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## Omicron BA.2.86 Pirola nightmare: Empirical formulas and thermodynamic properties (enthalpy, entropy and Gibbs energy change) of nucleocapsid, virus particle and biosynthesis of BA.2.86 Pirola variant of SARS-CoV-2

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**Abstract:** Similarly to a phoenix, SARS-CoV-2 has appeared periodically in waves. The new variants that appeared through mutations have suppressed earlier variants, causing new waves of the pandemic. The Omicron BA.2.86 Pirola variant is the latest in the sequence. An increased infectivity was noticed, which results in rapid spreading, as well as decreased pathogenicity, which results in a lower number of severe cases. However, in the public there is a fear of further development of the epidemic. This analysis was made with the goal to assess the risks in the period of early 2024. Mutations that were developed by the BA.2.86 variant have led to a change in empirical formula and thermodynamic properties. The empirical formula of the BA.2.86 virus particle is  $\text{CH}_{1.639023}\text{O}_{0.284130}\text{N}_{0.230031}\text{P}_{0.006440}\text{S}_{0.003765}$ . It is different than those of other variants of SARS-CoV-2, other virus species and cellular organisms. The driving force for the virus multiplication, Gibbs energy change of biosynthesis of the BA.2.86 variant is  $-221.75 \text{ kJ C-mol}^{-1}$ . It is more negative than that of its host tissue. According to the biosynthesis phenomenological equation, the more negative Gibbs energy change of biosynthesis allows the virus to achieve a greater biosynthesis rate and hijack the host cell metabolism. However, the Gibbs energy change of biosynthesis of the BA.2.86 variant is similar to those of the CH.1.1 and XBB.1.16 variants. This means that these variants should have similar multiplication rates and thus similar pathogenicity. Therefore, it seems that there is no ground for fear of an extensive spreading of severe forms, but there are reasons for caution and monitoring of the spreading of the epidemic and potential appearance of new mutations. Moreover, unlike the earlier pandemic waves, during the newest pandemic wave, the infections

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with influenza, RSV and BA.2.86 variant simultaneously appeared, which deserves an analysis.

*Keywords:* biothermodynamics; virus-host interaction; COVID-19; pandemic; variant under monitoring; pathogenicity; pathogen.

#### INTRODUCTION

Phoenix is an immortal bird that cyclically regenerates. Like a Phoenix, SARS-CoV-2 has cyclically regenerated several dozen times through mutations from Hu-1 to the newest Omicron BA.2.86 Pirola variant. With every new mutation and new variant, SARS-CoV-2 has obtained a new life appearing slightly different from its predecessor. Some of the variants have caused pandemic waves of high amplitude.<sup>1</sup> Differently from the mythological phoenix, the SARS-CoV-2 phoenix has disappeared and reborn in front of our eyes during the three years of the pandemic. Thus, SARS-CoV-2 has appeared in late-2019 in Wuhan and was labeled as the Hu-1 wild type.<sup>2</sup> The mutations of the virus have occurred mostly in the part of the genome that encodes the spike glycoprotein.<sup>3</sup> However, the mutations have occurred in other viral proteins as well.<sup>4</sup> The evolution of viruses and the formation of new variants has been described in the literature.<sup>5–9</sup>

BA.2.86 Pirola is the latest variant of SARS-CoV-2, which is characterized by many mutations.<sup>10</sup> The number of mutations in BA.2.86 variant, compared to the XBB.1.5 variant is similar to the difference between the first Omicron variant and its predecessor Delta variant.<sup>10</sup> This might give the BA.2.86 variant the ability to infect people who have previously had COVID-19 or who have received COVID-19 vaccines, which has raised public concerns.<sup>10</sup> During the late 2023 and early 2024, the BA.2.86 variant has become widely spread.<sup>11</sup> Even though there has been a decrease in number of daily infections worldwide since late 2022, with the appearance of the new Omicron BA.2.86 variant, the number of COVID-19 cases has increased since the mid-2023. Due to this situation, it would be good to perform a physicochemical analysis of the BA.2.86 variant to compare its ability to infect host cells with that of the previous variants of SARS-CoV-2.<sup>12</sup>

SARS-CoV-2 belongs to the *Coronaviridae* family.<sup>13</sup> It is an enveloped virus, with a single stranded positive sense RNA genome.<sup>13</sup> SARS-CoV-2 virus particles contain four kinds of structural proteins: nucleocapsid (N), membrane (M), envelope (E) and spike (S). The nucleocapsid protein binds to the viral RNA and forms the nucleocapsid.<sup>14</sup> The nucleocapsid is enclosed in a lipid bilayer envelope that contains membrane and envelope proteins.<sup>15</sup> The spike proteins point out from the surface of the virus particle. They represent the virus antigens that bind to host cell receptors.<sup>16–20</sup> Moreover, the SARS-CoV-2 genome encodes the viral proteins needed for multiplication, which have been identified as targets for the antiviral medicines.<sup>51,52</sup>

SARS-CoV-2 belongs to RNA viruses.<sup>21</sup> RNA viruses exhibit a great tendency to mutate.<sup>22,23</sup> Mutations lead to change in information content of the viral genome, chemical changes in elemental composition, as well as thermodynamic properties (enthalpy, entropy and Gibbs energy changes of formation and biosynthesis).<sup>24–26</sup> Mutation as a biological phenomenon, except through sequencing, can be detected through the atom counting method, which allows detection of changes in elemental composition that appear as a consequence of mutations.<sup>27</sup> Furthermore, changes in elemental composition lead to changes in thermodynamic properties.<sup>28–31</sup>

