

# C0r0n@ 2 Inspect

Review and analysis of scientific articles related to experimental techniques and methods used in vaccines against c0r0n@v|rus, evidence, damage, hypotheses, opinions and challenges.

**Thursday, August 12, 2021**

## **Lactose Free Dairy Products and Graphene Oxide Possible Cause of Lactose Intolerance?**

### **Reference**

Trusek, A. ; Dworakowska, D. ; Czyzewska, K. (2020). 3D enzymatic preparations with graphene oxide flakes and hydrogel to obtain lactose-free products. Food and Bioproducts Processing, 121, pp. 224-229. <https://doi.org/10.1016/j.fbp.2020.03.002>

### **Introduction**

1. Lactose intolerance is a problem caused when the small intestine is unable to produce "lactase" which is the enzyme in charge of processing and converting "lactose" into glucose and galactose. This can lead to diarrhea, gas, bloating, heavy digestion, after ingesting or eating dairy products.
2. Lactose-free dairy products are usually developed to facilitate digestion for intolerant people, avoiding the conditions and problems already mentioned. In this post, it is discovered that many methods used to make lactose-free products involve the use of graphene oxide.

### **Facts**

1. Research by (Trusek, A.; Dworakowska, D.; Czyzewska, K. 2020) shows that " *GO graphene oxide can be used as a vehicle in enzyme immobilization* " which allows inhibiting the enzyme responsible for generating lactose in fermentation processes of dairy products .
2. To this end, " *the article develops a method of chemical activation before the binding of the enzymatic molecules. This property of graphene oxide makes it possible to immobilize the enzyme  $\beta$ -galactosidase, after the activation of GO with divinyl sulfone* ".  $\beta$ -galactosidase (Beta galactosidase) is the enzyme used to obtain lactose-free products. The  $\beta$ -galactosidase is responsible for fermenting the sugars of lactose, allowing the production of cheese, yogurt and other dairy products, therefore it is an enzyme that catalyzes the hydrolysis of galactosides to monosaccharides. As for the " *divinyl sulfone* " it is a chemical compound with a molecular structure "C<sub>4</sub>H<sub>6</sub>O<sub>2</sub>S" that can be derived from mustard gas ( Grant, WM; Thomas, CC 1987), considered a dangerous product due to its toxic, corrosive and irritating properties. Divinyl sulfone has been used in other cases to make porous hydrogels (Sannino, A.; Madaghiale, M.; Conversano, F.; Mele, G.; Maffezzoli, A.; Netti, PA; Nicolais, L. 2004), drug encapsulating agents (Morales-Sanfrutos, J.; Lopez-Jaramillo, FJ; Elremaily, MA; Hernández-Mateo, F.; Santoyo-Gonzalez, F. 2015) or the activation of the properties of other compounds such as [chitosan](#) ( Pinheiro, BB; Rios, NS; Aguado, ER; Fernandez-Lafuente, R.; Freire, TM; Fachine, PB; Goncalves, LR 2019).

