Pseudomonas aeruginosa/Sera

Agglutinant sera for grouping *Bacillus pyocyaneus*

DEFINITION

Human and animal infections due to pyocyanic bacillus (*Pseudomonas aeruginosa*) are currently very frequent, due to the fact that this bacterium has become resistant to most antibiotics.

Within hospitals, these infections can take on epidemic proportions. They are then redoubtable and can lead to temporary closure of the department involved. It is then imperative to detect the epidemiological network of these hospital infections, so as to find the origin (it is often connected with the so-called "antiseptic" solutions used to disinfect of non-autoclavable medical equipment and probes) and to exonerate healthcare staff, possibly healthy carriers (throat, stools) of pyocyanic bacilli not responsible for the epidemic.

This epidemiological study involves investigating whether all pyocyanic bacilli isolated during the epidemic are identical or not.

Three methods can be used in this investigation:

1. Study of the biochemical characters of identification

This permits diagnosis of the species.

2. Determination of lysotype and pyocyano-type

A very precise method, but one that can only be used in specialized analytical laboratories.

3. Determination of O antigen group using agglutinant sera

This method is straightforward and provides essential and sometimes sufficient information for epidemiological investigation.

a) O antigen groups

Classification of strains of *Bacillus pyocyaneus* into O groups has been the subject of many studies. We have adopted the classification established by HABS and completed by the International Sub-committee on *Pseudomonas*. According to this classification, there are 16 O antigen groups numbered from 1 to 16.

b) Anti-pyocyanic agglutinant sera (ANTI-O)

Anti-O immune sera are obtained from rabbits inoculated with heated bacterial suspensions (the thermolabile H antigen being destroyed). Specificity is then obtained by cross-saturation.

Serum mixes

To facilitate typing, 4 mixes of sera are prepared and labelled PMA, PME, PMC and PMF:

PMA = $P_1 + P_3 + P_4 + P_6$
PME = $P_2 + P_5 + P_{15} + P_{16}$
PMC = $P_9 + P_{10} + P_{13} + P_{14}$
PMF = $P_7 + P_8 + P_{11} + P_{12}$

Monovalent sera

16 monovalent sera numbered from 1 to 16 are available. There are strong cross-reaction between groups O: 7 and O: 8 and also between groups O: 13 and O: 14.
The monovalent sera corresponding to these groups are absorbed in consequence.

PRESENTATION

Serum mixes and monovalent sera are presented in dropper bottles of 3 ml.

- **Serum mixes**

PMA	**code 355-8922**
PME	**code 355-8932**
PMC	**code 355-8942**
PMF	**code 355-8952**

- **Monovalent sera**

P_1	**code 355-8901**
P_2	**code 355-8902**
P_3	**code 355-8903**
P_4	**code 355-8904**
P_5	**code 355-8905**
P_6	**code 355-8906**
P_7	**code 355-8907**
P_8	**code 355-8908**
P_9	**code 355-8909**
P_{10}	**code 355-8910**
P_{11}	**code 355-8911**
P_{12}	**code 355-8912**
P_{13}	**code 355-8913**
P_{14}	**code 355-8914**
P_{15}	**code 355-8915**
P_{16}	**code 355-8916**

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Pseudomonas aeruginosa
Agglutinant sera for grouping *Bacillus pyocyaneus*

STORAGE
- Ready to use: + 2°C to 8°C.
- Expiration date and batch number are shown on the package.

METHODOLOGY
This consists of agglutination of a living bacterial suspension with an anti-O serum.

Method:
Start with detection of agglutination in the 4 serum mixes, then in the 4 specific sera corresponding to the mixture giving a distinct agglutination.
From the surface of a fairly dry agar, collect bacteria cultivated at 37°C for 16-24 hours.

Place these bacteria in suspension in a drop of each serum mix deposited on a slide, taking care to make a homogenous suspension by adding serum gradually using a platinum loop. O agglutination appears in a few minutes; it is fine and regular and quite distinctive from any fragments of poorly-emulsified bacterial colonies that may be present.

Proceed to detect agglutination in specific sera which are components of the serum mixes that produce agglutination.
In the event of failure (e.g. with strains producing a quantity of mucous antigens), it is recommended that a very thick suspension of colonies taken from the agar be prepared in distilled water, which is then heated at 120°C for 30 minutes (autoclave). This suspension is subjected to centrifugation, the supernatant is removed, and agglutinations are again looked for using the bacterial pellet.

N.B.:
- Approximately 5% of strains of *Pseudomonas aeruginosa* are unstable, autoagglutinable.

- Approximately 1% of stable strains of *Pseudomonas aeruginosa* belong to groups other than the 16 used here. Their typing would theoretically be possible with other sera, but the practical value of these new O groups seems limited.

QUALITY CONTROL OF MANUFACTURER
Every product manufactured and marketed by Bio-Rad is subject to a quality-assurance procedure at all stages, from the reception of raw materials to the marketing of the end-product. Each batch of finished product undergoes quality control and is marketed only if it satisfies the acceptability criteria.

Documentation relative to the production and control of each batch is kept on file.

BIBLIOGRAPHY

- **VIEU J.F., ALLOS G., HASSAN-MASSOUD B., SANTOS-FERREIRA M.O., TSELENTIS G. (1984):** Existe-t-il une épidémiologie géographique des sérogroupes O de *Pseudomonas aeruginosa* ? Bull Soc. Path. Ex. **77**: 288-294.

- **HABS I.Z. (1957):** Hyg. **144**: 218-228.

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