

# High-Dose $\gamma$ -Irradiation Tolerance of *Spirulina Platensis* in the Presence of Cesium Ions: Insights from DSC and UV-Vis Spectroscopy

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## Abstract

The study of optical absorbance and thermal denaturation spectra of *Spirulina platensis*, grown in a cesium-containing medium mixture and subjected to continuous  $\gamma$ -irradiation at a rate of 1.1 Gy/min for over one year (resulting in a total dose of 543 kGy), demonstrated the complete disappearance of both optical and thermal spectra. After 48 hours of recultivation in standard Zarrouk's medium, the previously  $\gamma$ -irradiated, colorless suspension showed significant recovery of the phycobilisome complex (PBCc) structure, accompanied by minor changes in the denaturation profile and a decrease in denaturation enthalpy of about 15–20%. After 7 days of recultivation, both optical and differential calorimetry (DSC) studies demonstrated complete restoration of structure and functions of *Spirulina platensis*. Based on our previous data demonstrating full restoration of the structural organization and biological functions of microalgae previously irradiated with 400 kGy  $^{137}\text{Cs}$ , we suggest that the changes observed after 48 hours of recultivation are caused by the influence of cesium ions on DNA organization within the nucleoprotein complex, leading to alterations in protein synthesis in *Spirulina platensis*. The complete restoration of the PBCc structure by day 7 of recultivation likely results from the release of cesium ions into the solution due to the rapid growth of the microalgae in the recultivation process. The restoration of the PBCc complex after recultivation of previously treated and irradiated samples, as well as the nature of the highly cooperative transition of the biopolymer that melts at 77.5 °C and fully recovers after heating to 110 °C, are discussed.

**Keywords:** Cesium Ions, *Spirulina Platensis*, UV-Vis Spectroscopy,  $\gamma$ -Irradiation

## 1. Introduction

We conducted *in vivo* experiments to study the influence of extreme conditions on the structure and functions of *Spirulina platensis*, a microalga that contains a phycobilisome complex (PBCc) comprising C-phycocyanin and allophycocyanin — which has been widely used as an anticancer and anti-inflammatory agent in pharmaceuticals, as well as a nutritional supplement. Multiple scientific studies have demonstrated that *Spirulina platensis* suspension is resistant to various extreme conditions, such as exposure to extremely high radiation doses of 400 kGy under anaerobic conditions at room temperature, freezing to -196 °C, and incubation at extreme high temperatures [1-3]. The removal of free water from the *Spirulina platensis* suspension enhances the resilience of the microalgae's bioactive components to high temperatures, with stability observed up to 115 °C. Vacuum-dried powder of the suspension exhibited resistance at 100 °C, although increasing the temperature to 200 °C led to the collapse of the biomass [3]. It should be noted that a significant number of studies have been devoted to the physical, chemical, and biological characteristics of

*Spirulina platensis* cultivated in Zarrouk's medium. However, there are also interesting findings regarding the improved extraction of biological components from the microalga through modifications of the Zarrouk's medium. Specifically, replacing  $\text{Na}^+$  ions with  $\text{K}^+$  has been shown to enhance algal growth and increase the yield of its bioactive compounds [4, 5]. These characteristics of *Spirulina platensis* – a unique living organism with a high capacity to synthesize distinct active biocomponents used both as food and as effective therapeutic agents for cancer and non-cancer diseases – highlight its potential for application not only under standard conditions but also in extreme environments on Earth and potentially in future space missions, including the gradually advancing Mars program [6-9].

The aim of our study was to investigate the ability of *Spirulina platensis* to survive under conditions in which the cultivation medium contains substances capable of affecting its DNA structure. For this purpose, we employed optical and differential scanning calorimetry (DSC) methods to assess the stability of the bioactive components of *Spirulina platensis* in

a modified medium, where the standard Zarrouk's medium was mixed with a modified Zarrouk's medium containing  $\text{Cs}^+$ . The samples were then exposed to a constant dose rate of 1.1 Gy/min for about one year, resulting in a total  $\gamma$ -radiation dose of 543 kGy under anaerobic conditions at a temperature of 22–26 °C. It should be noted that replacing  $\text{K}^+$  with  $\text{Cs}^+$  initiates immediate deep penetration of  $\text{Cs}^+$  into minor groove of DNA and facilitates interactions with the DNA bases. This behavior is attributed to the ability of  $\text{Cs}^+$  to more readily lose hydration water compared to  $\text{K}^+$  when interacting with phosphate groups in the DNA double helix. Depending on concentration,  $\text{Cs}^+$  can induce structural changes in DNA [10-11].

