteriaceae family* have a complex antigenic structure. The somatic antigen O (endotoxin) represents the polysaccharidic repetitive terminal units of the lipopolysaccharide (LPS) from the wall of the Gram-negative bacteria. LPS is also formed of a core, similar to all the Gram-negative bacteria and the lipid A, present in all the enterobacteriaceae, which is responsible for their toxic activity in the host body. The somatic antigen "O" is thermostable, resistant to alcohol. It can be detected by reactions of agglutination. It determines, in the host organism, creation of antibodies IgM type. *Enterobacteriaceae* are microbodies that can be found in the environment and/or have evolved to compose the normal intestinal flora. Still, some members needed extra genetic information assured by the plasmids, transposons and bacteriophage, which lead to the appearance of some factors of pathogenicity, these strains becoming pathogen. The additional genetic information can be coded by chromosomes or extra chromosomes.

The *Escherichia* type reunites species present in the human intestine highlighted for the first time in 1885 by the German Theodor Escherich in honor of who was given the name of the type. They were considered commensal bacteria of the large intestine until 1935 when it was demonstrated the etiologic role played by a strain of *Escherichia coli* involved in an episode of diarrhea in newborn. The *Shigella* type, called after the Japanese microbiologist Shiga who discovered these bacteria 100 ago, reunites four species. The *Salmonella* type belongs to the *Enterobacteriaceae* family and includes a single species *Enteric Salmonella*. The French bacteriologist Alexandre Yersin has isolated for the first time the bacteria causing the epidemics of swine fever from Hong Kong from the end of the 19th century.

MATERIAL AND METHOD

We accomplished a prospective study, based on the microbiologic diagnosis registered in the bacteriologic register of the laboratory of medical analysis, S.C. Diaser, Oradea.

For the performing of the study we used also the archive, registered in the specific program of the computer from the laboratory of S.C. Diaser, Oradea, in the computerized data base of the unit, respectively.

Necessary materials for the performing of the examination:

■ A recipient of collection (collection recipient with collecting spoon) with transport medium

- Wooden spatula
- Latex gloves

For the collection of fecal matter it has to be collected a sample of fecal matter of 5-10g introduced in the collection recipient of fecal matter with transport medium. If the stool is liquid, it will be collected 5 ml. It is recommended to be chosen a liquid, mucous and bloody portion, if there is one. Don't collect quantities larger than 10g because it will reduce the chances of isolating the pathogen bacteria.

RESULTS AND DISCUSSIONS

The weakly selective media, MC, EMB, allow the growth of all the equally lactose-positive and negative enterobacteriaceae, even of other groups of gram-negative bacilli as *Vibrio*, inclusively *V.cholerare*, *Pseudomonas*, *Aeromonas*, *Plesimonas*, *Alcaligenes*.

A special importance is represented also by the conditioned isolation of the enteric pathogens, especially *Escherichia Coli* in groups, represented by Enteropathogenic *E.Coli* (EPEC), enterohemorrhagic (CHEC), enterotoxigenic (ETEC), enteroinvasive (EIEC), enteroadherent (EAEC), enteroaggregative (EagEC).



Fig1. E. Coli, lactase-positive colonies, metallic green.

Another important role is represented by the estimation of the lactose negative flora/ lactose positive flora report, in regard to *E.Coli* or other lactase-positive, especially *Klebsiella* in children.

The moderate selective media have a higher selective capacity, considerably inhibiting the lactase-positive enterobacteriaceae. Thus, they allow the unhindered development of the lactase-negative enterobacteriaceae as would be: Salmonella, Shigella, Providencia, Proteus, Morganella and the late lactase-positive, Citrobacter, Serratia, Hafinia. Enterocolitis Yersinia is hardly developed, usually after the prolonging of the incubation 22-29°C maximum 24 hours, and on some media, as ADCL, XLD, IM, SMID is strongly inhibited. These media are usually used for the isolating of the pathogens of the Salmonella and Shigella type. Between these, the RA and SMID media, named chromogenic media, allow the absolute differentiation of the Salmonella type by distinctively colored colonies. Particularly the IM medium is recommended also for the isolation of the Pseudomonas type.

The highly selective media are inhibiting for all the groups of enterobacteriaceae, with the exception of the salmonella. The WB medium, chosen for the development of *Typhi* serotype, is recommended especially for the investigation of the suspects of typhoid fever and the chronic bearers. The AVB medium, partially inhibitor for the *Typhi* and *parathipy A* serotypes is recommended for the control of food products.

A remark is that, from all the media of enriching you can pass on any of the moderate selective media.

In the last years, it was extended very much a category of different selective media using the antibiotics and sulfonamides as pressing factors. The advantages to dose with precision the quantities for the associated flora and the more reduced price are taken into consideration in promoting these media.

The more decreased productivity of the weakly and moderate selective media on which Y. enterocolitica, can be isolated after an additional incubation of 24 hours on 22-29°C, at the room temperature, has promoted for these enterobacteriaceae different media. Among these the medium proposed by Schleman, cefsulodin-irgasan-novobiocin - Y. Enterocolitica (CIN-Ye) is more frequently used due to its advantages, as would be the fact that it is easily to be prepared, fast reading, in 18-24 hours at 32° C, the good selective capacity and it is relatively cheap. The Y.enterocolitica colonies are red with the dark red center and the transparent margins. Other enterobacteriacae, liquefaciens, Citrobacter freundii, as Serratia Enterobacter agglomerans, can determine colonies morphologically similar to those of Y. Enterocolitica. The differentiation is made afterwards by biochemical tests.

