Cancer Management and Research

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REVIEW

Efficacy of EGFR Targeting IR700-Related Photoimmunotherapy in Preventing Growth of Cancer Cells—A Systematic Review

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Introduction: Research of Near-Infrared Photoimmunotherapy (NIR-PIT) related to human epidermal growth factor receptor (EGFR) in cancer is very popular in recent years. By destroying the cell membrane, NIR-PIT can kill cancer cells and prevent tumor development. By using a statistical method, our study aims to find out the suitable indices for evaluating the efficacy of EGFR-targeted NIR-PIT.

Methods: Exploring strategy: (mice OR vivo study OR animal model) AND (NIR-PIT OR near-infrared photoimmunotherapy OR photoimmunotherapy) AND (EGFR OR EGFR receptor) AND (cancer OR tumor). Including standard: ① Reports could obtain full text from 2014 to 2024. ② Reports using tumor volume as an index of outcome. ③ Reports using IR-700, NIR-PIT, and EGFR antigens. The exclusion criteria were as follows: ① Reports could not obtain the full text. ② Reports that did not use the index above. ③ Reports that did not use IR-700, NIR-PIT, or EGFR antigens. ④ Reports could not obtain concrete data from articles.

Results: Using the above standards, we finally acquired 20 articles that conformed to requests. In this analysis, tumor volume showed statistical meaning of differentiation between experimental and control groups. However, 50% survival did not show the statistical meaning in the analysis.

Discussion: Through analysis, we found that the NIR-PIT groups had obvious differences compared with the control groups in tumor volume. However, we did not find any difference in the survival rates between NIR-PIT and control groups. This may be due to the following reasons: ① the number of samples was insufficient. ② Other indices of survival rate were more suitable in this study than 50% survival rate.

Conclusion: Compared with 50% survival rate, tumor volume may be better for evaluating the efficacy of NIR-PIT.

Keywords: EGFR, NIR-PIT, near-infrared photoimmunotherapy, cancer, tumor volume, survival rate, vivo study

Introduction

Background and Mechanism of NIR-PIT

Research on Near-Infrared Photoimmunotherapy (NIR-PIT) related to the human epidermal growth factor receptor (EGFR) in cancer has been very popular in recent years. As a novel and molecular-targeted therapy, it can selectively kill cancer cells as a consequence of photochemical reactions within antibody-photosensitizer conjugates on cancer cell membranes but does not damage normal tissues around the tumor.[1–3] NIR-PIT has the most obvious characteristic: the targeting specificity of the antibody.[4,5] IR-700 is a photosensitizer that can be activated by near-infrared (NIR) light. In recent years, it was used to treat cancer diseases, such as breast, neck, and head tumors.

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NIR-PIT can do harm to cancer cell by two functions. One is the direct function that it can destroy the tumor cell membrane by a photo-induced ligand release reaction.[6] Another one is activated host's immune system,[7] which can lead to immunogenic cell death. Upon irradiation with NIR light, as the direct function, IR700 is activated and produces a photo-induced ligand release reaction with minimal or no side effects on adjacent normal cells.[6] Cellular necrosis of tumoral tissue occurs, and tumoral antigens are released, which stimulate the immune system of the human body and induce tumoral immunity[7,8] (Figure 1). In this process, NIR-PIT releases DAMPs hallmarks such as calreticulin (CRT), high-mobility group box 1 (HMGB1), adenosine triphosphate (ATP), heat shock protein (Hsp) 70, and Hsp 90, which can improve the maturation of DCs and stimulate naive $CD8^+$ T cells.[9]

EGFR-Targeted NIR-PIT

As a transmembrane receptor that belongs to the ErbB family of receptor tyrosine kinases, EGFR is overexpressed in various types of cancers such as colon, breast, and rectal cancers.[10,11] One characteristic of solid tumors initiated by surface receptors is uncontrolled signal transduction, and EGFR plays a significant role in this process.[12]

However, EGFR still has limitations in treatment because of its complicated construction and function. It's signaling network involves 211-biochemical-reactions and 322 signaling molecules.[13] EGFR controls the activation of several pathways such as PI3K/Akt/mTOR and RAS-RAF-MEK-ERK.[14] These accessions modify the cell proliferation,