## 2. Materials and Methods

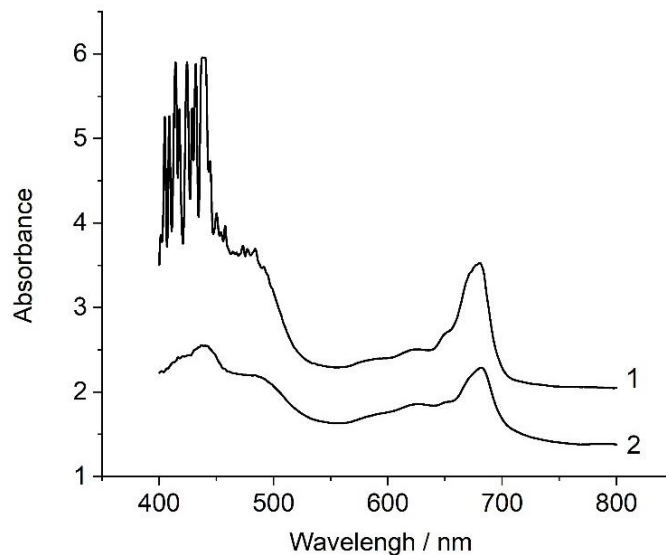
The *Spirulina platensis* strain IPPAS B-256 was cultivated in Zarrouk's alkaline water-salt medium under the following conditions: room temperature, illumination of approximately 5000 Lux, constant stirring, and periodic subculturing [12]. Prior to the experiments, the cells were grown for 7 days in standard Zarrouk's medium. In the standard Zarrouk's medium used for growing *Spirulina*, 2 g/L of ( $\text{K}_2\text{SO}_4 + \text{K}_2\text{HPO}_4$ ) was replaced with an equivalent concentration (2 g/L) of  $\text{CsCl}$ . The composition of Zarrouk's medium and its modifications are described in detail in [13]. A mixture of standard and modified Zarrouk's media (mixed medium) was used simultaneously for *Spirulina* cultivation. In the modified medium, the  $\text{CsCl}$  concentration was  $4.5 \times 10^{-3}$  M, while the potassium salt concentration was  $11.25 \times 10^{-3}$  M. This combined approach was implemented because *Spirulina* did not grow in the modified Zarrouk's medium lacking potassium salts when cesium was present. After 2 weeks of cultivation in this medium, *Spirulina platensis* samples were exposed to prolonged  $\gamma$ -irradiation up to a dose of 543 kGy. Irradiation was performed using a gamma source equipped with a  $^{137}\text{Cs}$  radioisotope at a dose rate of 1.1 Gy/min at the Applied Research Center, E. Andronikashvili Institute of Physics, TSU. Gamma radiation dosimetry was conducted using the certified "GUPOS" gamma facility. The primary radiation source,  $^{137}\text{Cs}$ , has a half-life of over 30 years. Therefore, the decrease in isotope activity during long-term irradiation is negligible, and the facility's certified dosimetry can be considered reliable. Following irradiation, the samples were recultivated in standard Zarrouk's medium. The potassium-to-cesium ratio was 2.5 before irradiation, and after irradiation and subsequent growth in standard Zarrouk's medium, it increased to 20. Growth was assessed by monitoring absorbance at 560 nm using a Cintra 10e UV-Vis spectrometer. Measurements were carried out in a 1 cm path-length quartz cuvette. Biological replicates were performed in triplicate (mean  $\pm$  0.06 SD). *Spirulina platensis* grows optimally in a pH range of 9–11. The suspension, adjusted to pH 10.9, was used to record absorption spectra in the 400–800 nm range. The concentration of *Spirulina platensis* was determined based on spectrophotometric data. Metal ion solutions were prepared in deionized water using analytical-grade inorganic salts. The  $\text{CsCl}$  reagent used was of analytical grade.

It is widely accepted that absorption at 681 nm corresponds to chlorophyll a (Chl a), absorption at 621 nm corresponds to phycocyanin (PC), and the peak at 440 nm corresponds to the Soret band of Chl a [14-17]. Heat absorption curves were recorded using a high-sensitivity differential scanning calorimeter (DSC) [18] capable of detecting thermal transitions in both dilute and concentrated solutions, suspensions, whole tissues, and other complex biological systems. Measurements were carried out using a DSC developed at the E. Andronikashvili Institute of Physics, I. Javakhishvili Tbilisi State University. The DSC has conical measuring cells with volumes of 20, 40, 60, 100, and 150  $\mu\text{L}$ . A volume of 100  $\mu\text{L}$  was used for the experiments in this study. These conical measuring cells were externally inserted into the differential measuring block [18]. It should be noted that the conical and cylindrical shapes of the measuring cells, unlike that of capillary calorimeters, makes it possible to measure not only dilute solutions but also complex biological systems such as cell suspensions, whole tissues, blood, and blood serum/plasma. The sample scanning rate can be selected in the range of 0.0025 to 1.6 °C/min [19]. This allows for the acquisition of valuable information about the behavior of macromolecules and complexes in vivo. Additionally, this type of DSC has been successfully used in the diagnostics and monitoring of both malignant and non-malignant diseases [20-23]. In our experiments, the heating rate was set at 1 °C/min over a temperature range of 20–110 °C, with a sensitivity of 0.5  $\mu\text{W}$ . Protein concentrations were determined directly in the calorimeter measuring cell at 110 °C by subtracting the dry mass of the Zarrouk's medium.