Since 2019, in the literature, elemental composition and thermodynamic properties have been reported for several virus species: Ebola,<sup>32</sup> Mpox,<sup>33</sup> SARS-CoV-2,<sup>8,9,16,20,24,25,34</sup> HIV,<sup>35</sup> arboviruses,<sup>35</sup> bacteriophages,<sup>36</sup> *etc.* Biothermodynamic mechanisms that influence infectivity and pathogenicity of different variants and the consequences on epidemiology and mechanisms of spreading of SARS-CoV-2 are available in the literature.<sup>37–42</sup>

The aim of this paper is to explore the changes in empirical formula, molar mass, biosynthesis reactions and thermodynamic properties (enthalpy, entropy, Gibbs energy changes) of formation and biosynthesis of the Omicron BA.2.86 Pirola variant. Based on the obtained results, the goal is to perform an assessment of the risk of spreading of an epidemic/pandemic of the BA.2.86 variant in early 2024. Moreover, the pathogenicity of the BA.2.86 variant will be compared to those of the earlier variants of SARS-CoV-2.

## METHODS

### *Data sources*

The genetic sequence of the Omicron BA.2.86 Pirola variant of SARS-CoV-2 was taken from GISAID, the global data science initiative.<sup>43</sup> It can be found under the accession number EPI\_ISL\_18138566 and is labelled hCoV-19/USA/OH-ODH-SC3032044/2023. Thus, the findings of this study are based on the metadata associated with one sequence available on GISAID up to September 24, 2023, and accessible at <https://doi.org/10.55876/gis8.230924yd> (please see the Supplementary material to this paper).

The sequence of the nucleocapsid phosphoprotein of SARS-CoV-2 was obtained from the NCBI database,<sup>44</sup> under the accession number QIK50455.1. The sequence of the membrane protein of SARS-CoV-2 was obtained from the NCBI database,<sup>44</sup> under the accession number QHR63293.1. The sequence of the spike glycoprotein of SARS-CoV-2 was obtained from the NCBI database,<sup>44</sup> under the accession number QHR63290.2. The number of protein copies in the virus particle was taken from literature.<sup>45</sup> In a SARS-CoV-2 particle, there are 2368 copies of the nucleocapsid phosphoprotein, 1184 copies of the membrane protein and 222 copies of the spike glycoprotein.<sup>45</sup>

The standard Gibbs energy changes of biosynthesis of the wild type Hu-1, Delta B.1.617.2, Zeta P.2, Eta B.1.525, Theta P.3, Iota B.1.526, Lambda C.37, Mu B.1.621, Kappa B.1.617.1, Omicron B.1.1.529, Omicron BA.2, Omicron BA.2.75, Omicron BQ.1, Omicron BQ.1.1, Omicron XBB, Omicron XBB.1, Omicron BA.5.2, Omicron BF.7, Omicron XBB.1.5,

Omicron BN.1, Omicron CH.1.1, Omicron XBC, Omicron XBB.1.9.1, Omicron XBF and Omicron XBB.1.16 variants of SARS-CoV-2 were taken from the literature.<sup>6-9,18,19,24-26</sup>

#### Empirical formulas

The empirical formulas and molar masses of the virus particle and nucleocapsid of the Omicron BA.2.86 Pirola variant of SARS-CoV-2 were determined through the atom counting method as described in references.<sup>27,46</sup> The atom counting method is implemented with a computer program, based on genetic sequences, protein sequences and morphological data.<sup>27</sup>

#### Thermodynamic properties of live matter

Thermodynamic properties of virus particle and nucleocapsid of the Omicron BA.2.86 variant were determined with the Patel–Erickson model<sup>29</sup> and Battley model.<sup>28</sup> To find enthalpy of live matter (*i.e.*, virus particle or nucleocapsid) with the Patel–Erickson model, the empirical formula is used to find the number of electrons transferred to oxygen during complete oxidation,  $E$ , with the equation:<sup>29</sup>

$$E = 4n_C + n_H - 2n_O - 0 n_N + 5n_P + 6n_S \quad (1)$$

$E$  is then used to find standard enthalpy change of combustion of live matter,  $\Delta_C H^0$ , with the equation:

$$\Delta_C H^0(\text{bio}) = -111.14 \frac{\text{kJ}}{\text{C-mol}} \times E \quad (2)$$

$\Delta_C H^0$  is then used to calculate standard enthalpy change of formation of live matter,  $\Delta_f H^0$ , with the equation:<sup>29</sup>

$$\begin{aligned} \Delta_f H^0(\text{bio}) = n_C \Delta_f H^0(\text{CO}_2) + \frac{n_H}{2} \Delta_f H^0(\text{H}_2\text{O}) + \frac{n_P}{4} \Delta_f H^0(\text{P}_4\text{O}_{10}) + \\ + n_S \Delta_f H^0(\text{SO}_3) - \Delta_C H^0 \end{aligned} \quad (3)$$

