

The Effect of Environmental Factors on the Growth of Bacteria

Temperature and Medium Composition

(*Arthrobacter atrocyaneus* and *Serratia marcescens*)

Every living organism is best fitted to live under a given set of environmental conditions, but is still able to exist outside of these conditions within a certain tolerance range. The optimum conditions (e.g., temperature) for growth of the organism may not necessarily be the same as those for some of the metabolic or physiological functions of the organism (e.g., motility, pigmentation). Thus it is important to specify the cultural conditions when recording the properties of an organism.

In this exercise you will study the effects of nutrients (especially glucose), pH, temperature, and time (age of cultures) on colony characteristics, pigmentation, cell morphology, Gram reaction, and motility of bacteria.

Materials required (per group)

Arthrobacter atrocyaneus or *Serratia marcescens*
Agar plates: 6 YGC, 3 SDA, 3 NP, 1 NA
Gram stain reagents
Anaerobe jar
Incubators at 22°C, 30°C, 37°C, and 45°C
Refrigerator at 4°C

Procedure

Day 1

Streak a culture of *Arthrobacter atrocyaneus* or *Serratia marcescens* on the following types and numbers of agar plates: 6 YGC, 3 SDA, 3 NP, 1 NA. Incubate the plates under the following conditions:

VARIOUS TEMPERATURES under AEROBIC CONDITIONS

Refrigerator (4°C)	YGC ¹			
Room temperature (22°C)	YGC ²	SDA ¹		
30°C	YGC ³	SDA ²	NP ²	NA ¹
37°C	YGC ⁴	SDA ³	NP ³	
45°C	YGC ⁵			

ANAEROBIC CONDITIONS

30°C	YGC ⁶
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Prepare a wet mount and Gram stain of the inoculum.

Environmental Effects

Days 2-4

Observe the cultures over a period of one week. Note both colony characteristics and cell morphology. Describe the relative amounts of growth on the plates and pigmentation of the colonies. Describe well isolated colonies, and write up your conclusions concerning the effects of environmental conditions on growth and pigmentation. Make the following specific comparisons:

a. Colony Characteristics

1. Effect of O₂: Compare plates YGC³ and YGC⁶.
2. Effect of temperature: Compare plates YGC¹ through YGC⁵. State the tolerance range and the temperature optima for growth and pigmentation.
3. Effect of glucose and pH: Compare plates SDA¹⁻³ and NP¹⁻³.
4. Effect of nitrogenous media components: Compare plates YGC³ and SDA²; compare plates NP² and NA¹.

b. Cell Morphology

Prepare wet mounts and Gram stains (place 2-3 smears on one slide) and note cell morphology, motility, and Gram reaction.

1. Compare YGC³ and NP².
2. Effect of time (age). Observe organisms from plates YGC³ and NA¹ in young and mature cultures.

Incubate the plates on which no colonies developed for an additional period of 48 hours at 30° in the presence of air. Identify the bacteriostatic and bacteriocidal environmental factors.