The error of temperature measurements did not exceed 0.1 °C. The error in determining the denaturation enthalpy ( $\Delta H_d$ ) and exit heat capacity ( $dQ/dT$ ,  $\Delta C_{\text{max}}$ ) did not exceed 7%. All DSC data were normalized per gram of dry biomass. The microcalorimeter processor was equipped with all necessary software for determining the thermodynamic parameters of melting/denaturation of the *Spirulina platensis* suspension. Calorimetric curves were plotted and deconvoluted using Origin graphing and analysis software [18]. DSC measurements were performed on *Spirulina platensis* suspensions containing 0.4–2.5 mg of biomass. In all DSC experiments, the pH of the suspension was maintained at 9.5, and the scanning rate was 1 °C/min. Control of the DSC heating program and data acquisition were carried out using the LabVIEW graphical programming environment. In total, approximately 32 DSC measurements and 21 optical measurements were performed.

## 3. Results and Discussion

The aim of this study was to investigate the optical properties of *Spirulina platensis* following alterations in the potassium ( $\text{K}^+$ ) to cesium ( $\text{Cs}^+$ ) ion ratio in the nutrient medium, combined with subsequent  $\gamma$ -irradiation.

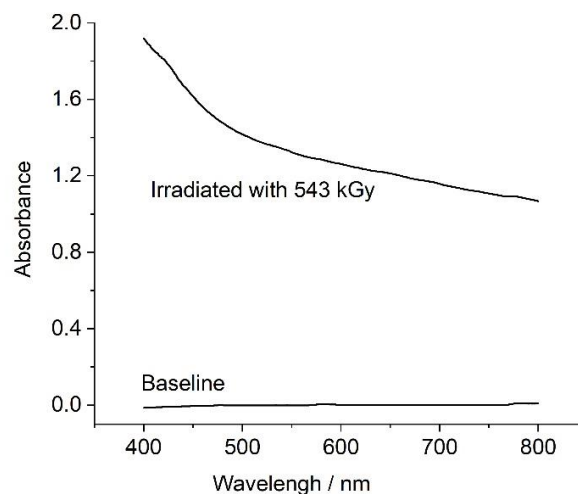


**Figure 1: Optical Absorption Spectra of *Spirulina Platensis***

1. *Spirulina platensis* was cultivated in the mixed system of standard and modified Zarrouk's media containing both  $\text{Cs}^+$  and  $\text{K}^+$  ( $\text{K}^+/\text{Cs}^+ = 2.5$  before irradiation).
2. The suspension, simultaneously cultivated in the mixed system of standard and modified Zarrouk's media, was irradiated with a dose of 543 kGy and subsequently recultivated in standard Zarrouk's medium only. After irradiation and recultivation, the  $\text{K}^+/\text{Cs}^+$  ratio increased to 20.

Fig. 1 presents the absorption spectra of *Spirulina platensis*: (1) cultivated in a mixed system of standard Zarrouk's and modified Zarrouk's media (mixed medium) containing  $\text{Cs}^+$  and  $\text{K}^+$ , and (2) cultivated under the same conditions, then suspension exposed to  $\gamma$ -irradiation at a dose of 543 kGy, followed by recultivation in standard Zarrouk's medium. The absorption spectrum of *Spirulina platensis* grown in the mixed medium is nearly identical to that of the irradiated

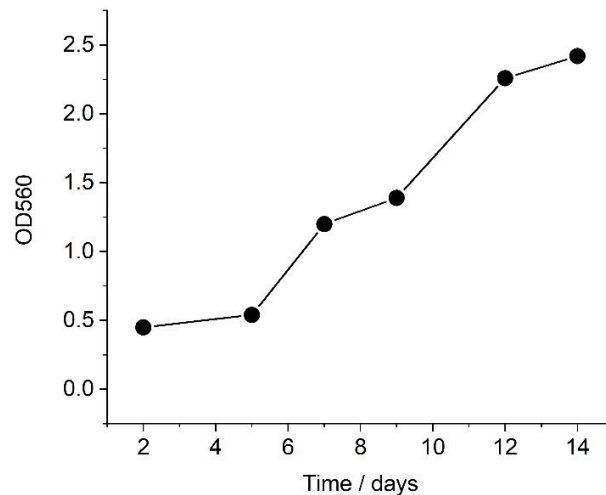
samples after recultivation in standard medium (curve 2). The spectral profiles remained unchanged across the samples, with differences observed only in the overall absorption intensity, indicating variations in biomass concentration. It should also be noted that no growth was observed when *Spirulina* was cultivated in modified nutrient medium containing cesium ions but lacking potassium ions [14].