Entropy of live matter is calculated with the Battley model, based on its elemental composition. Standard molar entropy of live matter,  $S_m^0$ , is given by the equation:

$$S_m^0(\text{bio}) = 0.187 \sum_J \frac{S_m^0(J)}{a_J} n_J \quad (4)$$

where  $S_m^0(J)$  is standard molar entropy of element  $J$ ,  $a_J$  number of atoms of element  $J$  in its standard state elemental form, and  $n_J$  the number of atoms of element  $J$  in the empirical formula of live matter.<sup>28</sup> The summation is over all elements  $J$  of which the live matter consists.<sup>28</sup> The Battley model can also be used to find standard entropy change of formation of live matter,  $\Delta_f S^0$ ,<sup>28</sup> if the constant 0.187 is changed to  $-0.813$ :

$$\Delta_f S^0(\text{bio}) = -0.813 \sum_J \frac{S_m^0(J)}{a_J} n_J \quad (5)$$

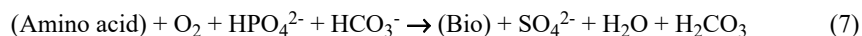
Finally,  $\Delta_f S^0$  and  $\Delta_f H^0$  are combined to find standard Gibbs energy change of formation,  $\Delta_f G^0$ , of live matter

$$\Delta_f G^0(\text{bio}) = \Delta_f H^0(\text{bio}) - T \Delta_f S^0(\text{bio}) \quad (6)$$

where  $T$  is temperature.<sup>47</sup>

*Biosynthesis reactions*

Biosynthesis reactions of the virus particle and nucleocapsid of the Omicron BA.2.86 variant were formulated based on their empirical formulas. Biosynthesis reactions are macrochemical equations of conversion of nutrients into new live matter in metabolism.<sup>47</sup> The general biosynthesis reaction for viruses has the form:



where (Amino acid) represents a mixture of amino acids, which has the empirical formula  $\text{CH}_{1.798}\text{O}_{0.4831}\text{N}_{0.2247}\text{S}_{0.022472}$ .<sup>6,8,9,17,23</sup> Newly synthesized live matter, (Bio), is represented with its empirical formula.<sup>6,8,9,17,23</sup> The source of energy, carbon, nitrogen and sulfur for biosynthesis are the amino acids.<sup>6,8,9,17,23</sup> The electron acceptor is  $\text{O}_2$ .<sup>6,8,9,17,23</sup> The source of phosphorus is  $\text{HPO}_4^{2-}$ .<sup>6,8,9,17,23</sup> Excess  $\text{H}^+$  generated during biosynthesis are absorbed by the  $\text{HCO}_3^-$ , which is a part of the bicarbonate buffer.<sup>6,8,9,17,23</sup> Excess sulfur atoms are released in the form of  $\text{SO}_4^{2-}$ , which is an additional metabolic product.<sup>6,8,9,17,23</sup> The oxidized carbon atoms are released in the form of  $\text{H}_2\text{CO}_3$ , which is also a part of the bicarbonate buffer.<sup>6,8,9,17,23</sup>

*Thermodynamic properties of biosynthesis*

Thermodynamic properties of biosynthesis of the virus particle and nucleocapsid of the Omicron BA.2.86 variant of SARS-CoV-2 were calculated with the Hess's law. They were found based on the biosynthesis reactions and thermodynamic properties of live matter. Thermodynamic properties of biosynthesis include standard enthalpy change of biosynthesis,  $\Delta_{\text{bs}}H^0$ , standard entropy change of biosynthesis,  $\Delta_{\text{bs}}S^0$ , and standard Gibbs energy change of biosynthesis,  $\Delta_{\text{bs}}G^0$ .<sup>47</sup> They can be found by application of the Hess's law to the biosynthesis reactions:

$$\Delta_{\text{bs}}H^0 = \sum_{\text{products}} \nu \Delta_f H^0 - \sum_{\text{reactants}} \nu \Delta_f H^0 \quad (8)$$

$$\Delta_{\text{bs}}S^0 = \sum_{\text{products}} \nu S_m^0 - \sum_{\text{reactants}} \nu S_m^0 \quad (9)$$

$$\Delta_{\text{bs}}G^0 = \sum_{\text{products}} \nu \Delta_f G^0 - \sum_{\text{reactants}} \nu \Delta_f G^0 \quad (10)$$

where  $\nu$  represents a stoichiometric coefficient.<sup>6,8,9,17,23,29,47</sup>

## RESULTS AND DISCUSSION

*Empirical formula and thermodynamic properties of live matter*

The empirical formula of the virus particle of the Omicron BA.2.86 variant of SARS-CoV-2 is reported for the first time:

$\text{CH}_{1.639023}\text{O}_{0.284130}\text{N}_{0.230031}\text{P}_{0.006440}\text{S}_{0.003765}$  (Table I).

Empirical formulas have been reported in the literature for other SARS-CoV-2 variants. The empirical formula of the virus particle of the Hu-1 wild type of SARS-CoV-2 is  $\text{CH}_{1.6390}\text{O}_{0.2851}\text{N}_{0.2301}\text{P}_{0.0065}\text{S}_{0.0038}$ .<sup>25</sup> The empirical formula of the virus particle of the Delta variant of SARS-CoV-2 is:

$\text{CH}_{1.6383}\text{O}_{0.2844}\text{N}_{0.2294}\text{P}_{0.0064}\text{S}_{0.0042}$ .<sup>25</sup>

The virus particle of the Omicron BA.1 variant of SARS-CoV-2 is characterized by the empirical formula  $\text{CH}_{1.6404}\text{O}_{0.2842}\text{N}_{0.2299}\text{P}_{0.0064}\text{S}_{0.0038}$ .<sup>25</sup> The empirical formula of the virus particle of the Omicron BA.2 variant of SARS-CoV-2 is

