



## TEMPERATURE CHANGE ON THE GROWTH OF BACILLUS ISOLATED FROM BEACH SEDIMENTS

RAMI HAMMOD<sup>1\*</sup>, AHMAD KARA ALI<sup>1</sup> AND BADR AL ALI<sup>1</sup>

<sup>1</sup>Department of Marine Biology at HIMR, Tishreen University, Lattakia, Syria.

### AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

*Received: 12 July 2021*

*Accepted: 17 September 2021*

*Published: 13 October 2021*

*Original Research Article*

### ABSTRACT

The study was conducted to determine the temperature change on the growth of marine bacterial isolates of the genus *Bacillus* isolated from the coastal sediments of Lattakia city. Marine sediment samples were collected from the Apamea region during the period (2019-2020) the identity of these bacterial isolates was determined through laboratory and microbiological analyses. A number of experiments were conducted to find out the effect of temperature changes on the growth of these bacterial isolates. The results showed that the best growth of bacterial isolates was at an incubation temperature of 30°C.

**Keywords:** Bacillus; marine sediments; biochemical tests.

### 1. INTRODUCTION

The species of the genus *Bacillus* are important microorganisms in the medical, industrial and agricultural fields, as they produce about 167 antibiotics, in addition to their production of many important enzymes in the medical and industrial fields [1]. In the industrial field, its use is no longer limited to the production of some industrial compounds only, but also includes the production of the most complex pharmaceutical chemical compounds, the most important of which are antibiotics, hormones, vitamins, pesticides and biofertilizers [2]. This genus is also complex at the genetic, phenotypic, metabolic, taxonomic and ecology levels. This allows it to be extremely versatile in different environments, especially in soil and water [3]. Temperature plays an important role in the microbial growth of bacteria. The growth of bacteria and their production of enzymes, antibiotics and chemical compounds are affected by physical factors (such as temperature, pH,

humidity and salinity) and chemical (such as the quality of the carbon and nitrogen sources in the culture medium, etc.). The effect of physical and chemical factors varies according to the type and strain of bacteria used, depending on its genetic makeup and its adaptation to the environment in which it lives [4]. Studies have shown that marine microorganisms have characteristics that differ from their counterparts on land due to the special environmental conditions in the marine environment, and this is what makes them the focus of researchers' attention. Therefore, the current study aimed to study the effect of some physical factors (such as temperature and humidity) and chemical (such as pH, salinity, The quality of the carbon and nitrogen sources in the culture media) On the growth of a number of marine bacterial isolates of the genus *Bacillus* isolated from the marine sediments of Lattakia city to determine the optimal conditions for their growth.

\*Corresponding author: Email: ramihammad1133@gmail.com;

## 2. RESEARCH MATERIALS AND METHODS

The study was conducted on the site of Apamea (35°76'19.1"N; 35°54'19.3"E) which is an open area affected by tourism activities and sewage canals. Marine sedimentary samples were collected by sterile 500ml glass bottles during the month of October (2019-2020). It was transferred directly to the laboratory for microbiological laboratory study. After obtaining marine isolates of different species of the genus *Bacillus sp.*, they were modeled as follows: (*B. cereus*, *B. subtilis*, *B. circulans*, *B. polymyxa*). These species were chosen because they are more prevalent in the Syrian coast, and are the most productive types of effective antibiotics. A number of experiments were conducted to find out the effect of temperature change and they were as follows: Liquid cultures of the four bacterial isolates of the marine *Bacillus* genus were prepared in the middle of the nutrient broth and incubated at a temperature of 30°C for 24 hours. Temperature variation was carried out on the growth of bacterial isolates using Sorenson broth medium (Table 1), The bacterial cultures were incubated at 30°C for 24 hours. The bacterial growth of cultures of *Bacillus* species was measured using a spectrophotometer to measure the optical density of bacterial growth at a wavelength of 600 nm during the incubation period [5].