**Figure 2: Effect of 543 kGy  $\gamma$ -Irradiation on the Absorption Spectra of *Spirulina Platensis* Previously Cultivated In a Mixed System of Standard and Modified Zarrouk's Media**

Fig. 2 illustrates the effect of  $\gamma$ -irradiation at a dose of 543 kGy on the absorption spectra of *Spirulina platensis* cultivated in the mixed medium in the presence of  $\text{K}^+$  and  $\text{Cs}^+$ . As shown in Fig. 2, no distinct changes in the spectral profile were observed following irradiation; however, the optical density relative to the baseline remained consistent in the 600–800

nm wavelength range. A comparison of Figures 1 and 2 suggests that cultivation in the modified medium followed by  $\gamma$ -irradiation induces structural alterations in *Spirulina platensis*. However, subsequent recultivation in standard Zarrouk's medium leads to the restoration of its structural characteristics.

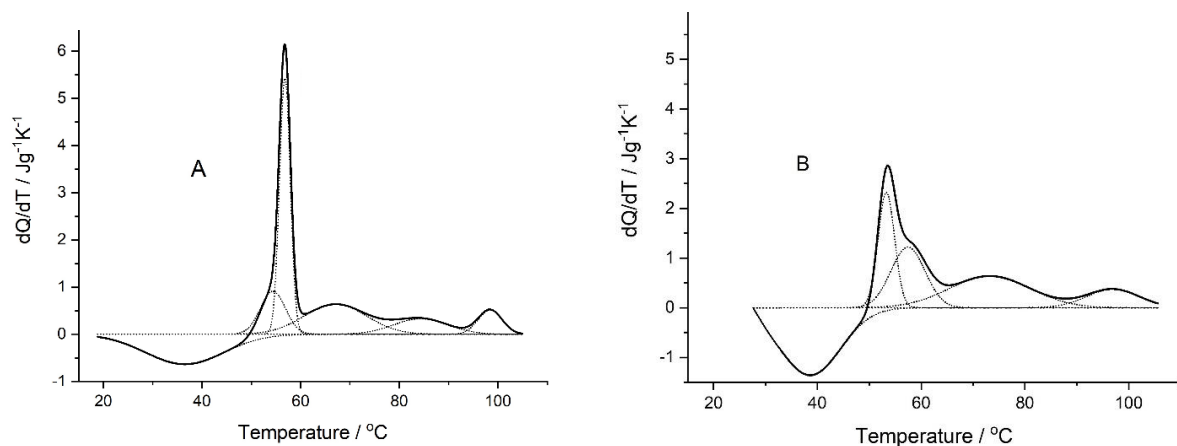


**Figure 3: Kinetic Growth Curve of *Spirulina Platensis* Following Cultivation in the Mixed System of Standard and Modified Zarrouk's Media, Subsequent Irradiation at a Dose of 543 kGy, and Recultivation in Zarrouk's Medium**

Fig. 3 illustrates the kinetic growth curves of *Spirulina platensis* during a 2-week recultivation period in standard Zarrouk's medium, following initial cultivation in the mixed medium and  $\gamma$ -irradiation. The vertical axis represents optical density (OD) at 560 nm, while the horizontal axis indicates cultivation time. As shown in Fig. 3, *Spirulina platensis* demonstrates the ability to recover its physiological characteristics during recultivation. Biological replicates were performed in triplicate, with data reported as mean  $\pm$  0.06 SD. Analysis of the results shows that no growth of *Spirulina platensis* occurred when it was cultivated in modified Zarrouk's medium containing only cesium ions and lacking potassium salts. In contrast, cultivation in the mixed medium containing both  $\text{Cs}^+$  and  $\text{K}^+$  ( $\text{K}^+/\text{Cs}^+ = 2.5$ )

supported *Spirulina* growth. Subsequent exposure to high-dose  $\gamma$ -irradiation for about one year led to structural disruption of *Spirulina* (Fig. 2). However, recultivation in standard Zarrouk's medium after irradiation, where the  $\text{K}^+/\text{Cs}^+$  ratio had increased to 20, resulted in the complete restoration of its structural and physiological components.