$\text{CH}_{1.6403}\text{O}_{0.2838}\text{N}_{0.2298}\text{P}_{0.0064}\text{S}_{0.0038}$ .<sup>26</sup> Moreover, empirical formulas of other virus species have been reported in the literature. The empirical formula of a *Poxviridae* virus particle is  $\text{CH}_{1.5876}\text{O}_{0.3008}\text{N}_{0.2538}\text{P}_{0.00223}\text{S}_{0.00554}$ .<sup>33</sup> A *Vaccinia* virus particle is characterized by the empirical formula  $\text{CH}_{1.5877}\text{O}_{0.3232}\text{N}_{0.2531}\text{P}_{0.00371}\text{S}_{0.00540}$ .<sup>33</sup> Therefore, every virus species and variant is characterized by a different empirical formula. Based on the empirical formula, it is possible to identify the virus. This provides a rapid method for virus identification through single particle inductively coupled plasma mass spectroscopy, as described by Degueldre.<sup>34</sup>

TABLE I. Empirical formulas and molar masses of the Omicron BA.2.86 Pirola variant of SARS-CoV-2. Empirical formulas have the general form  $\text{CH}_{n_{\text{H}}}\text{O}_{n_{\text{O}}}\text{N}_{n_{\text{N}}}\text{P}_{n_{\text{P}}}\text{S}_{n_{\text{S}}}$ , where  $n_{\text{H}}$ ,  $n_{\text{O}}$ ,  $n_{\text{N}}$ ,  $n_{\text{P}}$  and  $n_{\text{S}}$  are numbers of H, O, N, P and S atoms in the empirical formula, respectively. Molar masses were reported in two forms: molar mass of the empirical formula,  $Mr$ , and total molar mass of the macromolecular assembly (entire virus particle or entire nucleocapsid),  $Mr(\text{tot})$

Parameter	Virus particle	Nucleocapsid
$n_{\text{H}}$	1.639023	1.570946
$n_{\text{O}}$	0.284130	0.343118
$n_{\text{N}}$	0.230031	0.312432
$n_{\text{P}}$	0.006440	0.006007
$n_{\text{S}}$	0.003765	0.003349
$Mr / \text{g C-mol}^{-1}$	21.75	23.75
$Mr(\text{tot}) / \text{MDa}$	219.7	117.6

Empirical formulas have been reported in the literature for various species of cellular organisms. The empirical formula of *Escherichia coli* (bacteria) is:

$\text{CH}_{1.918}\text{O}_{0.528}\text{N}_{0.257}\text{P}_{1.76 \times 10^{-2}}\text{S}_{5.54 \times 10^{-3}}\text{K}_{5.87 \times 10^{-3}}\text{Mg}_{2.07 \times 10^{-3}}\text{Ca}_{8.36 \times 10^{-4}}\text{Mn}_{9.89 \times 10^{-6}}\text{Fe}_{7.82 \times 10^{-5}}\text{Cu}_{1.62 \times 10^{-6}}\text{Zn}_{2.41 \times 10^{-5}}$ .<sup>31</sup> The empirical formula of *Penicillium chrysogenum* (mold fungi) is:

$\text{CH}_{2.026}\text{O}_{0.511}\text{N}_{0.185}\text{P}_{9.15 \times 10^{-3}}\text{S}_{4.17 \times 10^{-3}}\text{K}_{3.45 \times 10^{-3}}\text{Mg}_{1.47 \times 10^{-3}}\text{Ca}_{3.69 \times 10^{-4}}\text{Mn}_{1.08 \times 10^{-5}}\text{Fe}_{9.51 \times 10^{-5}}\text{Cu}_{1.24 \times 10^{-6}}\text{Zn}_{2.15 \times 10^{-5}}$ .<sup>31</sup> *Saccharomyces cerevisiae* (yeast fungi) is characterized by an empirical formula:

$\text{CH}_{1.613}\text{O}_{0.557}\text{N}_{0.158}\text{P}_{0.012}\text{S}_{0.003}\text{K}_{0.022}\text{Mg}_{0.003}\text{Ca}_{0.001}$ .<sup>28</sup> The empirical formula of the human organism is:

$\text{CH}_{1.7296}\text{O}_{0.2591}\text{N}_{0.1112}\text{P}_{0.0134}\text{S}_{0.003}\text{Na}_{0.0027}\text{K}_{0.0031}\text{Ca}_{0.0173}\text{Cl}_{0.0018}$ .<sup>48</sup>

The empirical formula of the virus particle of the Omicron BA.2.86 variant of SARS-CoV-2 is  $\text{CH}_{1.639023}\text{O}_{0.284130}\text{N}_{0.230031}\text{P}_{0.006440}\text{S}_{0.003765}$  (Table I). Therefore, every class of organisms is characterized by a unique empirical formula different than those of other organisms.

