COMPOSITIONAL ANALYSIS OF Silybum Marianum plant at reduced pressure using calibration-free LIBS

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Abstract. Silybum marianum is an annual herb with a wide range of therapeutic values due to its high nutritional content. The compositional analysis of different parts of this plant, such as seeds and leaves, was performed using calibration-free laser-induced breakdown spectroscopy. The optical emission spectra from laser-generated plasma of seeds and leaves revealed the elemental profile, including Mg, Na, Ca, K, Si, Al, Zn, Rb, Sr, Fe, Ti, Ba, C, O, H, and N. The study was focused on improving LIBS based quantitative analysis by reducing self-absorption using two approaches. In the first approach, a theoretical model based on the internal reference line method and the relation between experimental and theoretical broadening was established to reduce self-absorption in the measured spectral line at atmospheric pressure. In the second approach, the self-absorption was evaluated at reduced pressure, which shows considerably reduced for most of the spectral lines at 10 mbar pressure. Thereafter, the theoretical model was applied at reduced pressure to further correct the line intensities. Finally, the self-absorption corrected line intensities were used for the compositional analysis of the plant using one-line CF-LIBS. The analytical results obtained with both approaches show that the results of the theoretical model at a reduced pressure are in close agreement with the ICP-MS results.

Key words: Silybum marianum plant, self-absorption, theoretical model, reduced pressure, one-line CF-LIBS.

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1. INTRODUCTION

The increased interest in medicinal plants and their global recognition may be attributed to their therapeutic and preventive role in fighting against diseases [1, 2]. The presence of macronutrients and trace metals with adequate concentrations in medicinal plants is important for the regular functioning of the key organs and
metabolic and cellular processes [3, 4]. Among the galaxy of medicinal plants, *Silybum marianum*, sometimes known as milk thistle, is a biennial herb that belongs to the family Asteraceae (Compositae), is native to Asia Minor, Southern Russia, Southern Europe, and Northern Africa, as well as the Mediterranean region, and is currently grown and cultivated all over the world [5, 6]. It is usually found as an annual wild herb in Pakistan and has become a widespread weed in the northwestern part of the country. The main active element of *Silybum marianum* is Silymarin, which is made up of a combination of silydianin, silychristin, silybin A and B, isosilychristin, isosilybin A and B, and taxifolin, among other flavonolignans. *Silybum marianum* has been used in traditional medicine for centuries, primarily to treat disorders of the liver, kidney, gallbladder, and spleen [7]. Various liver diseases, such as cirrhosis, adjuvant therapy in chronic inflammatory liver disease, and hepatitis were treated by *Silybum marianum* [8]. Aside from the hepatoprotective properties, it has anti-carcinogenic, cardioprotective, neuroprotective, anti-inflammatory, and cytoprotective properties [9]. Recent studies revealed that its ingredients are essential in treating cancer, diabetes, and neurodegenerative disorders [10–12]. However, few studies have been performed to determine the mineral elements in *Silybum marianum*. For example, Zafar et al. [13] determined the elemental composition of *Silybum marianum* and other medicinal plants by using atomic absorption spectrometry (AAS) and related its medicinal properties with their elemental distribution. Sobir o’g’li et al. [14] analyzed the macro and micro-elements in dry extract from asparagus seeds and reported that the extract contained balanced macro and micro-elements.

Laser-induced breakdown spectroscopy (LIBS) is a powerful analytical technique used for the qualitative and quantitative analysis of the sample in any phase [15–18]. This technique has become popular due to minimal or no sample preparation, simultaneous multi-elemental analysis, real-time analysis, and easy field-deployable [19–23]. It is widely used for the elemental analysis of different plants [24–26]. Recently, Qasim et al. [26] reported the compositional analysis of the *Maerua oblongifolia* plant using the LIBS technique. The quantitative analysis was performed by CF-LIBS, in which a simplified approach was used to correct the self-absorption for the determination of reliable plasma parameters. The conventional and modified forms of the Saha-Boltzmann plot method were employed to calculate the electron temperature. In comparison, the performance of the modified form of the Saha-Boltzmann plot was found to be better for the strongly self-absorbed lines. Umar et al. [27] probed the micronutrients and hazardous elements in the leaves of *Moringa oleifera* using time-resolved LIBS data and determined the elemental composition by the one-line CF-LIBS method, which utilizes electron number density and electron temperature. Andrade et al. [28] employed LIBS to detect essential and harmful metals in various medicinal plants from Eastern Europe, the principal component analysis (PCA) was combined with LIBS to distinguish the distinct morphological plant parts.