In Fig. 4A, B we have presented Gaussian deconvolution of the native *Spirulina Platensis* suspension DSC curves. The deconvolution is done based on the data, according to which the observed peaks, shoulders, and diffusional transitions in the DSC curve correspond to the independent melting of proteins and protein complexes in the *Spirulina platensis* suspension [24].



**Figure 4: The Heat Absorption Curve as a Function of Temperature for Fresh Native *Spirulina Platensis* and the Treated Suspension, Recalculated Per Dry Biomass:**

A. DSC curve recorded after cultivation in non-modified standard Zarrouk's medium

B. DSC curve recorded after cultivation in the mixed system of standard and modified Zarrouk's media containing  $\text{Cs}^+$  ( $\text{K}^+/\text{Cs}^+ = 2.5$ ), subsequent irradiation at a dose of 543 kGy, and recultivation in non-modified Zarrouk's medium for 48 hours ( $\text{K}^+/\text{Cs}^+ = 20$ )

Fig. 4A, B demonstrates DSC curves of native *Spirulina platensis* and recultivated microalgae previously cultivated in a mixed medium and  $\gamma$ -irradiated to a final dose of 543 kGy using  $^{137}\text{Cs}$ . Even a visual comparison of DSC curves A and B in Fig. 4 reveals significant differences in the denaturation profiles, directly indicating partial denaturation of the structural organization in sample B. To quantitatively

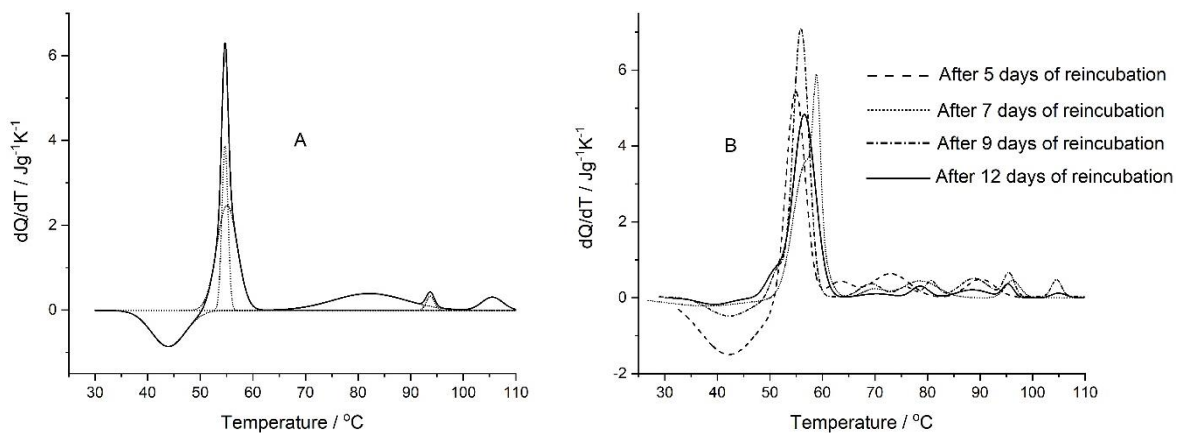
compare the parameters of these curves (Fig. 4A and Fig. 4B), we performed deconvolution, which showed that PBCc – the main component of *Spirulina platensis* denature at 50–60 °C in the native algae (Fig. 4A) (also see [1, 2]) and at 48–65 °C in the processed sample (Fig. 4B). In both cases, the main peak exhibits two distinct transition stages with significantly different parameters, which are presented in Table 1 below.

Sample	Denaturation temperature		Denaturation interval at half height		Denaturation enthalpy	
	$T_{d1}$	$T_{d2}$	$\Delta T_{d1}$	$\Delta T_{d2}$	$\Delta H_{d1}$	$\Delta H_{d2}$
Native <i>Spirulina platensis</i> cultivation in non-modified Zarrouk's medium (Fig. 4A)	54.4 °C	58.1 °C	5.9°	2.5°	5.8 Jg <sup>-1</sup>	15.8 Jg <sup>-1</sup>
<i>Spirulina platensis</i> after cultivation in the mixed medium containing Cs <sup>+</sup> ( $K^+/Cs^+ = 2.5$ ) with subsequent irradiation at dose 543 kGy, and recultivation in non-modified standard Zarrouk's medium for 48 hours ( $K^+/Cs^+ = 20$ ) (Fig. 4B)	53.3 °C	57.5 °C	3.8°	7.8°	7.2 Jg <sup>-1</sup>	8.5 Jg <sup>-1</sup>

**Table 1: DSC Parameters of The Main Transition Peak Of Native (Fig. 4A) and Processed (Fig. 4B) *Spirulina Platensis* Calculated after Deconvolution**

The comparison of data in Table 1 clearly shows that, although the denaturation temperatures of the main peak are similar in both native and processed samples, their integral enthalpies and denaturation intervals differ markedly. This

suggests that PBCc is partially unfolded after recultivation of samples that were previously cultivated in the mixed medium and then irradiated.