**Table 1. Chemicals used to prepare Sorenson broth medium [6]**

Quantity (g/L)	Subject
2	Xylan
1	NH <sub>4</sub> NO <sub>3</sub>
0.5	K <sub>2</sub> HPO <sub>4</sub>
0.2	NaCl
0.5	MgSO <sub>4</sub> .7H <sub>2</sub> O
0.02	FeSO <sub>4</sub> .6H <sub>2</sub> O

### 2.1 The Effect of Temperature Changes on the Growth of *Bacillus* Isolates

Test the effect of incubation at different temperatures (10, 20, 30, 40, 50) C on the growth of cultures of *Bacillus* isolates prepared as in the previous step with a light density of 0.05 at the beginning of the

experiment. The optical density was measured after a 24-hour incubation period for each bacterial isolate using a spectrophotometer at a wavelength of 600 nm to find the best growth temperature [5].

## 3. RESULTS AND DISCUSSION

The effect of temperature changes on the growth of *Bacillus* isolates after 24 hours of incubation:

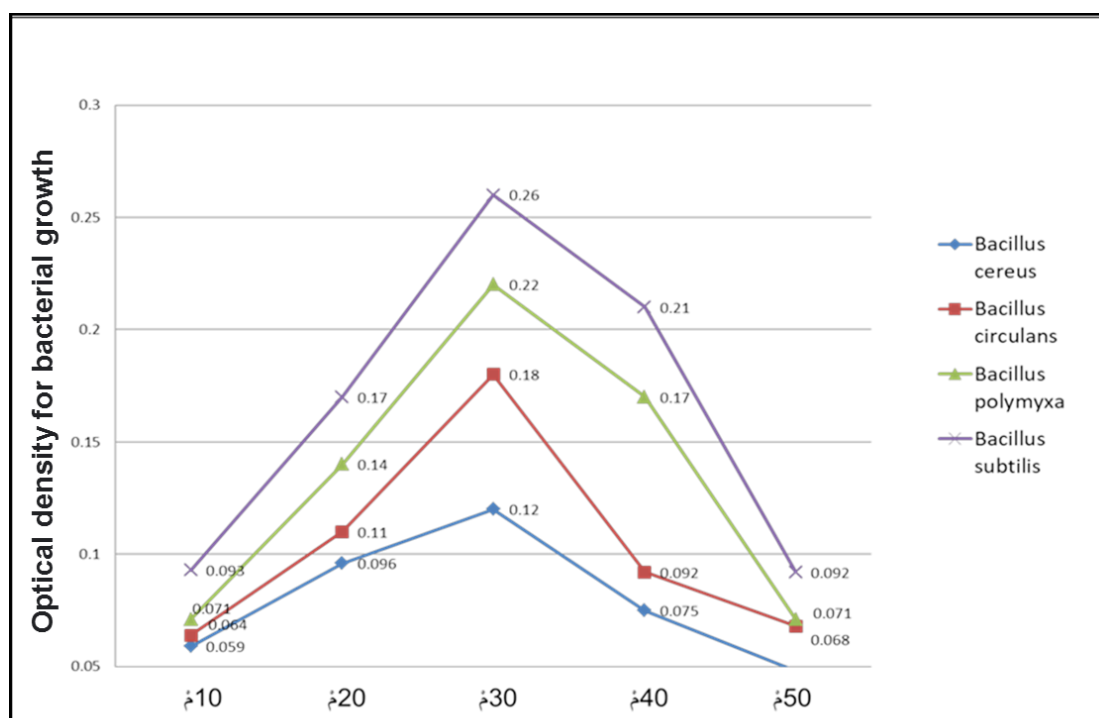
As for the isolate of *Bacillus subtilis*, the results showed that the highest optical density was recorded (0.26) at a temperature of 30° C, and the lowest optical density (0.092) was recorded at a temperature of 50° C (Fig. 1). The effect of temperature changes on the growth of *Bacillus* isolates after 24 hours of incubation:

For *Bacillus polymyxa*, the results recorded that the highest optical density was (0.22) at a temperature of 30° C, and the lowest optical density was (0.071) at a temperature of 50° C (Fig. 1).

For *Bacillus circulans*, the results showed that the highest optical density was recorded (0.18) at a temperature of 30° C, and the lowest optical density was recorded (0.064) at a temperature of 50° C (Fig. 1).

For *Bacillus cereus*, the results showed that the highest light density was (0.12) at a temperature of 30°C, and the lowest light intensity was recorded at a temperature of 50°C, which was 0.048 (Fig. 1).