The accuracy of elemental quantitative analysis with LIBS suffers from drawbacks such as self-absorption, matrix effect, and intensity fluctuations in spectral emission intensities. The self-absorption in the emission lines adversely affects the true line profile
of the emission line, resulting in poor accuracy in the measurement of plasma parameters and the quantitative elemental analysis. To reduce the self-absorption effects at atmospheric pressure, several theoretical models have been proposed in the literature [29–31]. Nevertheless, building an ideal self-absorption correction model is difficult due to the complexity of laser-matter interaction, plasma inhomogeneity, and temporal evaluation of the plasma [32]. Researchers developed different experimental approaches to exploit the self-absorption effect [33–35]. The use of reduced air pressure is considered an efficient way to eliminate self-absorption due to a fast expanding plasma plume [36, 37]. Moreover, reducing the air pressure causes better ablation and improved resolution [36]. Hao et al. [33] investigated the self-absorption effect at reduced pressures to perform the quantitative analysis of steel samples. The findings demonstrated that the nonlinearity in the Mn and Cu calibration curves caused by self-absorption was removed at reduced pressure. Similarly, Yalcin et al. [38] explored that the spectral line intensity and signal-to-noise ratio were significantly increased when plasma was produced at 4 Torr, presumably due to plasma cooling at low pressure.

In the literature, self-absorption correction has primarily been performed using theoretical models at atmospheric pressure, but to the best of our knowledge, no study has been reported where theoretical models were used at lower pressure. The objective of the present work is to develop a new approach for self-absorption correction in CF-LIBS to perform an accurate compositional analysis of Silybum marianum. The work for correcting the self-absorption in the measured intensities of the spectral lines is based on the two approaches. In the first approach, a theoretical model involving the internal reference line method and the relation between experimental and theoretical line broadening was developed to reduce self-absorption at atmospheric pressure. In the second approach, a theoretical model was applied to the experimental data recorded at reduced pressure and evaluated by plotting the Saha-Boltzmann plots for the Ca emission lines. The self-absorption corrected emission line intensities were used for the compositional analysis of Silybum marianum, and the results were compared with the ICP-MS results.

2. MATERIAL AND METHODS

2.1. SAMPLES

The seeds and leaves of fresh Silybum marianum plants were collected during the full growing season around the region of Sargodha district in Punjab province, Pakistan at GPS coordinates (32.1087° N, 72.6543° E). Figure 1a depicts a photo of the Silybum marianum plant, while Fig. 1b–c shows a close view of seeds and leaves. The aerial parts of the seeds and leaves were separated and washed properly before being put into the oven to make them fully dry. The samples were handled with care to avoid contamination. The seeds and leaves were powdered and hydraulically pressed at a pressure of 13440 psi for 5 min. The samples were prepared in the form of discs of 3 mm in thickness and 13 mm in diameter.
2.2. EXPERIMENTAL SETUP

The schematic of the experimental setup to produce plasma and record the emission spectra of different parts of *Silybum marianum* plant is shown in Fig. 2. For the generation of plasma plume, the experiments were performed with Q-switched Nd:YAG laser (Q-smart 850 by Quantel) operating at 355 nm, 10 Hz repetition rate, and 5 ns pulse width. The laser at 355 nm can deliver up to 230 mJ of energy. The 25 mJ laser energy was focused on the sample surface, which was placed on the sample holder inside a vacuum chamber. The laser energy was monitored using an energy meter (Maestro-Gentec). The vacuum chamber is made of stainless steel, having ports for laser pulse entrance, plume imaging, target holder, vacuum system, gas inlet, pressure gauge, and optical fiber feed through etc. The rotatory vane pump was used to evacuate the chamber at $10^{-2}$ mbar pressure. A quartz lens with a focal length of 50 cm was used to focus the laser light at 45° to the target surface. The sample holder was rotated after fixed time intervals to provide a fresh surface for every laser shot. To avoid air breakdown in front of the sample surface, the distance between the target and the lens was kept slightly smaller than its focal length. Under these arrangements, the laser induced plasma was produced, and the radiations emitted from the plasma were collected by an optical fiber having a collimator to collect the plasma emissions and transmitted to the LIBS spectrometer (LIBS2500+, Ocean Optics Inc.). The LIBS2500+ system is a multi-channel spectrometer equipped with six HR2000+ spectrometers having
each 10 μm slit width and 2048 element linear silicon CCD arrays. The LIBS2500+ system covers a spectral range of 230–805 nm with 0.1 nm spectral resolution.