**Figure 5: Heat Absorption Curve as a Function of Temperature. All Data are Recalculated Per Gram of Dry Biomass**

A – Representative deconvoluted DSC curve of *Spirulina platensis* after cultivation in the mixed system of standard and modified Zarrouk's media ( $K^+/Cs^+ = 2.5$ ), subsequent irradiation at a dose of 543 kGy, and recultivation in non-modified Zarrouk's medium for 5–10 days ( $K^+/Cs^+ = 20$ ).

B – DSC curves of four other *Spirulina platensis* samples recorded after different durations of recultivation ( $K^+/Cs^+ = 20$ ).

Fig. 5A, B demonstrate DSC curves of recultivated *Spirulina platensis* previously cultivated in a mixed medium and  $\gamma$ -irradiated to a final dose of 543 kGy using  $^{137}\text{Cs}$ . The recultivation was performed over different time periods

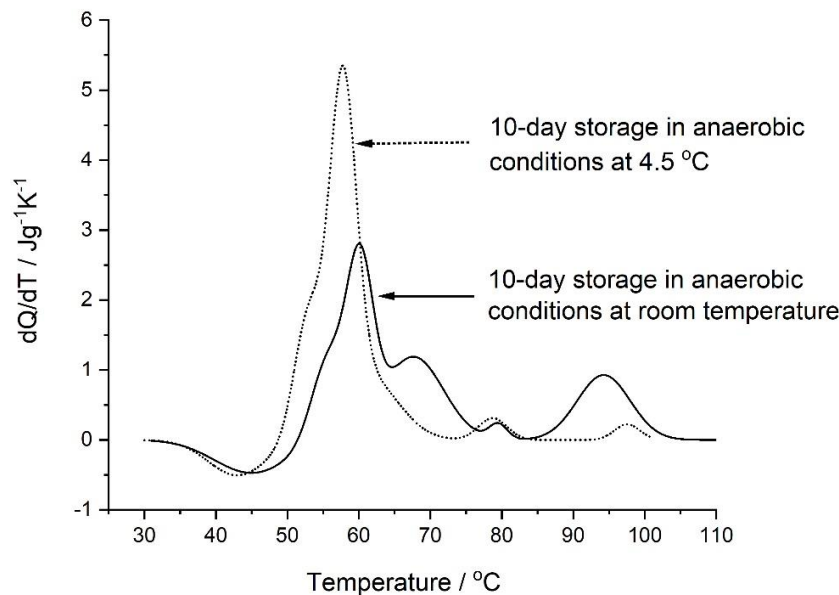
ranging from 5 to 12 days. The thermodynamic parameters of the main peak of the DSC curve shown in Fig. 5A are listed in Table 2.

Sample	Denaturation temperature		Denaturation interval at half height		Denaturation enthalpy	
	$T_{d1}$	$T_{d2}$	$\Delta T_{d1}$	$\Delta T_{d2}$	$\Delta H_{d1}$	$\Delta H_{d2}$
<i>Spirulina platensis</i> after cultivation in the mixed medium ( $K^+/Cs^+ = 2.5$ ) with subsequent irradiation at dose 543 kGy, recultivation in non-modified standard Zarrouk's medium for 10 days ( $K^+/Cs^+ = 20$ ) (Fig. 5A)	55.2 °C	56.0 °C	1.3°	4.2°	6.9 Jg <sup>-1</sup>	13.8 Jg <sup>-1</sup>

**Table 2: DSC Parameters of the Main Transition Peak of Processed and Recultivated *Spirulina Platensis* Suspension (Fig. 5A), Calculated after Deconvolution**

A comparison of the parameters calculated from the curve in Fig. 5A with those in the control (Fig. 4A) clearly demonstrates similarities between the native and recultivated algae. Fig. 5B presents several DSC curves recorded after different