Except for its empirical formula, the Omicron BA.2.86 variant of SARS-CoV-2 has its characteristic thermodynamic properties of live matter (enthalpy, entropy and Gibbs energy change), which were determined in this research

(Table II). The Gibbs energy change of the formation of the Omicron BA.2.86 virus particle is  $-24.64 \text{ kJ C-mol}^{-1}$ , while that of the BA.2.86 nucleocapsid is  $-33.32 \text{ kJ C-mol}^{-1}$  (Table II). Therefore, the virus particle has a greater (less negative) Gibbs energy change than the nucleocapsid. This means that the virus particle has a greater usable energy content. The reason for this are the lipids in the viral envelope. The SARS-CoV-2 virus particle contains a lipid envelope.<sup>13</sup> The lipids in the envelope have a high energy content.<sup>49</sup> Therefore, the usable energy content of the virus particle is greater than that of the nucleocapsid.

TABLE II. Thermodynamic properties of live matter of the Omicron BA.2.86 variant of SARS-CoV-2: standard enthalpy change of formation,  $\Delta_f H^0$ , standard molar entropy,  $S_m^0$ , and standard Gibbs energy change of formation,  $\Delta_f G^0$

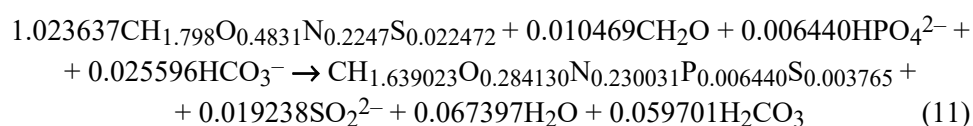
Name	$\Delta_f H^0 / \text{kJ C-mol}^{-1}$	$S_m^0 / \text{J C-mol}^{-1} \text{K}^{-1}$	$\Delta_f G^0 / \text{kJ C-mol}^{-1}$
Virus particle	-64.43	30.70	-24.64
Nucleocapsid	-75.41	32.47	-33.32

The Gibbs energy changes of formation have been reported in the literature for other virus species and variants. The virus particle of the Hu-1 wild type of SARS-CoV-2 is characterized by a Gibbs energy change of formation  $-24.8 \text{ kJ C-mol}^{-1}$ .<sup>25</sup> Gibbs energy change of formation of the virus particle of the Omicron BA.2.86 variant of SARS-CoV-2 is  $-24.64 \text{ kJ C-mol}^{-1}$  (Table II). Thus, Gibbs energy change of formation of the BA.2.86 variant is different than that of the Hu-1 wild type. Moreover, Gibbs energy change of a *Poxviridae* virus particle is  $-25.3 \text{ kJ C-mol}^{-1}$ ,<sup>33</sup> while that of a *Vaccinia* virus particle is  $-30.0 \text{ kJ C-mol}^{-1}$ .<sup>33</sup> Thus, the virus particle of the Omicron BA.2.86 variant of SARS-CoV-2 has a different Gibbs energy change of formation than those of the *Vaccinia* and *Poxviridae* virus particles. Therefore, every virus species and variant has a characteristic Gibbs energy change of formation.

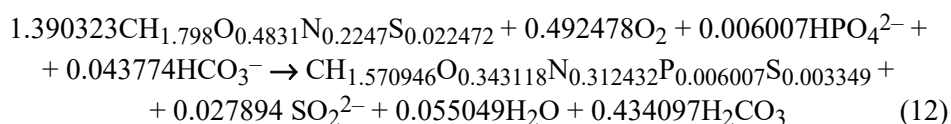
Gibbs energy changes of formation of cellular microorganisms can also be found in the literature. Gibbs energy change of formation of some cellular microorganisms are:  $-66.98 \text{ kJ C-mol}^{-1}$  for *Escherichia coli* bacteria,  $-87.07 \text{ kJ C-mol}^{-1}$  for *Saccharomyces cerevisiae* yeast fungi and  $-18.99 \text{ kJ C-mol}^{-1}$  for *Penicillium chrysogenum* mold fungi.<sup>30</sup> Thus, the Gibbs energy changes of these cellular microorganisms are different than that of the Omicron BA.2.86 variant of SARS-CoV-2 ( $-24.64 \text{ kJ C-mol}^{-1}$ ). Furthermore, the Gibbs energy change of formation of the human organism is  $-37.54 \text{ kJ C-mol}^{-1}$ ,<sup>48</sup> which is different than that of the Omicron BA.2.86 variant of SARS-CoV-2. This means that every class of organisms should have a characteristic Gibbs energy change of formation, summarizing the usable energy content in its life matter.

*Biosynthesis reaction and thermodynamic properties of biosynthesis*

Based on the empirical formulas of the virus particle and nucleocapsid of the Omicron BA.2.86 Pirola variant of SARS-CoV-2, biosynthesis reactions were formulated (Table III). The biosynthesis reaction of the virus particle of the Omicron BA.2.86 variant is:



where  $\text{CH}_{1.798}\text{O}_{0.4831}\text{N}_{0.2247}\text{S}_{0.022472}$  is the empirical formula of amino acids and  $\text{CH}_{1.639023}\text{O}_{0.284130}\text{N}_{0.230031}\text{P}_{0.006440}\text{S}_{0.003765}$  is the empirical formula of the BA.2.86 virus particle (Table I). The biosynthesis reaction of the nucleocapsid of the Omicron BA.2.86 variant is:



where  $\text{CH}_{1.570946}\text{O}_{0.343118}\text{N}_{0.312432}\text{P}_{0.006007}\text{S}_{0.003349}$  is the empirical formula of the BA.2.86 nucleocapsid (Table I). The biosynthesis reaction of the BA.2.86 virus particle contains both amino acids and carbohydrates as an energy source, while that of the BA.2.86 nucleocapsid contains only amino acids. This means that the biosynthesis of the BA.2.86 virus particle takes more energy than the biosynthesis of the nucleocapsid alone. The reason for this is the higher energy content in the virus particle, due to the lipids in the viral envelope, as discussed above. The lipids in the viral envelope have a high energy content.<sup>49</sup> This means that the virus particle that contains the lipid envelope takes more energy for biosynthesis than the nucleocapsid which doesn't contain lipids. This energy comes