To reduce the impact of continuum radiations, the spectra were acquired at a detector gate delay of 2.5 μs and at a fixed integration time of 2.1 ms. The emission signals were corrected by removing the dark signals from the observed signals. The spectral emission obtained through a multi-channel spectrometer was stored on a computer using OOLIBSplus software for further analysis.

2.3. ICP-MS ANALYSIS

The procedure for ICP-MS analysis of seeds and leaves of *Silybum marianum* is the same as described in earlier work [26]. In brief, 50 mg sample powder was digested with 2.0 mL of concentrated nitric acid (HNO₃) at 95°C for 2 hours. After that, 4.0 mL of hydrofluoric acid (HF) solution was added. Then again, 2.0 mL of HNO₃ reagent was digested at 190°C for 30 min. After cooling down the sample, the digestive solution was diluted, filtered, and finally transferred to 100 mL volumetric flask and evenly shaken before putting into ICP-MS system (iCAP Q, ThermoFisher, Waltham, USA).

![Fig. 2 – Schematic of LIBS experimental setup.](image-url)
3. RESULTS AND DISCUSSION

3.1. OPTICAL EMISSION ANALYSIS

The optical emission spectra of seeds and leaves of *Silybum marianum* plant samples were acquired after optimizing the experimental parameters. The spectra were carefully analyzed to identify the emission lines using the NIST database [39]. Moreover, the sample spectra were compared with spectra of various pure elements to further confirm the elemental analysis. In the emission spectra, most of the emission lines were neutral, but a few singly ionized emission lines of sample constituents were also detected.

Figure 3(a, b) shows the emission spectra of seeds and leaves acquired in the 230–805 nm spectral region. The neutral and ionic emission lines of elements are labelled in both spectra, which confirms the presence of Mg, Na, Ca, K, Si, Al, Zn, Rb, Sr, Fe, Ti, Ba, C, O, H, and N. It is observed that the emission lines in the leaf’s spectra are more intense than that of the seed’s spectra, except the emission lines of H and O, which are relatively weaker in the emission spectra of leaves. The spectra of seeds and leaves are dominated by neutral lines of Ca at 443.50 nm, 445.48 nm, 558.88 nm, 585.75 nm, 612.22 nm, 616.22 nm, 643.91 nm, 646.26 nm, 649.38 nm. Besides, the strong resonance line corresponding to a transition 4s4p 1P1 → 4s 2 1S0 at 422.67 nm, and ionic lines at 315.89 nm, 317.93 nm, 370.60 nm, 373.69 nm, along with a strong resonance doublet of ionic lines corresponding to the 4p 3P1/2, 3/2 → 4s 3S1/2 transitions at 393.37 nm, 396.85 nm were also observed.

![Optical emission spectra](image-url)
In the shorter wavelength region, the neutral emission lines of Si at 288.16 nm and C at 247.85 nm were observed, whereas, in the visible region, the CN molecular band was detected at 388.31 nm. Furthermore, the singly ionized Mg resonance doublet consisting of 3p ²P₁/₂, 3/₂ → 3s ²S₁/₂ transitions were detected at 279.55 nm and 280.27 nm, along with multiple structures consisting of 3d ²D₁/₂, 3/₂ → 3p ²P₁/₂, 3/₂ transitions, while resonance line of neutral Mg was detected at 285.21 nm. The spectra show unresolved spectral lines of neutral Mg doublet 3s3d ²D₂, 3 → 3s3p ²P₁, 2 at 383.23 nm and 383.83 nm and triplet structure 3s4s ³S₁ → 3s3p ³P₀, 1, 2 at 516.73 nm, 517.27 nm, and 518.36 nm. The major lines from neutral Al at 308.21 nm, 309.27 nm, 394.40 nm, and 396.15 nm and a few weak lines of neutral Fe along with a prominent line at 438.35 nm were observed in this region of the spectrum. The spectral lines with weak intensities of neutral Zn at 481.05 nm, multiple of neutral Ti lines around 498–501 nm and singly ionized Sr line at 407.77 nm, Ba at 455.40 nm and 493.41 nm were identified in the spectra with good signal-to-background ratio.