4. The authors state that the lactose separation process with this method was effective and fast, being " possible to obtain a low concentration of lactose in the effluent stream at a very low temperature, 6 ° C, which corresponds to the storage conditions of the refrigerated milk ". For this, two fundamental problems had to be solved; on the one hand, the enzymatic separation, solved by combining the GO flakes with divinyl sulfone to separate and isolate the  $\beta$ -galactosidase enzyme, and on the other, the development of a less expensive manufacturing method. The authors summarize it like this "The main disadvantage of flake graphene-based vehicles is the difficulty of separating them from solution due to the size and density of the particles. This disadvantage creates problems when preparing the carrier and during its application. In previous work, a GO flake separation method was developed, based on ultra-fast centrifugation (Trusek, A. 2019). The method was efficient, but expensive, particularly in industrial-scale application. To eliminate this problem, 3D preparations based on GO flake encapsulation were proposed. The new method combines hydrogel and flake applications of GO" Although the method allows to eliminate lactose at low cost, the authors do not explain its elimination process, in fact they assume the release of graphene flakes in the solution combined with the dairy product, as stated in the following paragraph." The encapsulation of graphene flakes in the hydrogel allowed an easy creation of the preparations, as well as their packaging in the column of the chemical distillation reactor. There were no hydraulic resistances as the substrate stream flowed through the column. In addition, the alginate capsules were not destroyed, which prevented the release of graphene flakes into the solution." This raises many unresolved questions and problems in the article, since the food product is in direct contact with the graphene flake solution.
5. Among the conclusions, it should be noted that " the procedure developed for the preparation of 3D carriers can be used for any enzyme ". This means that graphene oxide together with other components can be used to inhibit or immobilize all kinds of enzymes. This statement is corroborated by many other researchers, as can be seen in (Zhang, J.; Zhang, F.; Yang, H.; Huang, X.; Liu, H.; Zhang, J.; Guo, S. 2010 ), who consider graphene oxide as " the matrix for enzyme immobilization ", in fact state that " immobilization of the enzyme on GO sheets could easily take place without using crosslinking reagents or further modification of the surface. This is due to the adsorption capacity of graphene, which causes enzyme inhibition, as seen in the immobilization reaction rate distribution graph per minute, see figure 1.

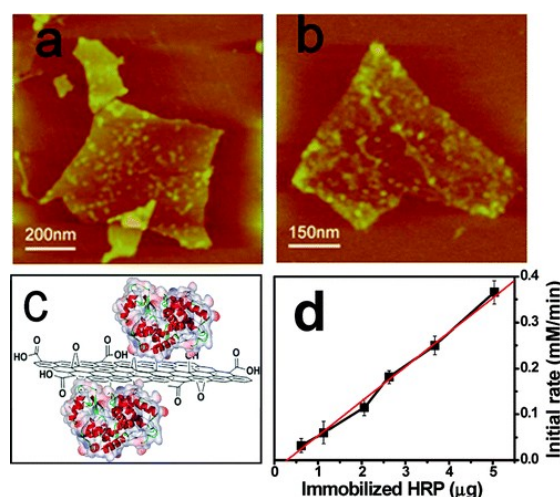


Fig. 1. Graphene oxide samples, enzyme immobilization process diagram and immobilization distribution per minute. (Zhang, J.; Zhang, F.; Yang, H.; Huang, X.; Liu, H.; Zhang, J.; Guo, S. 2010)

Since graphene oxide has special properties for the immobilization of enzymes, this technique has been studied for the immobilization of hydrolases ( Husain, Q. 2016 ), which are a family of digestive enzymes, among which is the " lactase " for being in the EC3 classification under the code " 3.2.1.108 " of the subfamily of " glucosidases ". According to ( Husain, Q. 2016 ) it is possible to immobilize hydrolases with magnetic nanoparticles, including [graphene oxide in combination with magnetite Fe3O4](#). Therefore, it is possible that GO graphene oxide alone, or combined with other components in the small intestine, may inhibit " lactase ", responsible for digesting lactose, causing " lactose intolerance ". This immobilization effect is also confirmed by ( Chen, L.; Wei, B.; Zhang, X.; Li, C. 2013 ) who curiously uses graphene and Fe2O3 aerogels (or what is the same trioxide of dihierro), with " a higher saturation magnetization ( 23–54 emu / g-1 ) ". The " emu / g"(electromagnetic unit / gram) is a magnetic measurement unit to define the ratio of magnetization per mass. Another similar study is that of ( Jiang, B .; Yang, K .; Zhao, Q .; Wu, Q .; Liang , Z .; Zhang, L .; Zhang, Y. 2012 ) in which " *magnetic graphene oxide nanocomposites modified with Fe3O4 nanoparticles managed to immobilize trypsin .* " Trypsin is an essential enzyme of digestion, produced in the pancreas and secreted in the duodenum to hydrolyze peptides that facilitate the absorption of proteins from food.

