Nanoparticles for Glioblastoma Treatment

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Abstract. Glioblastoma multiforme is a primary brain tumor whose diagnosis carries with it a dismal prognosis for survival. The development of nanomedicine would lay a path to cross the hurdles that current treatments fail to overcome: the blood-brain barrier (BBB) and the tumor's immune microenvironment. Targeted drug delivery systems are responsible for releasing the chemotherapeutic drug into specific tumor cells, which in addition to allowing crossing the BBB, reduces the damage caused to healthy cells in conventional chemotherapy. However, this type of therapy is still in its infancy and its health effects are still being studied using murine models. The present project aims to determine whether the use of nanoparticles in targeted drug delivery for the treatment of glioblastoma has an inhibitory effect on tumor cell growth, so a systematic review was developed using a defined search strategy using the key terms focused on the research question. The steps and guidelines defined in the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) for systematic reviews were followed. The analysis of the data extracted from the articles included in the review indicates that there is an inhibitory effect on the proliferative activity of tumor cells and a reduction in tumor size when nanoparticles are used to encapsulate drugs in targeted delivery.

Keywords: Nanoparticles · Drug delivery · Cancer

1 Introduction

Glioblastoma multiforme (GBM) is the most common and most harmful primary brain tumor (Fig. 1). It accounts for 57% of gliomas and 48% of all primary malignant tumors of the central nervous system. In Mexico, available statistical data indicate that gliomas account for 33% of all brain tumors and the average age at diagnosis of GBM is 46.4 years [1]. In general, the survival prognosis is less than two years. Standard treatment is multimodality. It includes maximal resection surgery, followed by radiotherapy and chemotherapy treatments using temozolomide (TZM), which administered to GBM cells causes double-strand breaks in DNA, cell cycle arrest and eventual cell death. However, TMZ attacks DNA indiscriminately, causing damage to the patient's hematopoietic stem

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cells as well. Due to the low effectiveness in eliminating the tumor in its entirety and the high recurrence rate, there is a need to look for better treatment options, one of them being targeted drug delivery [2, 3].

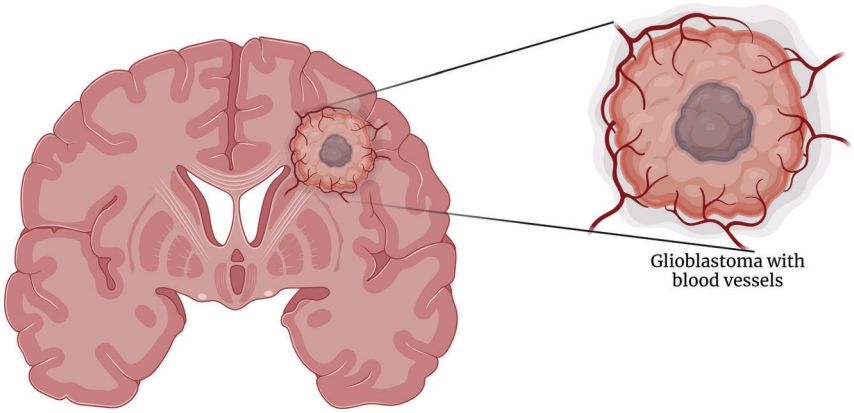

Fig. 1. Representation of the brain tumor

In recent years, vast scientific evidence has accumulated from in vitro and in vivo experiments explaining the influence of the physicochemical properties of nanomaterials on their distribution and effects in the nervous system. The study of such interactions ranges from an overview of the interaction of nanoparticles with cells [4] to the internal benchmarking of a cellular model of the human blood-brain barrier for screening nanoparticle uptake and transcytosis [5] through toxicity and modeling studies [6–10]. An interesting review paper reported in ref. [11] shows the importance of elucidating the covalent and non-covalent interactions of nanoparticle surfaces with biomolecules and biological barriers. The interactions between engineered nanomaterials and biosystems are complex and their net effects on effector activity are largely unknown, thus requiring robust and precise characterization to advance the development of nanomedicine systems.

Biological factors such as the blood-brain barrier (BBB) (Fig. 2) and the immune microenvironment of the tumor hinder the development of new therapies. In recent years, the use of nanoparticles in drug delivery systems has received much attention as they represent a possible alternative to cross the BBB [12]. The biological surface interactions of nanomaterials are strongly influenced by physicochemical properties, such as surface charge and morphology. In the literature there are interesting works dedicated to elucidating how the variation of the shape of the NPs impacts BBB passage, systemic circulation cellular uptake, and hemorheological dynamics [11].