In the longer wavelength region of the spectra, a well-known doublet resonance of neutral Na 3p ²P₁/₂, 3/₂ → 3s ²S₁/₂ at 588.99 nm and 589.59 nm and neutral K 4p ²P₁/₂, 3/₂ → 4s ²S₁/₂ at 766.48 nm and 769.90 nm were observed. The spectra also showed that there were weak marker peaks of neutral Rb at 780.02 nm and 794.76 nm. Aside from this, spectral analysis revealed the CaO molecular band around 620–625 nm and the existence of H, N, and O lines. The analysis indicates that Silybum marianum plant can provide sufficient minerals and nutrients. The presence of Ca, K, and Mg as well as the identification of trace elements, is a distinguishing trait that has a metabolic role and can be used medicinally for therapeutic effect [40].

3.2. SELF-ABSORPTION CORRECTION

3.2.1. Theoretical model at atmospheric pressure

A theoretical model was developed to reduce the self-absorption in the measured intensities of the spectral lines acquired from the laser-induced plasma at atmospheric pressure. The theoretical model for self-absorption correction is based on the combination of the internal reference line method and the relation between experimental and theoretical broadening. The internal reference method exploits the internal reference line to determine the self-absorption coefficient (SA) of a spectral line. A line with higher upper energy $E_m$ and lower transition probability $A_{mn}$ can be selected as an internal reference line since such lines are less prone to self-absorption [31]. The self-absorption coefficient (SA)$_{int}$ of the spectral line based on the internal reference line can be evaluated as follows [31]:

$$ (SA)_{int} = \frac{I_{m}^{j} A_{mn} g_{m}}{I_{R}^{mn} A_{ji} g_{j}} \exp \left( \frac{E_{j} - E_{m}}{k_{B}T} \right) $$

where $I_{R}^{mn}$, $A_{mn}$, $g_{m}$ are the signal intensity, transition probability, and statistical weight of internal reference line of a transition having upper-level $m$, respectively.
The parameters $I_{ji}, A_{ji}, E_j,$ and $g_j$ are the measured signal intensity, transition probability, energy, and statistical weight of the spectral line of transition with upper-level $j$ respectively, whereas $k_B$ is the Boltzmann constant, and $T$ is the electron temperature. The SA is then used to correct the spectral line intensity of emission lines. The SA coefficient ranges from 0 to 1, where a smaller value means stronger self-absorption [41]. The self-absorption in the spectral lines of each element was initially checked using Eq. (1) by selecting the internal reference lines in the spectra of the seed and leaf recorded at atmospheric pressure. The determination of coefficients showed self-absorption in Ca, Mg, Na, and K lines. The first correction in the intensity of the self-absorbed spectral line was obtained by dividing its measured intensity by the self-absorption coefficient

$$f_{ji} = \frac{I_{ji}}{(SA)_{int}}$$

The corrected line intensities were used for the determination of electron temperature and for the compositional analysis. As self-absorption affects the line width, and inaccurate full width at half maxima (FWHM) results in an erroneous electron number density. Therefore, $f_{ji}$ may further be corrected by estimating the self-absorption coefficient $(SA)_{Stark}$ of the Stark broadened self-absorbed spectral line as [42]

$$(SA)_{Stark} = \left( \frac{\Delta \lambda}{\Delta \lambda_o} \right)^{1/\alpha}$$

where the value of $\alpha$ is $-0.54$, $\Delta \lambda$ is the full width at half maximum (FWHM) of the measured spectral line determined by Lorentz fitting and $\Delta \lambda_o$ is the expected or theoretical line width of the spectral which is free from self-absorption and correlated with electron number density $n_e$ as:

$$\Delta \lambda_o = 2 \omega \left( \frac{n_e}{10^{16}} \right)$$

where $\omega$ denotes the Stark parameter of the spectral line [43].