TABLE III. Biosynthesis stoichiometry for the Omicron BA.2.86 variant of SARS-CoV-2. The general biosynthesis reaction has the form (Amino acid) +  $\text{CH}_2\text{O}$  +  $\text{O}_2$  +  $\text{HPO}_4^{2-}$  +  $\text{HCO}_3^-$  → (Bio) +  $\text{SO}_4^{2-}$  +  $\text{H}_2\text{O}$  +  $\text{H}_2\text{CO}_3$ . "Amino acid" represents a mixture of amino acids with the formula  $\text{CH}_{1.798}\text{O}_{0.4831}\text{N}_{0.2247}\text{S}_{0.022472}$ . "Bio" represents the empirical formula of live matter from Table I

Role	Name	Virus particle	Nucleocapsid
Reactants	Amino acid	1.023637	1.390323
	$\text{CH}_2\text{O}$	0.010469	0.000000
	$\text{O}_2$	0.000000	0.492478
	$\text{HPO}_4^{2-}$	0.006440	0.006007
	$\text{HCO}_3^-$	0.025596	0.043774
Products	Bio	1.000000	1.000000
	$\text{SO}_4^{2-}$	0.019238	0.027894
	$\text{H}_2\text{O}$	0.067397	0.055049
	$\text{H}_2\text{CO}_3$	0.059701	0.434097



from the carbohydrates in the biosynthesis reaction. The biosynthesis reaction of the BA.2.86 virus particle requires more hydrogen phosphate ion than that of the nucleocapsid.  $\text{HPO}_4^{2-}$  is the phosphorus source for biosynthesis. The higher amount of  $\text{HPO}_4^{2-}$  in the biosynthesis reaction is due to phospholipids in the envelope of the virus particle.

Based on the biosynthesis reactions, the thermodynamic properties of biosynthesis of the BA.2.86 variant were determined for the first time. The enthalpy change of biosynthesis of the BA.2.86 variant nucleocapsid is  $-232.88 \text{ kJ C-mol}^{-1}$  (Table IV). This means that the enthalpy of biosynthesis contributes favourably to the biosynthesis process. The entropy of biosynthesis of the BA.2.86 nucleocapsid is  $-37.48 \text{ kJ C-mol}^{-1}$  (Table IV). The negative entropy change is unfavourable for the biosynthesis reaction. The Gibbs energy change of biosynthesis of the BA.2.86 variant is  $-221.75 \text{ kJ C-mol}^{-1}$ . The negative Gibbs energy change, which is due to the negative enthalpy change of biosynthesis, means that the biosynthesis process is thermodynamically favourable.

TABLE IV. Thermodynamic properties of biosynthesis for the Omicron BA.2.86 variant of SARS-CoV-2: standard enthalpy change of biosynthesis,  $\Delta_{\text{bs}}H^0$ , standard entropy change of biosynthesis,  $\Delta_{\text{bs}}S^0$ , and standard Gibbs energy change of biosynthesis,  $\Delta_{\text{bs}}G^0$

Name	$\Delta_{\text{bs}}H^0 / \text{kJ C-mol}^{-1}$	$\Delta_{\text{bs}}S^0 / \text{J C-mol}^{-1} \text{ K}^{-1}$	$\Delta_{\text{bs}}G^0 / \text{kJ C-mol}^{-1}$
Virus particle	-4.80	6.94	-6.94
Nucleocapsid	-232.88	-37.48	-221.75

#### *Virus–host and virus–virus interactions*

Gibbs energy change of biosynthesis represents the driving force for the biosynthesis process.<sup>47</sup> A more negative Gibbs energy change of biosynthesis,  $\Delta_{\text{bs}}G$ , implies a greater biosynthesis rate,  $r_{\text{bs}}$ , according to the biosynthesis phenomenological equation:

$$r_{\text{bs}} = -\frac{L_{\text{bs}}}{T} \Delta_{\text{bs}}G \quad (13)$$

where  $L_{\text{bs}}$  is the biosynthesis phenomenological coefficient and  $T$  is temperature.<sup>8,23,25</sup> Gibbs energy change of the biosynthesis of the nucleocapsid of the BA.2.86 Pirola variant of SARS-CoV-2 is  $-221.75 \text{ kJ C-mol}^{-1}$  (Table IV). On the other hand, Gibbs energy change of biosynthesis for the lung tissue is  $-49.76 \text{ kJ C-mol}^{-1}$ .<sup>32</sup> Therefore, the BA.2.86 variant has a much more negative Gibbs energy change of biosynthesis. This means that, according to the biosynthesis phenomenological equation, the biosynthesis rate of the BA.2.86 variant will be much greater than that of its host tissue. Due to this, the infected host cells will produce the virus particles at a much greater rate than their own building blocks. This allows the hijacking of the host cell metabolism by the virus. The virus and its host cell compete for the cellular metabolic machinery and resources. The

competition occurs in the host cell cytoplasm, at the ribosomes. The virus has a much greater driving force of biosynthesis, in the form of negative Gibbs energy. This means that the virus will have a much greater biosynthesis rate, which will allow it to hijack the host cell metabolism.