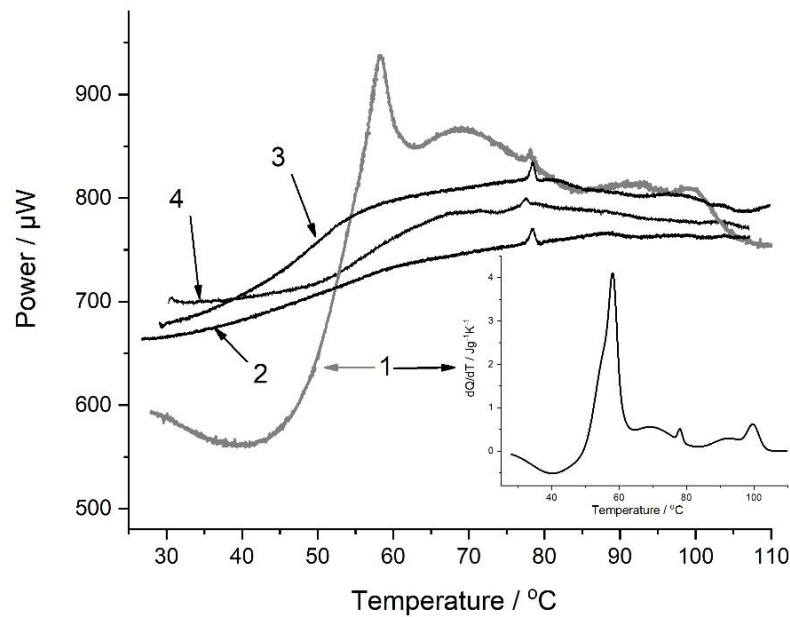
cultivation durations. The shapes of the curves indicate that *Spirulina platensis* gradually recovers its thermodynamic properties with increasing incubation time.



**Figure 6: Two Representative DSC Curves of *Spirulina Platensis* After 10 days of Storage, Following Cultivation in the Mixed System of Standard And Modified Zarrouk's Media ( $K^+/Cs^+ = 2.5$ ), Subsequent Irradiation at a Dose of 543 kGy, and Recultivation in Non-Modified Zarrouk's Medium ( $K^+/Cs^+ = 20$ )**

We also studied the influence of storage duration and temperature on the thermodynamic parameters of *Spirulina platensis* suspension. Fig. 6 presents two representative examples of algae that were cultivated in the mixed medium, irradiated, recultivated, and then stored in non-modified Zarrouk's medium for 10 days. As shown in Fig. 6, storage at 4.5 °C did not cause unfolding of PBCc, as evidenced by the absence of significant changes in transition temperatures

and enthalpies (see Fig.4A). However, at room temperature, a decrease in enthalpy of PBCc was observed, along with a broadening of the main peak and the emergence of a new high-temperature peak. Moreover, the total denaturation enthalpy does not change, which indicates a redistribution of enthalpy between the PBCc and the newly emerged peaks. The nature of the newly emerged high-temperature peaks is not yet known.



**Figure 7: DSC Curves of *Spirulina Platensis* Recorded After Cultivation in the Mixed System Of Standard And Modified Zarrouk's Media ( $K^+/Cs^+ = 2.5$ ), Subsequent Irradiation at Dose 543 kGy, and Recultivation in Non-Modified Zarrouk's Medium for 14 days ( $K^+/Cs^+ = 20$ )**

1. The first scan from 25 °C to 110 °C
2. The same sample cooled to 25 °C for 3 hours, and then re-scanned to 110 °C
3. The same sample cooled again to 25 °C for 3 hours, kept in a measuring cell for 4 days, and scanned again to 110 °C
4. Native *Spirulina platensis* kept in anaerobic conditions at 5 °C for 8 months (long storage)
5. Insert – Processed curve of the first scan

The main transition peak of curve 1 in Fig. 7 is similar to other relevant peaks observed in *Spirulina platensis* [Fig. 5A, B]. This figure reveals the following significant findings: A narrow, distinct peak appears at approximately 77.5 °C in all four curves, indicating that *Spirulina platensis* cells contain a biopolymer or biopolymer domain with a highly cooperative transition ( $\Delta T = 1.0 \pm 0.1$  °C), resembling protein crystals that remain stable after multiple heating cycles to 110 °C. Storage of samples at 25 °C in a sealed calorimeter cell initiated a partial renaturation process, as evidenced by the reshaping of Curve 3 compared to Curve 2. This observation suggests that *Spirulina platensis* possesses a restoration potential even without cultivation. Curve 4 in Fig. 7 represents native *Spirulina platensis* maintained in standard medium under anaerobic conditions at 5 °C for eight months. In this state, the biological activity of the microalga is effectively paused or conserved, yet it remains viable and can be successfully recultivated to yield functionally normal *Spirulina platensis*. It is noteworthy that Curve 3 increasingly resembles Curve 4, which may indicate the sample's ability to fully restore its biological activity and regain its physical and chemical characteristics upon cultivation.