The electron number density $n_e$ can be determined by Stark width of hydrogen ($H_\alpha$) line using following relation [44]

$$n_e = 8.02 \times 10^{12} \left( \frac{\Delta \lambda_H}{\alpha_{1/2}} \right)^{3/2} \text{ cm}^{-3}$$

where $\alpha_{1/2}$ is the half width of the reduced Stark profile which is a weak function of electron number density and electron temperature [44]. $\Delta \lambda_H$ is the FWHM of hydrogen $H_\alpha$ line.
Thus, after measuring the value of self-absorption \((SA)_{Stark}\) from Eq. (3), \(I^{ji}\) may further be corrected by the following relation [42]

\[
I_{corrected} = \frac{I^{ji}}{(SA)_{Stark}^{0.46}}
\]  

(6)

The self-absorption coefficients of the spectral lines of the elements, measured using equations (1) and (3) are given in Table 1.

\begin{table}[h]
\centering
\caption{List of self-absorption coefficients of the spectral lines calculated using a theoretical model of self-absorption correction}
\begin{tabular}{lllll}
\hline
Species & Wavelength (nm) & 1st correction of self-absorption coefficient calculated using equation (1) & 2nd correction of self-absorption coefficient calculated using equation (3) \\
\hline
Ca I & 422.67 & 0.13 & 0.05 & \\
& 428.30 & 0.63 & 0.30 & \\
& 428.94 & 0.62 & 0.35 & \\
& 429.90 & 0.71 & 0.30 & \\
& 430.25 & 0.42 & 0.31 & \\
& 430.77 & 0.79 & 0.33 & \\
& 443.50 & 0.61 & 0.31 & \\
& 445.48 & 0.52 & 0.24 & \\
& 612.22 & 1.00 & 0.25 & \\
& 612.22 & 1.00 & 0.26 & \\
& 643.91 & 0.92 & 0.45 & \\
& 646.26 & 0.71 & 0.27 & \\
& 649.38 & 1.00 & 0.49 & \\
Ca II & 315.89 & 0.11 & 0.04 & \\
& 370.60 & 0.40 & 0.07 & \\
& 373.69 & 0.49 & 0.06 & \\
& 393.37 & 0.12 & 0.02 & \\
& 396.85 & 0.11 & 0.02 & \\
Mg I & 278.30 & 0.12 & 0.06 & \\
& 285.21 & 0.01 & 0.002 & \\
& 383.23 & 0.31 & 0.11 & \\
& 383.83 & 0.36 & 0.13 & \\
& 516.73 & 1.00 & 0.11 & \\
& 517.27 & 1.00 & 0.11 & \\
& 518.36 & 0.69 & 0.10 & \\
Mg II & 279.55 & 1.00 & 0.9 & \\
& 280.27 & 0.02 & 0.01 & \\
Na I & 388.99 & 0.40 & 0.11 & \\
& 589.59 & 0.48 & 0.11 & \\
K I & 404.72 & 1.00 & 0.41 & \\
& 693.88 & 0.33 & 0.30 & \\
& 766.49 & 0.21 & 0.10 & \\
& 769.90 & 0.22 & 0.19 & \\
\hline
\end{tabular}
\end{table}
Fig. 4 – Boltzmann plots of the species in the seed spectra: a) before self-absorption correction; b) after self-absorption correction.

The Boltzmann plots of Ca, Mg, and K lines were used as the evaluation parameter for self-absorption correction. Figure 4a shows the Boltzmann plots of Ca, Mg, and K, before SA correction using the spectroscopic parameters taken from the NIST database [39]. The data points are scattered, indicating poor fitting with adjusted $R^2$ values ~0.4, which results in less accurate electron temperature. The average value of electron temperature was extracted as 1.45 eV for the seed spectrum and 1.5 eV for the leaf spectrum. Figure 4b shows the reconstructed Boltzmann plots after SA correction. The data points are now stretched along the fitted line, and the linear fitting coefficients (slope and intercept) were significantly improved. In this case, the average electron temperature of seed and leaf spectra was extracted as 0.90 eV and 0.93 eV, respectively.