The ability to cross the BBB would reduce the therapeutic limitations faced by targeted drug delivery systems. The use of nanoparticles has been studied for their potential use to be accumulated in tumor areas to inhibit tumor cell proliferation and metastasis effectively, either on their own or in combination with other therapies such as photothermal therapy, photodynamic therapy, or chemotherapy, among others [13]. It is becoming increasingly clear that altering the shape, size and charge of nanoparticles can influence

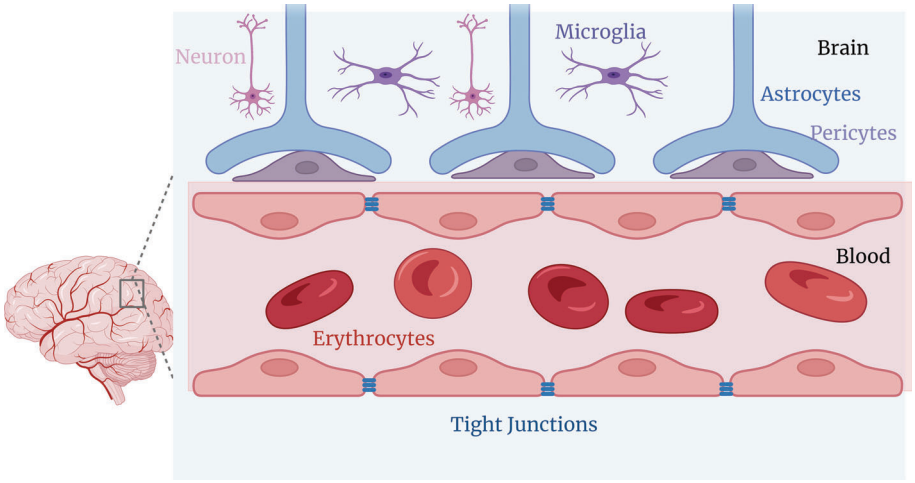

Fig. 2. Representation of the blood-brain barrier

their uptake in the brain, thereby increasing efficacy and bioavailability for the treatment of glioblastoma multiforme. Elucidating the physicochemical properties of nanoparticles in the treatment of glioblastoma that penetrate the BBB and have an inhibitory effect on tumor volume may guide efforts to find the most promising nanomedicine systems in the fight against this type of cancer. Therefore, we have posed the following research question: Does the use of nanoparticles in targeted drug delivery to treat glioblastoma multiforme have an inhibitory effect on tumor cell growth in preclinical animal models?

2 Methods

The protocol was developed using the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) registered in [14]. The search for articles published in ScienceDirect, PubMed, Web of Science and Nature was defined from the search strategy: (glioblastoma) AND (nanoparticles) AND ("drug delivery") AND ("mice") AND ("tumor growth" OR "cell growth" OR "tumor size") AND ("inhibition" OR "inhibit$"). Papers were included in the review if they met the following inclusion criteria: the article is a research of the use of nanoparticles in the targeted administration of drugs for the treatment of glioblastoma that mention whether or not there were changes in tumor growth or cell growth; the article reports the ability of nanoparticles to cross the blood-brain barrier and their effects on tumor cells, whether in inhibiting tumor growth, reducing the proliferative activity of cancer cells or changes in tumor volume; the studies carried out in murine model; the article must have been published from 2015 to 2021 in the English language. On the other hand, articles that fell into one or more of the following exclusion criteria were not considered in the review: the article reports the use of nanoparticles for the localization of glioblastoma by imaging, not including their use in therapy; the article is a review, or a book chapter, or an editorial, or a paper of conference, or unpublished data; full access to the text is not available. As can be

seen, physicochemical properties such as size or shape were not stated in the criteria. Therefore, we speculate to find reports of various morphologies and sizes. Risk of bias assessment was conducted with SYRCLE for animal studies.

3 Results and Discussion

Initially, there were a total of 411 records, of which 96 were catalogued as systematic reviews or book chapters and 174 did not focus on the use of nanoparticles in the therapy of glioblastoma but referred to their use in other types of cancer, or mentioned glioblastoma, but their focus was on another type of therapy Fig. 3.