## Other studies

1. Other studies confirm the involvement of graphene oxide in the preparation of lactose-free dairy products. It is the case of ( Morelos-Gomez, A .; Terashima, S .; Yamanaka, A .; Cruz-Silva, R .; Ortiz-Medina, J .; Sánchez-Salas, R .; Endo, M. 2021 ) that develop graphene oxide membranes for lactose-free milk. The membranes are capable of filtering lactose with a lactose permeation capacity greater than 2.5kg per m2 per day. The authors state that " *Molecular dynamics (MD) simulations demonstrate that lactose exhibits primarily weak van der Waals interactions with GO layers, allowing lactose to diffuse through nanochannels in GO membranes, whereas fat and protein are preserved in milk*". These filtration properties are similar to those found in wastewater filtration ( Fathizadeh, M.; Xu, WL; Zhou, F.; Yoon, Y.; Yu, M. 2017 | Wang, J.; Zhang, P.; Liang, B.; Liu, Y.; Xu, T.; Wang, L.; Pan, K. 2016), including uranium (Hu, X.; Wang, Y.; Yang, JO; Li, Y.; Wu, P .; Zhang, H.; Liu, Z. 2020 | Li, Z.; Chen, F.; Yuan, L.; Liu, Y.; Zhao, Y.; Chai, Z.; Shi, W. 2012) However , like the other studies, residues or traces of graphene in milk and other dairy products are not analyzed, which can [cause intoxication or poisoning of consumers, see all side effects and damage caused by graphene oxide](#).
2. Relying on the enzymatic immobilization properties of graphene oxide, the work of (de-Brito, AR; de-Carvalho-Tavares, IM; de-Carvalho, MS; de-Oliveira, AJ; Salay, LC; Santos deserves special attention, AS ; Franco, M. 2020) that study the interaction of "lactase" in a matrix of carbon nanotubes CNT, which are cylindrical graphene nanosheets. The researchers demonstrate that " *there was adsorption of the enzyme lactase in the tubular area of the carbon nanotubes. By fluorescence spectroscopy analysis, it was observed that the fluorescence emission of lactase is mainly due to the tryptophan (Trp) residue, and this fluorescence was reduced in the presence of CNT, demonstrating the interaction between these components*". This statement reinforces the thesis that graphene oxide could inhibit or immobilize many of the digestive enzymes, causing problems of lactose intolerance, among other possible incidents that have not yet been described.