3.2.2. Theoretical model at reduced pressure

The theoretical model was employed to correct the self-absorption in the spectral lines of the emission spectra acquired at reduced pressure. The use of reduced pressure is considered an efficient approach for self-absorption reduction. The reduced pressure lowers the plasma density and increases the SA coefficient due to the fast expansion of the plasma plume. The second approach involved a two-step self-absorption correction. In the first step, the seed and leaf spectra of *Silybum marianum* were recorded at various reduced pressures to investigate the self-absorption in the measured spectral lines. The results revealed that the self-absorption was found to be significantly reduced for most of the spectral lines at 10 mbar, with self-absorption coefficients close to 1 except for the few lines including Mg (279.55 nm, 280.27 nm), Ca (393.37 nm, 396.85 nm), Na (588.99 nm, 589.59 nm), and K (766.49 nm, 769.90 nm).
These are resonant and doublet spectral lines that are highly affected by self-absorption. However, self-absorption reduction for these spectral lines was observed at 10 mbar pressure. The variation in the line widths and the relative intensities of these spectral lines at atmospheric and 10 mbar pressure is shown in Fig. 5. Increased line broadening showed significant self-absorption in all the doublet lines of the spectra recorded at atmospheric pressure. However, when the pressure was reduced to 10 mbar, the widths of the doublet lines reduced, the relative intensity ratio improved, and the lines were well separated.

Fig. 5 – Variation in line width and relative intensity of the doublet Mg II, Ca II, Na I, and K I at atmospheric pressure (bottom x-axis, right y-axis) and 10 mbar pressure (bottom x-axis, left y-axis).

Fig. 6 – Variation of: a) full width at half maximum (FWHM); b) self-absorption coefficient (SA) of the spectral lines at various reduced air pressures.
Furthermore, Fig. 6a shows that the FWHMs of the doublet lines reduced with pressure, whereas Fig. 6b shows that the self-absorption coefficients increased at reduced pressure, showing relatively less self-absorption. However, the increase in the SA coefficients of the resonant lines was less compared to that of the non-resonant lines. In the second step, the theoretical model was applied at reduced pressure for further correction of those spectral intensities whose self-absorption coefficients were not close to 1.

The electron number density and electron temperature were extracted after a two-step self-absorption correction using a theoretical model at reduced pressure. The electron number density was determined using the Stark broadened line profile of Ca (612.22 nm), and the electron temperature was determined using Ca lines of two consecutive stages in the Saha-Boltzmann plot [45]. Figure 7 indicates that as the pressure was reduced, the electron temperature and electron number density decreased. At reduced pressures, the plasma expands rapidly due to weak atmospheric confinement, and the electron temperature drops due to weak ionization of the plasma [46]. Besides this, the plasma shielding became weaker at reduced pressures, and more material was ablated, making plasma less prone to self-absorption [47]. From atmospheric pressure to 10 mbar, the electron temperature of seed plasma was decreased from 0.95 to 0.78 eV, and the electron number density was decreased from $2.30 \times 10^{17}$ to $1.26 \times 10^{17}$ cm$^{-3}$.

Similarly, the electron temperature and number density of leaf spectra were reduced from 1.2 to 0.80 eV and $2.9 \times 10^{17}$ to $2.01 \times 10^{17}$ cm$^{-3}$ respectively. Figure 8 shows the comparison of Saha-Boltzmann plots before self-absorption correction at atmospheric pressure and after the two-step self-absorption correction.

Fig. 7–Variations in electron temperature and electron number density at various reduced pressures.
The local thermodynamic equilibrium condition (LTE) can be verified using the McWhirter criteria [48]:

\[ n_e \geq 1.6 \times 10^{12} \frac{T^{1.5}}{(\Delta E)^3} \]  

(7)

where \( \Delta E \) (eV) represents the maximum energy difference between the upper and lower level of a transition detected in the emission spectra, and \( T \) (eV) is the electron temperature. The LTE criteria determined that all the species are subject to the same electron temperature and have a minimum electron number density \( \sim 10^{15} \text{ cm}^{-3} \). In this case, the values of electron number densities determined after self-absorption correction using both approaches were two orders of magnitude greater than the measured ones. Hence, it fulfils the criteria to satisfy the LTE condition. It should be noted that the McWhirter criteria is necessary but insufficient for the plasma to be in LTE.

