

TESTS WITH A PIGMENT (VIOLACEIN) PRODUCED BY A
PROBABLE NEW SPECIES OF CHROMOBACTERIUM *CLEOMENES REIS ** EDITH BLAU *** JOSÉ JOAQUIM
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SUMMARY

The results of spectrophotometric and chromatographic tests with a pigment produced by a probable new species of chromobacterium are presented. This pigment shows the characteristics described for violacein.

INTRODUCTION

In a previous paper, the isolation of a probable new species of chromobacterium from polluted water in the municipality of Goiânia (Goiás) was reported ⁴.

Ballantine et al. ¹ elucidated the structure of violacein, the pigment produced by known species of chromobacterium. They concluded that the violacein molecule consists of a 5-hydroxyindole, an alpha pyrrolidone and an oxindole unit.

In recent years, DeMoss and Evans ³ demonstrated that the

only substance required for the biosynthesis of violacein was tryptophan.

Sneath ⁵ studied the absorption spectra of violacein and found that the ethanolic solution shows an absorption maximum at 580 mu. and a minimum at 430 mu; he also observed an ultraviolet absorption peak in the region of 200 mu. When 10% H₂SO₄ was added to this solution, it showed an absorption maximum at 700 mu.

The genus *Chromobacterium* contains only two well-defined species: *C. violaceum* and *C. lividum*.

The aim of the present study was to investigate if the pigment isolated from a probably new species of chromobacterium presents the features described for violacein.

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MATERIALS AND METHODS

The pigmented material which had been obtained from cultures was submitted to the following methods of study:

- a) Spectrophotometry
- b) Thin-layer chromatography

Two solutions of the bacterial pigment were used for spectrophotometric analysis: one in 96% ethanol and the other in 96% ethanol to which 10% (v/v) sulphuric acid had been added. The former solution gave a violet colour, whereas the latter turned green. (This finding is in accordance with the characteristics described by Sneath)⁵. Absorption spectra were obtained with a Zeiss spectrophotometer; 2,0 ml silica cells were used.

Thin-layer chromatography was carried out on silica gel G. The

following solvent systems were used: benzene/ethanol (8:2); chloroform/acetone (6:4) and ethyl acetate.

RESULTS

Fig. 1 presents the absorption spectra of the pigmented material. The ethanolic solution shows an absorption maximum at 580 m μ , an ultraviolet absorption peak in the region of 200 m μ and an absorption minimum at 430 m μ . The pigment in 10% (v/v) sulphuric acid in 96% ethanol showed an absorption maximum at 700 m μ and a strong ultraviolet absorption peak at 220 m μ .

A single spot was detected by thin-layer chromatography. Rf values are presented in Table I.

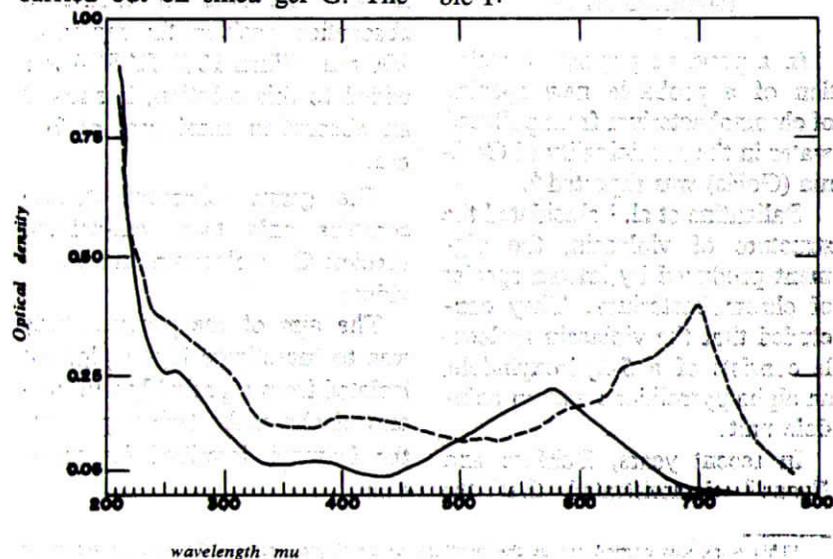


FIGURE 1
— violacein, ethanolic solution
... violacein, 10% (v/v) sulphuric acid in 96% ethanol

TABLE I

Solvent systems	Rf values (at room temperature)
benzene/ethanol	0,60
chloroform/acetone	0,50
ethyl acetate	0,86

DISCUSSION

On the basis of the findings obtained by spectrophotometry and thin-layer chromatography, it was concluded that the pigmented material produced by a probable new species of chromobacterium is violacein.

The results of chromatographic analysis are in agreement with the findings reported by Corpe²

RESUMO

TESTES COM UM PIGMENTO (VIO-LACEINA) PRODUZIDO POR POSSÍVEL NOVA ESPÉCIE DE CHROMO-BACTERIUM

Os Autores fizeram um estudo espectrofotométrico e cromatográfico de um pigmento extraído de uma possível espécie nova de chromobacterium. O pigmento apresentou as características descritas para a violaceina.

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