The Gibbs energy change of biosynthesis is proportional to the biosynthesis rate of a virus, according to the biosynthesis phenomenological equation. In case that several virus species or virus variants are simultaneously in circulation in the population, the virus with the most negative Gibbs energy change of biosynthesis will have a competitive advantage.<sup>18,50</sup> The virus characterized by a more negative Gibbs energy change of biosynthesis will have a greater biosynthesis rate.<sup>18,50</sup> This will allow it to dominate over other viruses circulating in the population.<sup>18,50</sup> Gibbs energy changes of biosynthesis of SARS-CoV-2 variants are shown in Fig. 1. The Gibbs energy change of biosynthesis of the nucleocapsid of the BA.2.86 Pirola variant of SARS-CoV-2 is  $-221.75$  kJ C-mol<sup>-1</sup> (Table IV). Gibbs energy changes of biosynthesis of nucleocapsids of other variants under monitoring are  $-221.21$  kJ C-mol<sup>-1</sup> for the Omicron CH.1.1 variant<sup>6</sup> and  $-221.19$  kJ C-mol<sup>-1</sup> for the Omicron XBB.1.16 variant.<sup>8</sup> Therefore, the Gibbs energy changes of biosynthesis of the BA.2.86, CH.1.1 and XBB.1.16 variants are very similar. This means that in case these SARS-CoV-2 variants appear in a population, they will have very similar biosynthesis rates. This means that no variant will have an advantage in the competition in a short time period. As a result, all three variants should circulate in the population during a pandemic. However, having in mind that even though it is small, the difference in Gibbs energy changes of biosynthesis exists, which is the most negative for the BA.2.86 variant, which means that in a long time period, it will be able to suppress the CH.1.1 and XBB.1.16 variants.

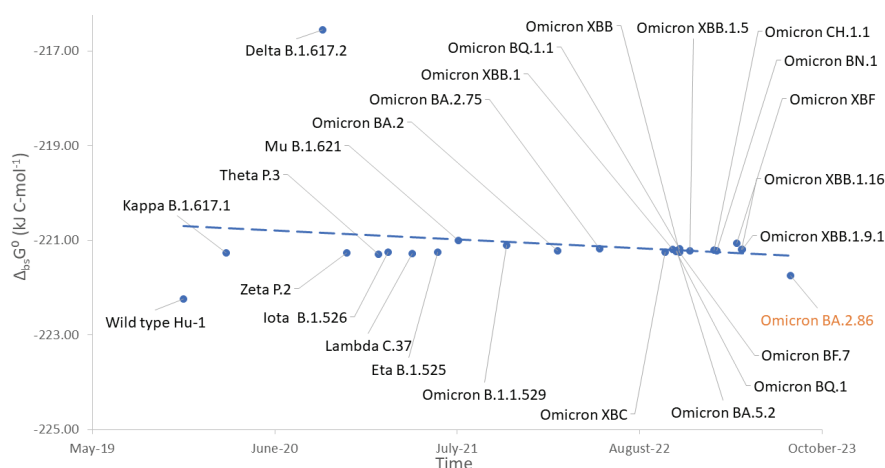


Fig. 1. Gibbs energy changes of biosynthesis of SARS-CoV-2 variants.

The concern expressed in the social media, concerning the greater pathogenicity of the new BA.2.86 variant seems not to be reasonable, since its Gibbs energy change of biosynthesis is only slightly different than that of the other variants. The epidemiological measures that were undertaken in the fight against the other variants that caused the pandemic should result in an adequate response against the spreading of the BA.2.86 variant. However, the data related to kinetics of binding of the new variant to the host cell receptors are still not available. Therefore, in this work, it is not possible to predict with certainty the potential changes in infectivity of the new BA.2.86 variant compared to the other variants of SARS-CoV-2.

The latest wave caused by the Omicron BA.2.86 variant has shown a specific aspect. Unlike the earlier waves caused by earlier SARS-CoV-2 variants, during the latest wave, the epidemics appeared in parallel caused by other viruses. For example, in Serbia, the cases caused by infection with influenza were registered simultaneously. Having in mind that in the same place at the same time, at least 3 different viruses appeared, there was competition between them. The competition between SARS-CoV-2 and other viruses was reported in literature.<sup>6, 50</sup> We must have in mind that the pandemic wave that was reported in literature<sup>50</sup> was of high intensity, with a large number of infected people and a small number with people with natural or artificial immunity. The reported biothermodynamic properties have shown that in that epidemic wave, there was interference. This means that SARS-CoV-2 dominated, while influenza, parainfluenza and RSV were suppressed, because their biothermodynamic properties were not favourable. In the late 2023 and early 2024, the situation is completely different, since the extent of vaccination against COVID-19 was far greater and also the natural immunity of the population was also greater. Thus, the intensity of the pandemic caused by the Omicron BA.2.86 variant is much lower and is about several thousand new cases daily. This has led to a “dilution” of the virus in the population and therefore decreased the ability of spreading. Therefore, despite the still unfavourable thermodynamic properties, there has been a parallel development of epidemics, caused by different viruses. From this we can conclude that even though thermodynamic properties of the antigen–receptor binding and the thermodynamic properties of multiplication of the virus play a biologically important role in the development of the pandemic, an important role is played by epidemiological measures, in the sense of isolation and vaccination. Therefore, vaccination remains the primary method in the fight against the epidemic.