3.3. COMPOSITIONAL ANALYSIS

The compositional analysis of the *Silybum marianum* plant was performed by one-line calibration free-LIBS (OLCF-LIBS). Like calibration free LIBS, it does not require a reference sample and uses a single spectral line of the species to measure the composition of an element [49, 50]. It relies on the accurate electron temperature and electron number density, followed by fulfilling the LTE assumption, stoichiometric ablation, and an optically thin plasma condition. Therefore, without considering self-absorption in laser-produced plasma, the integrated intensity related to one of
the neutral \( (s = I) \) and ionic \( (s = II) \) emission lines of an element \( \alpha \) can be written as [51]:

\[
\tilde{I}_\alpha^s(\lambda) = \int_{\lambda - \infty}^{\lambda + \infty} I^s_\alpha(\lambda)d\lambda = F\left(\frac{A^s_j g^s_j}{U^s(T)}\right) n^s_\alpha \exp\left(-\frac{E^s_j}{k_B T}\right)
\]

where \( U^s_\alpha(T) \) denotes the specie’s partition function at the electron temperature \( T \), \( (A^s_j)\alpha \) is the transition probability, \( \lambda_j \) denotes the central wavelength of the transition, \( (g^s_j)\alpha \) denotes the specie’s statistical weight of the upper level, \( n^s_\alpha \) denotes the number density of the specie, \( E^s_j \) denotes the energy of the upper level, \( k_B \) is Boltzmann constant, and \( F \) denotes the experimental factor. Due to the constant efficiency of the collection system, the experimental factor is assumed to be constant [52].

In this method, the Boltzmann equation [16, 53] was used to determine the number density of the neutral species in the plasma for elemental composition. The number density of the neutral species \( n^I_\alpha \) was calculated by the integrated intensity \( \tilde{I}_\alpha^I(\lambda) \) of the emission line using the Boltzmann equation, which can be written as:

\[
n^I_\alpha = \left(\frac{U^I(T)\tilde{I}^I(\lambda)}{A^I_j g^I_j}\right) \exp\left[\frac{E^I_j}{k_B T}\right]
\]

The number density of the ionized species \( (n^{II}_\alpha) \) can be determined by the Saha-Boltzmann equation after measuring the \( n^I_\alpha \) in the plasm [54] which is written as:

\[
R_\alpha \equiv \frac{n^{II}_\alpha}{n^I_\alpha} = \frac{6.04 \times 10^{21}(k_B T)^2 U^{II}_\alpha(T)}{n_e U^I_\alpha(T)} \exp\left[-\frac{\chi_\alpha}{k_B T}\right]
\]

where \( \chi_\alpha \) (eV) is the ionization energy of the ionized species.

Since the concentration of the species is related to its number density, Eq. (10) can be written as, \( R_\alpha \equiv \frac{n^{II}_\alpha}{n^I_\alpha} = \frac{c^{II}_\alpha}{c^I_\alpha} \). In the case of stoichiometric ablation, the elemental concentration is determined by adding the concentration of neutral and ionized species, i.e.

\[
C_\alpha \equiv C^I_\alpha + C^{II}_\alpha
\]

The above equation may also be written as:

\[
C_\alpha = C^I_\alpha \left[1 + \left(\frac{c^{II}_\alpha}{c^I_\alpha}\right)^q\right] = C^I_\alpha \left[1 + (R_\alpha)^q\right]
\]
In Eq. (12), $C_\alpha$ can be represented in terms of concentration of neutral atoms if $(s = 1,q = 1)$ and concentration of ionized atoms if $(s = 2,q = -1)$.

The concentration of an element can now be calculated using equations (8) and (12), as follows:

$$C_\alpha = \left[ \frac{\bar{I}^s(\lambda) U^s(T)}{A_j^s J_j^s} \exp \left( \frac{E_j^s}{k_B T} \right) \right]_\alpha [1 + (R_\alpha)^q]$$

Finally, the percentage composition of the element can be measured by the following relation:

$$C_\alpha \% = \frac{C_\alpha \times A_\alpha}{\sum \left( C_\alpha i \times A_\alpha i \right)} \times 100$$

where $A_\alpha$ is the atomic weight of element $\alpha$ in a sample, and $n$ is an integer representing the species index of an element.