From 442 samples of meat examined by Stiles, 86% have a content of enteric bacteria, all the 127 samples of beef milled being positive. The most frequent were involved *Escherichia coli* biotype I (29%), *Serratia liquefaciens* (17%) and *Pantoea agglomerans* (12%). A total of 721 of isolates (32%) were represented by *Citrobacter freundii, Klebsiella pneumoniae, Enterobacter cloacae and E. Hafniae*. On the examination of 702 food for fecal coliforms, the most probable number of germs (MPN), representing 10 categories of food, was found in 119 samples of milled beef, the geometrical average being obtained by the AOAC procedure was of 59 /g.

CONCLUSIONS

- 1. Escherichia coli is a gram-negative bacillus that presents sometimes filamentous forms.
- 2. The majority of the species presents peritrich cillia and are not capsulated.
- 3. There are though strains of immovable Escherichia coli, some presenting capsule.
- 4. It grows on simple media where the glucose is the only organic constituent.
- 5. It is an aerobe germ, facultative anaerobe that can have fermentative or respiratory metabolism.
- 6. On solid media it grows under the form of colonies of "S" type and in the liquid media it determines uniform agitation and adherent ring on the wall of the tube.
- 7. The weakly selective media, MC, EMB, allow the growth of all the equally lactose-positive and negative enterobacteriaceae, even of other groups of gram-negative bacilli as *Vibrio*, inclusively *V.cholerare*, *Pseudomonas*.
- 8. The moderate selective media have a higher selective capacity, considerably inhibiting the lactase-positive enterobacteriaceae.
- 9. The highly selective media are inhibiting for all the groups of enterobactericae, with the exception of salmonella.
- 10. The strains of Escherichia coli that poses the "col' plasmid emit colicines, toxic substances for other bacterial strains.

REFERENCES

1. Buiuc D. 2003 – Medical Microbiology: guide for the study and practice of medicine, Ed. "Gr. T. Popa" Iași.

2. Cepoi V., Azoicăi D. 2012 – Guide of management of nosocomial infections. Ed. Arte, București.

3. Constantiniu S., Ionescu G. 2005 – Acinetobacter type in human pathology. Bacteriology, Virusology, Parasitology, Epidemiology, pp. 50:1-2, 157-173.

4. Crisan A., Nicoara E. 2015 - Course of Infectious diseases, Ed. de Vest, Timișoara.

5. CORNELISSEN C. N. HOBBS M. M. 2020 – Microbiology, fourth edition, Lippincott Illustrated reviews.

6. Campfield T, Braden G, 2010. Urinary Oxalate Excretion by Very Low Birth Weight Infants Receiving Parenteral Nutrition. In Pediatrics, pp. 84(5):860-3.

7. CAROLL K.C., PFALLER M.A., LANDRY M.L., MCADAM A.J., PATEL R. RICHTER S.S., WAENOCK D.W, 2019 - Manual of Clinical Microbiology, 2 volume, (ASM Books), 12th edition.

8. Dumitrașcu V., Laboratory Medicine. Biochemistry of urine, EdituraOrizonturiUniversitare, Timișoara, 2002

9. Earnest DL. Enteric Hyperoxaluria. In Adv Intern Med, 1979.LaboratorSynevo. Specific references to the work technology used in 2015. Ref Type: Catalogue. pp.24:407-27 (review).

10. Dumitrașcu V. and collab. 2007 – Farmacology– antimicrobial medicine, Ed. de Vest, Timișoara,.

11. Engemann JJ, Carmeli Y, Cosgrove SE, et al, 2003. Adverse clinical and economic outcomes attributable to methicillin resistance among patients with Staphylococcus Aureus surgical site infection. Clin Infect Dis, pp. 36(5):592-598.

12. Francis JS, Doherty MC, Lopatin U, et al,2005. Severe community-onset pneumonia in healthy adults caused by methicillin-resistant Staphylococcus Aureus carrying the PantonValentine leukocidin genes. Clin Infect Dis, pp.40(1):100-107.

13. Fridkin SK, Hageman JC, Morrison M, et al, 2005. Methicillin-resistant Staphylococcus Aureus disease in three communities. N Engl J Med, pp. 352(14):14361444.

14. Głuszek, J., 1998. The effect of glucose intake on urine saturation with calcium oxalate, calcium phosphate, uric acid and sodium urate, International Urology and Nephrology, pp. 20 (6), 657-663.

15. GOERING VG, DOCKRELL HM, ZUCKERMAN M, CHIODINI PL 2019 – Mims Medical Microbiology and Immunology, Elsevier, sixth edition.

16. Garrity G.M., Bell J.A., and Timothy G.I. 2004 –Taxonomic outline of the Prokaryotes, Bergey's Manual of Systematic Bacteriology –II-nd edn. Bergey Manual Trust, Springer, New York.

17. Heymann D.L. 2012 - Manual of management of transmissible diseases, Ed. Amaltea, București.

18. Holtmann H., Nitschke J., 2017 – Basics Medizinische Mikrobiologie, Hygiene und Infektiologie, 4 Auflage, Elsevier GmbH Deutschland.