#### CONCLUSIONS

This research reports for the first time the empirical formula, molar mass, biosynthesis reactions and thermodynamic properties (enthalpy, entropy and Gibbs energy changes) of formation and biosynthesis of the Omicron BA.2.86

Pirola variant of SARS-CoV-2. The empirical formula of the BA.2.86 virus particle is  $\text{CH}_{1.639023}\text{O}_{0.284130}\text{N}_{0.230031}\text{P}_{0.006440}\text{S}_{0.003765}$ , which has a molar mass of 21.75 g/C-mol. The empirical formula of the BA.2.86 variant is different than the empirical formulas of other SARS-CoV-2 variants, other virus species and cellular organisms.

The standard Gibbs energy change of formation of the BA.2.86 virus particle is  $-24.64 \text{ kJ C-mol}^{-1}$ , while that of the BA.2.86 nucleocapsid is  $-33.32 \text{ kJ C-mol}^{-1}$ . Gibbs energy change of formation of the virus particle is less negative than that of the nucleocapsid, which implies a greater usable energy content of the virus particle. This is due to the structure of the virus particle. The virus particle is enveloped and contains lipids, which have a high usable energy content and are not present in the nucleocapsid.

The nucleocapsid of the BA.2.86 variant is characterized by a Gibbs energy change of biosynthesis of  $-221.75 \text{ kJ C-mol}^{-1}$ . Gibbs energy change of biosynthesis of the BA.2.86 variant is more negative than that of its host tissue. The more negative Gibbs energy change of biosynthesis means that the virus will have a greater biosynthesis rate than the host tissue, according to the biosynthesis phenomenological equation. The greater biosynthesis rate means that an infected host cell will produce more virus particles than its own building blocks. This allows the virus to hijack the host cell.

Gibbs energy change of biosynthesis of the BA.2.86 variant is very similar to those of the other variants under monitoring: CH.1.1 and XBB.1.16. The Gibbs energy change of biosynthesis represents the driving force for biosynthesis of virus particles and is proportional to their biosynthesis rate. Since the BA.2.86, CH.1.1 and XBB.1.16 variants have similar Gibbs energy change of biosynthesis, they will have similar biosynthesis rates. This means that they will have very similar pathogenicity.

#### SUPPLEMENTARY MATERIAL

Additional data and information are available electronically at the pages of journal website: <https://www.shd-pub.org.rs/index.php/JSCS/article/view/12862>, or from the corresponding author on request.

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ИЗВОД  
ОМИКРОН ВА.2.86 ПИРОЛА НОЋНА МОРА: ЕМПИРИЈСКЕ ФОРМУЛЕ И  
ТЕРМОДИНАМИЧКЕ ОСОБИНЕ (ПРОМЕНЕ ЕНТАЛПИЈЕ, ЕНТРОПИЈЕ И ГИБСОВЕ  
ЕНЕРГИЈЕ) НУКЛЕОКАПСИДА, ВИРУСНЕ ЧЕСТИЦЕ И БИОСИНТЕЗЕ ВА.2.86  
ПИРОЛА ВАРИЈАНТЕ

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Слично фениксу, SARS-CoV-2 се периодично појављивао у таласима. Нове варијанте које су се појавиле кроз мутације потиснуле су раније варијанте, што је изазвало нове таласе пандемије. Омикрон ВА.2.86 Пирола варијанта је најновија у низу. Уочена је повећана инфективност, што резултира брзим ширењем, као и смањена патогеност, што резултира мањим бројем тешких случајева. Међутим, у јавности постоји страх од даљег развоја епидемије. Ова анализа је урађена са циљем да се процене ризици у периоду од почетка 2024. године. Мутације које је развила варијанта ВА.2.86 довеле су до промене емпиријске формуле и термодинамичких особина. Емпиријска формула ВА.2.86 вирусне честице је  $\text{C}_{\text{H}_{1,639023}\text{O}_{0,284130}\text{N}_{0,230031}\text{P}_{0,006440}\text{S}_{0,003765}}$ . Она се разликује се од других варијанти SARS-CoV-2, других врста вируса и ћелијских организама. Погонска сила за умножавање вируса, промена Гибсове енергије биосинтезе варијанте ВА.2.86 је  $-221,75 \text{ kJ C}^{-1}\text{mol}^{-1}$ . Она је негативнија од промене Гибсове енергије биосинтезе ткива домаћина. Према феноменолошкој једначини биосинтезе, негативнија промена Гибсове енергије биосинтезе омогућава вирусу да постигне већу брзину биосинтезе и преузме метаболизам ћелије домаћина. Међутим, промена Гибсове енергије биосинтезе варијанте ВА.2.86 је слична оној код варијанти СН.1.1 и ХВВ.1.16. То значи да ове варијанте треба да имају сличне брзине размножавања, а самим тим и сличну патогеност. Дакле, чини се да нема основа за страх од екстензивног ширења тешких облика, али постоје разлози за опрез и праћење ширења епидемије и потенцијалне појаве нових мутација. Штавише, за разлику од ранијих пандемијских таласа, током најновијег пандемијског таласа, истовремено су се појавиле инфекције инфлуенце, RSV и варијанте ВА.2.86, што заслужује анализу.

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