Temperature plays an important role in the chemical and physiological behavior of vital components of an aquatic ecosystem [3]. The effect of water temperature usually varies depending on the seasons, geographic location, sampling time and the temperature of the water source entering the aquarium [7]. It is shown (Table 2 and Fig. 1) that the best growth of bacterial isolates was at an incubation temperature of 30°C, followed by their growth at a temperature of 40°C, and the lowest growth appeared at a temperature of 50°C, followed by a temperature of 10°C. This discrepancy in growth at different temperatures can be attributed to the effect of temperature change on the morphological and physiological characteristics and enzyme activity of marine bacteria [4].



**Fig. 1. Effect of different incubation temperature (10, 20, 30, 40, 50) C on the bacterial growth of Bacillus after incubation (24) hours**

Bacteria grow well only when optimum temperatures are secured, as most bacteria grow at a temperature of 30-37° C, which is specified for bacterial growth. The optimum temperature for growth varies according to the type of bacteria and the study area [2]. This can be attributed to the effect of temperature on enzymatic activity (particularly transport enzymes) and other important physiological functions in the microbial cell [8]. Temperature plays an important role in the growth of bacteria, which depends on the adaptation of bacteria and the nature of its cytoplasmic membrane and cell wall, and thus affects the physiology, production of enzymes and the production of chemical compounds [9]. The rate of bacterial growth increases with the increase in the temperature of the medium, and this causes the consumption of organic materials and an increase in the production of antibiotics, which leads to a decrease in the production of enzymes [10].

#### 4. CONCLUSIONS AND RECOMMENDATIONS

The results showed that studying the effect of temperature change on the growth of Bacillus isolates is of importance in the medical, industrial and agricultural fields. Where the growth of bacteria and their production of enzymes, antibiotics and chemical compounds are affected by physical and chemical factors (such as temperature, pH, humidity, salinity).

The effect of physical and chemical factors varies according to the type and strain of bacteria used This is in line with Adam et al.'s 2014 study.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

#### REFERENCES

- Schmidt A, Kochanowski K, Vedelaar S, et al. The quantitative and condition-dependent Escherichia coli proteome. *Nature Biotechnology*. 2016;34(4):104-110.
- Maddamsetti R, Lenski RE, Barrick JE. Adaptation, clonal interference, and frequency-dependent interactions in a long-term evolution experiment with Escherichia coli. *Genetics*. 2015;200(2):619-631.
- Prakash P, Jayalakshmi S, Prakash B, et al. Production of alkaliphilic, halotolerant, thermostable cellulase free xylanase by Bacillus halodurans PPKS-2 using agro waste: single step purification and characterization. *World Journal of Microbiology*. 2012; 28(1):183-192.
- Bear SE, Nguyen MT, Jasper JT, et al. Removal of nutrients, trace organic contaminants, and bacterial indicator organisms

- in a demonstration-scale unit process open-water treatment wetland. *Ecological Engineering*. 2017;109(3):76-83.
5. Gupta U, Kar R. Technology. Xylanase production by a thermo-tolerant *Bacillus* species under solid-state and submerged fermentation. *Brazilian Archives of Biology*. 2009;52(6):1363-1371.
  6. Yilmaz M, Soran H, Beyatli Y. Antimicrobial activities of some *Bacillus* spp. strains isolated from the soil. *Microbiological Research*. 2006;161( 2):127-131.
  7. Singh D, Singh B. Utility of acidic xylanase of *Bacillus subtilis* subsp. *subtilis* JJBS250 in improving the nutritional value of poultry feed. *Biotech*. 2018;8(12):1-7.
  8. Chasanah U, Nuraini Y, Handayanto E. The potential of mercury-resistant bacteria isolated from small-scale gold mine tailings for accumulation of mercury. *Journal of Ecological Engineering*. 2018;19(2).
  9. Abad M, Bedoya L, Bermejo P. Marine compounds and their antimicrobial activities. *Science against Microbial Pathogens: Communicating Current Research*. 2011; 51(3):1293-1306.
  10. Adam SI, Ahmed AA, Omer AK, et al. In vitro Antimicrobial activity of *Rosmarinus officinalis* leave extracts. *Agri-Food Appl Sci*. 2014;1(2):15-21.