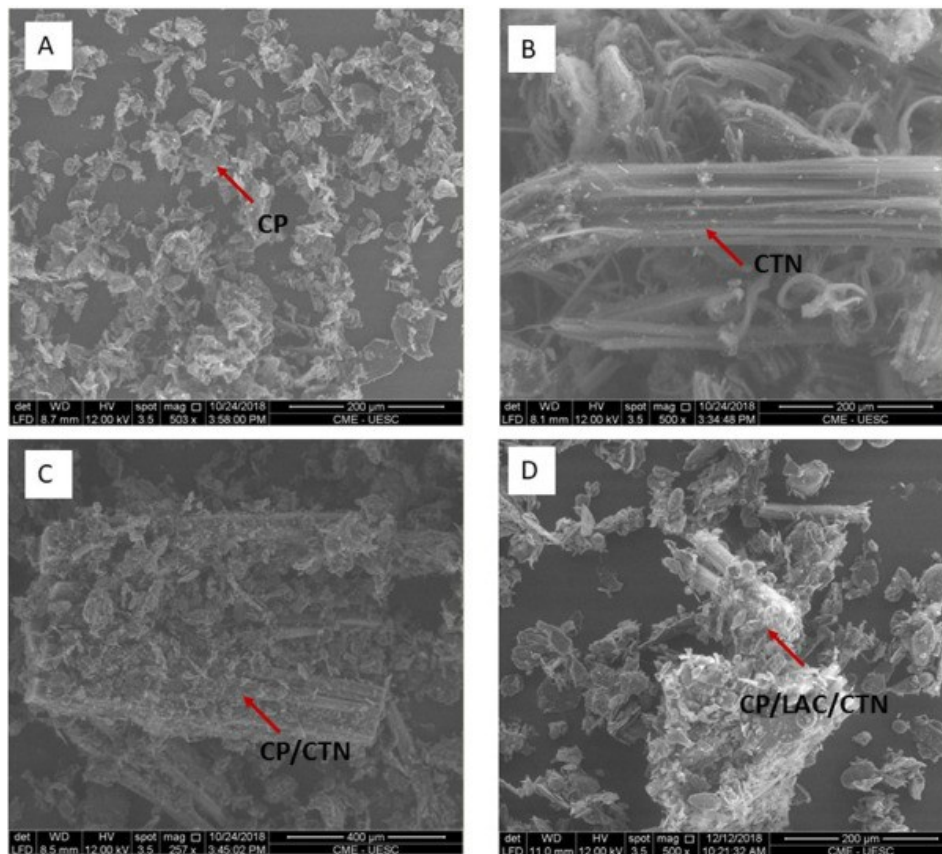


Fig. 2. SEM microscopy of the enzyme immobilization experiment with carbon nanotubes CNT (graphene oxide). (de-Brito, AR; de-Carvalho-Tavares, IM; de-Carvalho, MS; de-Oliveira, AJ; Salay, LC; Santos, AS; Franco, M. 2020)

- Another example of enzymatic immobilization is that of ( Zhou, L.; Jiang, Y.; Gao, J.; Zhao, X.; Ma, L.; Zhou, Q. 2012 ), in whose study " *glucose oxidase*" with graphene oxide. Glucose oxidase is the enzyme that helps break down sugars to facilitate metabolism. Obviously, if glucose oxidase is immobilized, metabolic dysfunctions could occur. Although the study was oriented to the development of bioelectrodes and biosensors, This shows that graphene oxide can be dangerous for the body's metabolic function, as suggested and stated (Papi, M .; Lauriola, MC; Palmieri, V .; Ciasca, G .; Maulucci, G .; De-Spirito , M. 2015 | Volkov, Y .; McIntyre, J .; Prina-Mello, A. 2017 | Zhang, Y .; Qin, L .; Sun, J .; Chen, L .; Jia, L .; Zhao, J .; Sang, W. 2020 | Jastrzębska, AM, Kurtycz, P. and Olszyna, AR 2012 | Singh, Z. 2016 | Jarosz, A .; Skoda, M .; Dudek, I .; Szukiewicz, D. 2016 | Montagner, A .; Bosi, S .; Tenori, E .; Bidussi, M .; Alshatwi, AA; Tretiach, M .; Syrgiannis, Z. 2016 ).

## Reviews

- It has been shown that lactose-free dairy products could be the result of the application of graphene oxide filtration techniques, aimed at the elimination of lactose. However, in none of the studies consulted, the possible residues of graphene oxide in food products are analyzed and neither does it point to the toxicity and adverse effects it causes in the human body. It is essential to identify which lactose-free dairy products have been subjected to these processes in order to detect food contamination. This requires laboratory analysis.

4. The researchers corroborate the ability of graphene oxide in nanoparticles in isolation and in combination with other magnetic elements such as Fe<sub>2</sub>O<sub>3</sub> and Fe<sub>3</sub>O<sub>4</sub>, to inhibit or immobilize all types of enzymes present in the small intestine. The incidence of enzymes in other organs has not yet been reviewed, but the scientific literature confirms that the involvement could be more extensive, since graphene oxide affects the metabolism of cells, see (Jarosz, A.; Skoda, M.; Dudek, I.; Szukiewicz, D. 2016) where [oxidative stress and the disruption of mitochondrial homeostasis are explained](#).

## Hypothesis

1. People inoculated with the COVID-19 vaccine could develop lactose intolerance and problems derived from the immobilization of enzymes, given the ability of graphene oxide to infer its proper functioning. It could also be the case that people with lactose intolerance have high concentrations of graphene oxide or metallic nanoparticles in the small intestine. On the other hand, graphene oxide could affect the normal functioning of trypsin, given its ability to inhibit enzymes, which would explain indigestion, nausea, reflux, abdominal pain and even diarrhea.

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