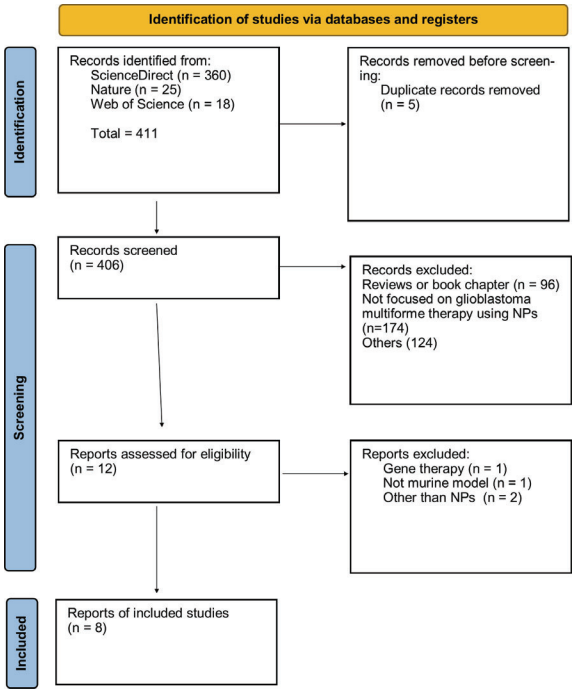

Fig. 3. PRISMA flow chart

Of the remaining 136 articles, only 12 could be accessed in full text. Four articles were eliminated for the following reasons: one article mentioned the use of nanoparticles to encapsulate siRNA for use in GBM therapy, however, the project is focused on targeted drug delivery and the use of siRNA is aimed at gene silencing, so it would not be possible to make a comparison between the two topics; One article was eliminated because no in vivo studies were carried out in murine animal models, it only mentioned in vitro assays; two articles were eliminated because despite mentioning nanobioconjugates, they did not mention the use of nanoparticles in drug delivery systems [15–22].

In all the studies, a saline group was used as a control, a group with the free drug and a combination of the drug encapsulated in the NPs in question. It can be observed

that the groups of NPs that apart from encapsulating the drug were also functionalized with some compound, tend to present better results than their counterpart of NPs and encapsulated drug only. Using the SYRCLE tool for risk of bias assessment in animal model studies, most of the articles did not include key points to minimize the risk of bias in their wording. As a result, the generation of the random sequence as well as the concealment of sequence assignment are unclear, the studies only mentioned that the mice were randomly divided into the test groups without mentioning how this was determined. It is also unclear whether the researchers were unaware of who the mice belonging to the different groups were at the time of treatment.

The characteristics of the nanoparticles used in the articles comprising the review are presented in Table 1. PEG/PLGA polymeric nanoparticles are among the most widely used due to their high level of biocompatibility and biodegradability [15]. The prevalent shape in the case of polymeric NPs tends to be spherical and the surface charge is negative, as indicated by the zeta potential. NPs with values between −10 and 10 mV are approximately neutral, as is the case in refs. [16, 17] NPs with values greater than +30 or −30 mV are considered strongly anionic and cationic, respectively. The PLGA NPs of reference [15] have a zeta potential value that confers stability, since they are the closest to −25 mV, a value that usually leads to a certain degree of stability [23]. The zeta potential also affects the tendency of the NPs to permeate the membrane, since the membrane is negatively charged. Negative zeta potential values are associated with a disruption of the cell wall and therefore tend to exhibit a certain degree of toxicity [24].

Table 1. Characteristics of formulations. NA = Not Available

Ref.	NP + drug	Size (nm)	Shape	Zeta potential (mV)
[15]	PLGA + cis platinum	SEM: 123 ± 31 (CPT) 119 ± 37 (DiR) DLS: 206 ± 32 (CPT) 204 ± 41 (DiR)	Spherical with smooth surface	−21.1 (CPT) −23.7 (DiR)
[16]	PLA-PEG + quisionostat	128 ± 8.5	NA	−6.0 ± 1.0
[17]	PEG-PCL + luteolin	34.7	Spherical	−9.2
[18]	PEG-Tf + ZOL	NA	NA	NA
[19]	Albumin-SP + paclitaxel	NA	Spherical	−12
[20]	mPEG-PLGA + PT and TMZ	206.3 ± 14	Smooth surface and uniform morphology	NA
[21]	PLGA-M1 + DOX	156.9 ± 7.1	Spherical	NA
[22]	PEG-Pep1 + PTX	95.7	Spherical	−34.5 ± 1.74

In all, except in ref. [15], drug release was controlled and systematic, ending within a couple of days. When observing the values of the zeta potential presented in the table, it stands out that the more negative the value of the zeta potential, as is the case in ref. [15], the faster the drug release will be. The less negative the value of the zeta potential, the more sustained release over a longer period. The drug loading efficiency (LE) and drug encapsulation efficiency (EE) are presented as a percentage in Table 2, both are related to the amount of drug encapsulated in the NP and the amount of drug released.