#### 4. Conclusion

Differential scanning calorimetry (DSC) and optical investigations demonstrated that continuous  $\gamma$  irradiation of *Spirulina platensis* suspension at 1.1 Gy/min to a total dose of 543 kGy in a cesium-containing mixed system of standard and modified Zarrouk's media did not cause irreversible

damage to the microalga. On the 2nd and 4th days of recultivation, when *Spirulina platensis* exhibited a clear white color and the biomass concentration was 30–40  $\mu\text{g}/\text{mL}$ , DSC measurements showed a widening of the melting interval and a decrease in the melting enthalpy of the phycobilisome complex (PBCc) compared to the native norm, without a significant change in the denaturation temperature. These results indicate partial unfolding of the highly ordered PBCc. However, after 7 days of recultivation in standard Zarrouk's medium, the DSC profile closely resembled that of the native state, and the  $\Delta H_d$  value was comparable to that of the native *Spirulina platensis* suspension, suggesting recovery of the phycobilisome complex. Based on our previous data demonstrating full restoration of the structural organization and biological functions of microalgae previously irradiated with 400 kGy  $^{137}\text{Cs}$ , we suggest that the changes observed after 48 hours of recultivation are caused by the influence of cesium ions on DNA organization within the nucleoprotein complex, leading to alterations in protein synthesis in *Spirulina platensis*. The changes observed during the early stages of recultivation (days 2–4) are likely due to the influence of  $\text{Cs}^+$  on DNA conformation, as  $\text{Cs}^+$  can induce alterations in the structure of individual segments of double-stranded DNA [10, 11].

Specifically,  $\text{Cs}^+$ , compared to  $\text{K}^+$ , penetrate more deeply into the minor groove of DNA and directly interact with the base pairs [10]. According to  $\text{Cs}^+$  preferentially bind to GC-rich sites that form four-stranded G-quadruplex structures

with diverse conformations, which play an important role in gene expression [25-27]. It is well known that the eukaryotic genome contains more than 300,000 GC-rich sites capable of forming G-quadruplex structures [28-30]. At low concentrations of Cs<sup>+</sup> and K<sup>+</sup> (~10<sup>-3</sup> M), Cs<sup>+</sup> induces weak unfolding of G-quadruplex structures due to competition with K<sup>+</sup>, which increases the thermostability of the G-quadruplex [31]. This potentially leads to a slight decrease in the thermostability of DNA within the nucleoprotein complex of *Spirulina platensis*, which may in turn affect protein synthesis. Increasing the Cs<sup>+</sup> concentration to 200 mM, with K<sup>+</sup> at 0.6 mM, shifts the transition temperature of G-quadruplex structures by 25 °C, leading to complete unfolding of the G-quadruplex [25]. As reported in [10], Cs<sup>+</sup>, as a heavy ion deeply embedded in the minor DNA groove, can alter the internal dynamics of the double helix and influence protein synthesis [32]. Nevertheless, the rapid growth of *Spirulina platensis* in standard Zarrouk's medium, likely associated with the loss of Cs<sup>+</sup>, was accompanied by a rapid restoration of normal PBCc stability.

DSC studies also demonstrate that native *Spirulina platensis*, when heated to 110 °C and cooled to room temperature multiple times, is capable of fully restoring its single peak at 77.5 ± 1.5 °C, with a ΔT of 1 ± 0.1 °C and a ΔC<sub>max</sub> of 0.4 ± 0.1 Jg<sup>-1</sup>K<sup>-1</sup> (see Fig. 7). We suggest that this sharp peak is most likely associated with the melting of a biopolymer with a high degree of structural organization. Analogous highly ordered structures have not been reported for eukaryotic proteins during denaturation in solutions [33]. However, this phenomenon may occur in *Spirulina platensis*, given that such a complex biological system as PBCc — and its main component, C-phycoyanin, which accounts for 20% of PBCc — denatures within a narrow temperature interval of 1.3 °C (see Fig. 5A and Table 2). Storage of the microalga suspension at ~5 °C did not affect PBCc denaturation, whereas storage at 24 °C caused thermal distribution changes between the PBCc and high-temperature transitions that emerged during the storage period. It was also demonstrated that *Spirulina platensis* suspensions exhibit partial self-recovery after thermal denaturation when maintained at room temperature for several days.

The optical data obtained align well with the calorimetry results. After 7 days of recultivation, spectral analysis of the optical characteristics of *Spirulina platensis* revealed a high degree of restoration of bioactive component structures, even after exposure to high doses of ionizing radiation in the cesium-containing medium mixture. These results confirm the high radioresistance of the microalga, enabling it to withstand significant irradiation without severely compromising growth, development, or post-radiation recovery. These findings provide new insights into the resilience of *Spirulina platensis* under extreme conditions and radiation stress, highlighting its potential applications in biotechnology, nutraceutical production, and radiation-protection research – including in future space missions, such as the advancing Mars exploration programs.

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