

IDENTIFICATION OF YERSINIA IN FOOD TOXINFECTIONS

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Abstract

The term of "biota" is used in this text, instead of "flora" as general reference to bacteria. The flora is referring to the life of plants. The term of "bacterial flora" goes back to the period when it is considered that the bacteria would have been primitive plants. Because the bacteria are not plants it is preferred instead of "flora" the term of "biota" or of "bacterial microbiota". In general, biota reflects the mediums of cutting and of processing mentioned above, the gram-negative bacteria being predominant. Among the gram positive bacteria the most frequent are the enterococci and lactobacilli.

Due to their presence in all the mediums of processing of meat it is expected that the number of yeasts to be very large, including, Penicillium, Mucor and Cladosporium. The yeasts that are found most frequently in the red meat and the poultry are members of the genders Candida and Rhodotorula.

Key words: biota, bacterial flora, enterococci.

INTRODUCTION

Yersinias are gram negative Enterobacteriaceae with coccobacillary morphology. In the Yersinia genre are included 12 species of which only three, *Y. pestis*, *Y. pseudotuberculosis* and *Y. enterocolitica* were isolated from the human being, the others being present only in the soil and waters, or pathogen for the wild mammals, birds and fish.

Yersinia pestis has as main fountains the rodents. From the *Y. pestis* animals is transmitted to the human being by the fleas of the rats, in the proventricol and stomach of which it lives for a long time, or by dust of excretions or animal products. Between the human beings the pest is transmitted by the *Pulex irritans* flea and, most frequently, by inhaling of Flügge drops from the patient in the prodromal stage or the acute stage of pulmonary pest.

Yersinia pseudotuberculosis has as natural fountain rodents and wild birds from which it gets on the soil and in the waters where it survives actively, even with low temperatures. In human being it is transmitted on digestive way and is the cause of some sub-acute enteritis accompanied by marked mesenteric adenopathy that can mime appendicitis. The clinical

picture includes fever, diarrhea and abdominal pain, with the duration of 1-3 weeks; the nausea and vomit are added in 15-40% of the cases. In the fecal matters can be detected leucocytes, blood or mucus. The severe cases can be complicated with ileac perforations or rectoragias. The patients with mesenteric adenitis or terminal ileitis present fever pain and muscular defense in the right iliac fossa accompanied by leukocytosis; the older children and adolescents are affected predominantly. In these cases the picture can't be differentiated from the acute appendicitis and often is intervened surgically.

The sepsis induced by *Yersinia pseudotuberculosis* is rarely reported; approximately 50% of the patients with sepsis present a basic chronic affection. A syndrome similar to the scarlatina was described in association to some strains of *Y. pseudotuberculosis* in the Eastern Russia and Japan. This condition is explained by the production of a mitogen, a unique super antigen similar to the one involved in the syndrome of toxic shock induced by staphylococci and streptococci. Moreover the recent data indicate the involving of the bacteria in the Kawasaki disease, the infection being associated with an increased frequency of the arterial lesions.

MATERIAL AND METHODS

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

The duration for which was extended the study is of 5 years, included in the period 01.01.2014-31.12.2019.

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

Necessary materials for the performing of the examination:

- A recipient of collection (collection recipient of fecal matter with collecting spoon) with transport medium
- Wood 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 larger quantities than 10 g because will reduce the chances to isolate the pathogen bacteria.

RESULTS AND DISCUSSIONS

The antibodies detected by the method Western blot are guided against the 3 species of *Yersinia*: *enterocolitica*, *pseudotuberculosis* and *pestis*. The test uses the secretor antigens coming from *Yersinia* (Yop: *Yersinia* outer proteins) relevant serologically, that are separated based on the molecular weight by the electrophoresis in the gel of polyacrylamide in the presence of sodium dodecyl sulfate (SDS-PAGE) and transferred afterwards electrophoretically on a membrane of nitrocellulose (Western blotting). The free connecting sites from the membrane are saturated with a solution of proteins, and afterwards the matrix is washed and cut in strips. For the detection of specific antibodies anti-*Yersinia* the strip loaded with antigens is incubated together with the diluted serum of the patient. If in the serum are present specific antibodies they are connected to the corresponding antigens from the strip. After a phase of washing, the strip is incubated, depending on the tested class of antibodies, with a human anti-IgG or anti-IgA conjugated marked with the alkaline phosphatase. The conjugate will be attached to the antigen-antibody complex formed. After the elimination by washing of the conjugate not connected it is added the chromogenic substrate. If the connected conjugate is present the enzymatic reaction will generate a product of violet color on the level of the bands occupied by the specific antibodies. Thus the bands visualized on the level of the strip are the result of the connection of the specific antibodies to the individual antigens. Each strip includes in the upper part a band of control that represents the control of the reaction necessary to confirm the correct performing of the test. The test is validated if the band YopD (35 kDa) of the control IgA and IgG is present. The evaluation of the intensity of the bands that appeared in the serum of the patients is made using as reference the band of control. The bands whose intensity is larger or equal to the one of the control are marked with X in the protocol of work. The very intense bands will be marked with XX; the bands with intensity smaller than the one of the control won't be considered. **The antibodies IgG** are produced in case of chronic yersiniosis, the reactive arthritis associated with *Yersinia* and of acute yersiniosis. In the beginning stage of a yersiniosis the antibodies are rarely detectable. The IgG antibodies persist minimum 5 months from the beginning of the disease, but most often for a longer period (over 5 years). The antibodies IgG are guided against all the proteins secreted by *Yersinia*, but most often against YopE (23 kD), YopD (35 kD), YopB (41kD) and YopH (51 kD). **The antibodies IgA** appear pregnantly in the beginning phase of the acute yersiniosis. In case of the reactive arthritis associated with *Yersinia*, the response of IgA is guided in 90% of the cases against the antigen YopD (35kD). In case of chronic yersiniosis the

antibodies IgA are guided in a visible way against YopE (23 kD), YopD (35 kD) and YopB (41kD). In case of the complicated yersiniosis the antibodies IgA persist in the majority of the case for more years and in case of the simple yersiniosis, usually, only a few months.



Fig.1. colonies of Yersinia. The medium of culture endo – agar.<http://www.bacteriainphotos.com>

In the study “Enterocolitis Yersinia: the charisma continues”, affirms the fact that the majority of the human pathogen strains are found in distinct blood groups (for example, O: 3, O: 5,27, O: 8, O: 9) and include factors of virulence mediated by chromosomes, and by plasmids (60 up to 75 kb), absent in the “non virulent” strains. While Enterocolitis Yersinia is first of all a pathogen agent of the gastric-intestinal tube, it can produce extra-intestinal infections in hosts with basic predisposing factors. The post infection sequelae include arthritis and nodular erythema, that are observed mainly in Europe in the patients with blood groups: O: 3 and O: 9 infection and antigen HLA-B27. Enterocolitis Yersinia is achieved on an oral way and is connected epidemiologically to the pig sources.

CONCLUSIONS

For *Yersinia* a current procedure of enriching is keeping the tapped swab in tampon solution of phosphate 2-3 weeks at 4-5°C after which is seeded after selective mediums. Because the bacteria from the *Yersiniagenre* are developed preferentially at 22-29°C, the simple incubation at this temperature performs the enriching on the broth for gram-negative bacilli.

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