Table 2. Results of *in vivo* experiments of the different nanoparticles formulations. NA = Not Available

Ref.	Tolerated doce	Average survival (days) reduced	LE (%)	EE (%)	Drug release	Tumor weight (% reduced)
[15]	20 mg/kg	NP 20: 36.5 NP-10 33.5	9.6 - CPT 0.5 - DiR	NA	Released 80% in 6h and finished in 24 h	NA
[16]	NA	27.5	NA	NA	NA	NA
[17]	NA	NA	5	98.5	Sustained release at 120 h, 46%. Release continued at 70 h	81.2
[18]	NA	42	NA	NA	NA	41
[19]	NA	NA	7.89	85.70	Less than 40% withn 48 h	NA
[20]	PTX 4 mg/L TMZ 20 mg/L	NA	0.871 – PTX 3.15 – TMZ	90.7 – PTX 65.2 – TMZ	Maximum amount accumulated at 80h	NA
[21]	NA	38.5	4.35	NA	40% released in 12 h and 73% released in 24 h	NA
[22]	10 mg/kg	NA	NA	77.27	Rapid release for the first 6 h followed by a sustained release for 10 h	73.4

Nanomedicine systems such as PEG-PCL-Fa-Lut NPs, modified with folic acid to bind with the folate receptor overexpressed in tumor tissue, produce up to 81% tumor inhibition. Something similar occurs with ZOL-Tf-NPs as they exhibit 41% tumor inhibition and were modified on their surface with transferrin to bind with transferrin receptors overexpressed on endothelial cells of the BBB to regulate the passage of iron. The functionalization of the nanoparticles allows them to be more specific, adhering to cancer cells that express specific receptors for the ligands used in the functionalization and to be engulfed by the cell by means of different mechanisms, such as receptor-mediated transcytosis and, therefore, to have an inhibitory effect by accumulating in greater quantities in tumor areas and directly releasing the drug at the expected site.

Our study has some limitations. We did not have access to the full text of a large percentage of articles related to the topic that would potentially help answer the research question, so the type of nanoparticles and drugs presented here is limited and represents a generalization of results of studies on polymeric nanoparticles. Accumulating evidence is lacking to determine a relationship between tumor inhibitory effect and the use of nanoparticles as drug delivery vehicles. In addition, in evaluating the risk of bias we found that the studies report in an unspecific way the randomization process of the control groups, since mentioning that it is randomized and not delving into how it was determined and who was aware of it limits the critical assessment within the sources of evidence.

4 Conclusion

This work addresses for the first time the systematic study of the use of nanoparticles in targeted drug delivery and their relationship with an inhibitory effect on glioblastoma growth. The findings indicate that polymeric nanoparticles are commonly used as a vehicle for drug delivery, specifically those of spherical shape with a smooth surface. According to the data collected, there is a trend towards the use of polymeric nanoparticles in drug encapsulation and their size ranges from 35 to 250 nm. The most common synthesis method is the emulsion-solvent evaporation method for polymeric nanoparticles. In summary, evidence is still lacking to affirm that the use of nanoparticles has an inhibitory effect on tumor size as well as on the proliferative activity of tumor cells in preclinical animal models.

The studies included in this review show a significant difference in survival time and percentage tumor inhibition between groups regardless of the type of nanoparticle used. For the use of nanoparticles in targeted drug delivery to be effective and have an impact on tumor growth reduction, it is necessary to control the size, shape, and surface charge of the nanoparticle, as they have effects on both the health of the test subject and the specific drug release efficacy. An advantage of using nanoparticles in drug delivery is the ability to functionalize them to make them more specific and improve their ability to cross the BBB, which will impact tumor inhibition. These findings provide information for scientists seeking to develop nanomedicine systems for cancer treatment, in particular, glioblastoma multiforme.