Table 2

<table>
<thead>
<tr>
<th>Elements</th>
<th>Seeds concentration (wt.%)</th>
<th>Leaves concentration (wt.%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>OLCF-LIBS Before SA correction</td>
<td>OLCF-LIBS After SA correction</td>
</tr>
<tr>
<td>Ca</td>
<td>36.01</td>
<td>42.60</td>
</tr>
<tr>
<td>Mg</td>
<td>12.33</td>
<td>16.59</td>
</tr>
<tr>
<td>Na</td>
<td>5.05</td>
<td>0.99</td>
</tr>
<tr>
<td>K</td>
<td>25.94</td>
<td>29.87</td>
</tr>
<tr>
<td>Al</td>
<td>0.88</td>
<td>1.49</td>
</tr>
<tr>
<td>Fe</td>
<td>15.03</td>
<td>2.07</td>
</tr>
<tr>
<td>Si</td>
<td>0.26</td>
<td>5.30</td>
</tr>
<tr>
<td>Sr</td>
<td>2.77</td>
<td>0.35</td>
</tr>
<tr>
<td>Zn</td>
<td>0.22</td>
<td>0.34</td>
</tr>
<tr>
<td>Ba</td>
<td>0.03</td>
<td>0.18</td>
</tr>
<tr>
<td>Ti</td>
<td>1.36</td>
<td>0.14</td>
</tr>
<tr>
<td>Rb</td>
<td>0.10</td>
<td>0.90</td>
</tr>
</tbody>
</table>

In the OLCF-LIBS method, the integrated line intensity $\bar{I}_\lambda^s(\lambda)$ is experimentally measured from the plasma emission spectrum, while other parameters can be taken from the atomic spectra NIST database [39]. After determining the electron temperature $T$
and the electron number density $n_e$ to obtain the value of $R_\alpha$, the concentration of the element can be measured. The OLCF-LIBS gives good results without self-absorption; however, self-absorption is inevitable in the experimental spectra. Therefore, the spectral lines of each element were examined, and self-absorption in the selected spectral lines was reduced using both approaches. After self-absorption correction, the plasma parameters, and the partition functions of the species, which are dependent on the measured temperature and used in the computation of $R_\alpha$ and concentration of an element, experience improvements towards their actual values. Finally, the compositional analysis of leaves and seeds of the *Silybum marianum* was performed by the OLCF-LIBS method after the self-absorption correction using both approaches. The consistency of the results of both approaches was validated using ICP-MS results, which are given in Table 2 and shown in Fig. 9. The findings exhibited that OLCF-LIBS gives better results using a theoretical model at reduced pressure than atmospheric pressure.

![Figure 9](image_url)

Fig. 9 – The OLCF-LIBS results of relative concentrations of the elements in the sample: a) seeds; b) leaves of *Silybum marianum* plant obtained after self-absorption correction using theoretical model at atmospheric pressure and two-step correction using theoretical model at reduced pressure. The ICP-MS technique is used to cross-validate the results.

4. CONCLUSIONS

The work demonstrated is a comparative study of compositional analysis using the LIBS technique with two different self-absorption correction approaches to improve the quantitative analysis. The qualitative analysis of the *Silybum marianum* plant confirmed the presence of major (Mg, Na, Ca, K), trace (Si, Al, Zn, Rb, Sr, Fe, Ti, Ba), and organic (O, H, N, C) elements in seeds and leaves. In the first approach, a theoretical model based on the internal reference line method and the relationship between experimental and theoretical broadening was developed to reduce self-absorption.
This approach was used to evaluate and minimize self-absorption in the measured spectral line intensities at atmospheric pressure. The correction was evaluated by drawing the Boltzmann plots for the neutral lines of Ca, Mg, and K. In the second approach, a two-step correction involving a theoretical model at reduced pressure was used. In the first step, the self-absorption at 10 mbar shows that the spectral widths of the self-absorbed lines decreased, and the self-absorption coefficients for most of the lines were close to 1. In the next step, a theoretical model was used to further correct those spectral line intensities whose self-absorption coefficients were not close to 1. The correction was evaluated by drawing the Saha-Boltzmann plots for the Ca lines. Finally, the validated with the ICP-MS results. However, the theoretical model at reduced pressure yields better results than at atmospheric pressure.

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**REFERENCES**