References

1. Moreno-Jiménez, S., Alegría-Loyola, M.A., Sonabend, A.M., et al.: Management of glioblastoma: a perspective from Mexico. Chin. Clin. Oncol. **10**, 44 (2021)
2. Tan, A.C., Ashley, D.M., López, G.Y., et al.: Management of glioblastoma: state of the art and future directions. CA Cancer J. Clin. **70**, 299–312 (2020)
3. Fang, C., Wang, K., Stephen, Z.R., et al.: Temozolomide nanoparticles for targeted glioblastoma therapy. ACS Appl. Mater. Interfaces **7**, 6674–6682 (2015)
4. Mailänder, V., Landfester, K.: Interaction of nanoparticles with cells. Biomacromol **10**, 2379–2400 (2009)
5. Ragnaill, M.N., Brown, M., Ye, D., et al.: Internal benchmarking of a human blood-brain barrier cell model for screening of nanoparticle uptake and transcytosis. Eur. J. Pharm. Biopharm. **77**, 360–367 (2011)
6. Pietroiusti, A., Campagnolo, L., Fadeel, B.: Interactions of engineered nanoparticles with organs protected by internal biological barriers. Small **9**, 1557–1572 (2013)
7. Yokel, R.A.: Physicochemical properties of engineered nanomaterials that influence their nervous system distribution and effects. Nanomed. Nanotechnol. Biol. Med. **12**, 2081–2093 (2016)
8. Chen, R., Riviere, J.E.: Biological and environmental surface interactions of nanomaterials: characterization, modeling, and prediction. Wiley Interdiscip. Rev. Nanomed. Nanobiotechnol. **9**, e1440 (2017)
9. Hussain, Z., Thu, H.E., Elsayed, I., et al.: Nano-scaled materials may induce severe neurotoxicity upon chronic exposure to brain tissues: a critical appraisal and recent updates on predisposing factors, underlying mechanism, and future prospects. J. Control. Release **328**, 873–894 (2020)
10. Cui, W., Fu, W., Lin, Y., Zhang, T.: Application of nanomaterials in neurodegenerative diseases. Curr. Stem Cell Res. Ther. **16**, 83–94 (2020)
11. Tsou, Y.H., Zhang, X.Q., Zhu, H., et al.: Drug delivery to the brain across the blood-brain barrier using nanomaterials. Small **13**, 1701921 (2017)
12. Pinel, S., Thomas, N., Boura, C., Barberi-Heyob, M.: Approaches to physical stimulation of metallic nanoparticles for glioblastoma treatment. Adv Drug Deliv. Rev. **138**, 344–357 (2019)
13. Kong, X., Cheng, R., Wang, J., et al.: Nanomedicines inhibiting tumor metastasis and recurrence and their clinical applications. Nano Today **36**, 101004 (2021)
14. Guerra Sánchez, K.J., Chapa, C.: Review Research Protocol. Advances in the administration of nanoparticles for the treatment of glioblastoma. In: Ingeniería Biomédica. Universidad Autónoma de Ciudad Juárez., Ciudad Juárez (2021)
15. Householder, K.T., Diperna, D.M., Chung, E.P., et al.: Intravenous delivery of camptothecin-loaded PLGA nanoparticles for the treatment of intracranial glioma. Int. J. Pharm. **479**, 374–380 (2015)
16. Householder, K.T., DiPerna, D.M., Chung, E.P., et al.: pH driven precipitation of quisinostat onto PLA-PEG nanoparticles enables treatment of intracranial glioblastoma. Colloids Surf. B Biointerfaces **166**, 37–44 (2018)
17. Wu, C., Xu, Q., Chen, X., Liu, J.: Delivery luteolin with folacin-modified nanoparticle for glioma therapy. Int. J. Nanomed. **14**, 7515 (2019)
18. Porru, M., Zappavigna, S., Salzano, G., et al.: Medical treatment of orthotopic glioblastoma with transferrin-conjugated nanoparticles encapsulating zoledronic acid. Oncotarget **5**, 10446–10459 (2014)
19. Ruan, C., Liu, L., Lu, Y., et al.: Substance P-modified human serum albumin nanoparticles loaded with paclitaxel for targeted therapy of glioma. Acta Pharm. Sin. B **8**, 85–96 (2018)

20. Xu, Y., Shen, M., Li, Y., et al.: The synergic antitumor effects of paclitaxel and temozolomide co-loaded in mPEG-PLGA nanoparticles on glioblastoma cells. Oncotarget **7**, 20890–20901 (2016)
21. Pang, L., Zhu, Y., Qin, J., et al.: Primary M1 macrophages as multifunctional carrier combined with PLGA nanoparticle delivering anticancer drug for efficient glioma therapy. Drug Deliv. **25**, 1922 (2018)
22. Wang, B., Lv, L., Wang, Z., et al.: Improved anti-glioblastoma efficacy by IL-13Rα2 mediated copolymer nanoparticles loaded with paclitaxel. Sci. Rep. **5**, 16589 (2015)
23. Shnoudeh, A.J., Hamad, I., Abdo, R.W., et al.: Synthesis, characterization, and applications of metal nanoparticles. In: Biomaterials and Bionanotechnology, pp. 527–612 (2019)
24. Clogston, J.D.: Patri AK Zeta potential measurement. Methods Mol. Biol. **697**, 63–70 (2011)