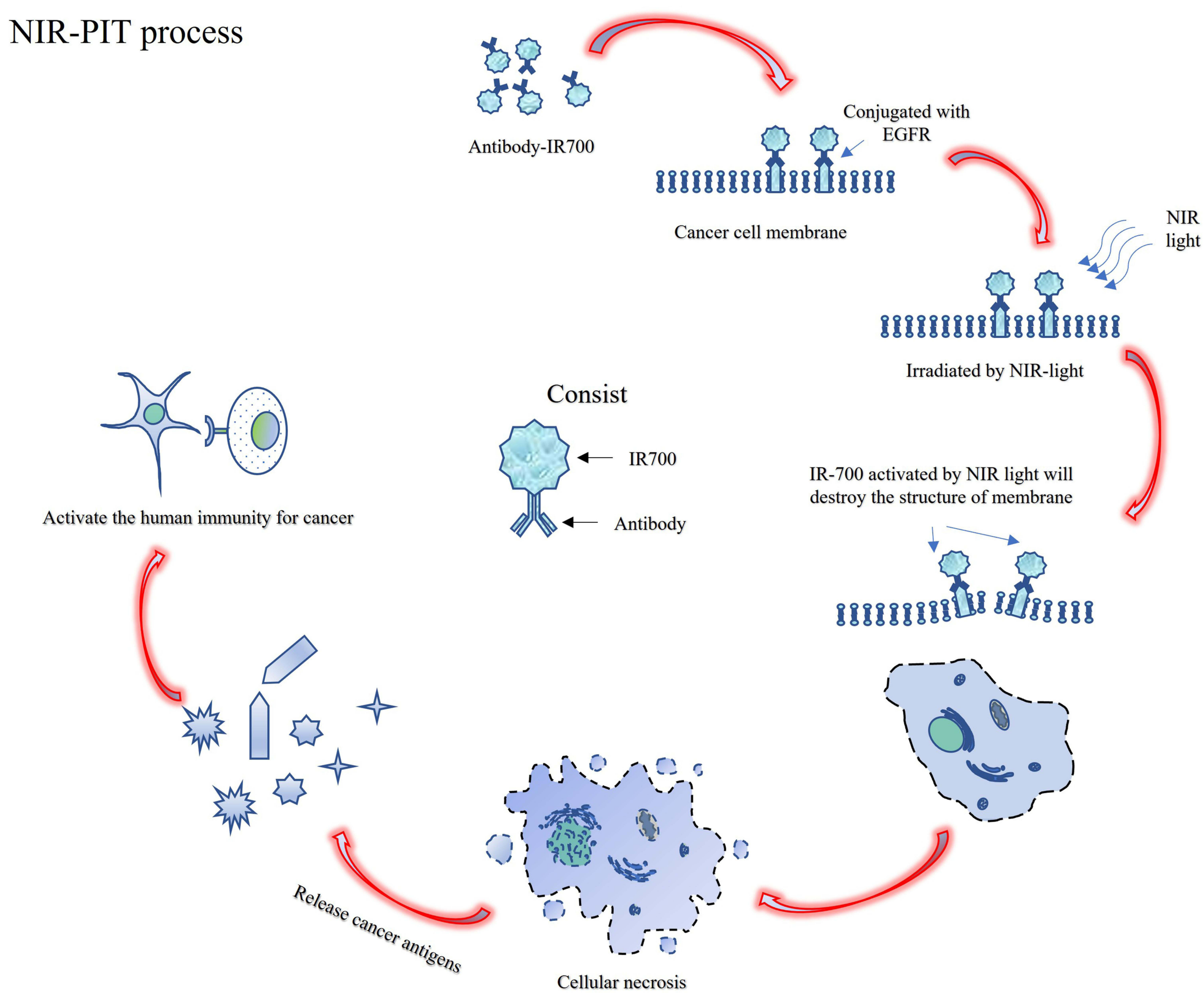

Figure 1 Mechanism of NIR-PIT in killing cancer cell.

survival, and migration. Although new therapies for molecule-targeted drugs, such as ErbB tyrosine kinase receptor family ErbB1/EGFR or ErbB2/human epidermal growth factor 2 (HER2), have been used in some clinical trials, the outcomes of these experiments have been unsatisfactory.[15] Thus, it is necessary to better understand the pathogenesis of EC to develop novel targeted therapies.

It is worth noting that using EGFR-targeted NIR-PIT which combines Cet-IR700 conjugate with NIR laser system to treat patients with head and neck cancer has been selectively sanctioned in Japan in 2020.[16]

Tamagawa et al[17] reported a case in which a patient with locoregionally recurrent oropharyngeal squamous cell carcinoma at the tongue base was cured using NIR-PIT after chemoradiotherapy. Ten months after therapy, patient had no recurrence or metastasis.

Okada et al[18] described a case of mixed reality-supported near-infrared photoimmunotherapy in oropharyngeal cancer in their article. They used the mixed reality (HMD-MR) technology of head-mounted displays to perform NIR-PIT on patients and demonstrated the utility of HMD-MR technology in optimizing NIR-PIT outcomes. Three months post-treatment, no residual lesion was observed.

Recently, to minimize adverse events and maximize therapeutic efficacy, some scientists have found the best dose of NIR light in the study of hEGFR-targeted mouse experiments.[19] They found that in mouse models, intratumoral fluorescence was still visible at 25 J/cm^2 irradiation but completely disappeared when the dose was increased to 50 J/cm^2 or greater.

According to these outcomes, using EGFR-targeted NIR-PIT to treat the colorectal cancer also has the possibility.

Recent Studies in Other Cancer

Although NIR-PIT remains in the period of basic experiments in other types of cancer, some studies have obtained significant results and improvements.

Remarkable tumor suppression and a prolonged survival rate have been observed in athymic nude mice suffering from EGFR-positive human tumors (such as triple-negative breast cancer (TNBCs)) following treatment with cetuximab-IR700 or panitumumab-IR700 and in combination with NIR-light.[20,21]

Suzuki et al[22] performed in vitro and in vivo experiments on osteosarcoma and found that NIR-PIT using Pan-IR700 had strong efficacy in curing osteosarcoma.

Hanaoka et al[23] used two normal cell lines of humanity in an NIR-PIT experiment and found that NIR-PIT could induce the death of senescent cells, which were induced by conventional anti-cancer treatments such as chemotherapy and radiotherapy.

Yamaguchi et al[24] used the EGFR Affibody-IR700Dye conjugate and found that NIR-PIT could selectively induce the death of EGFR-positive SGC cells and limit tumor tissue growth and metastasis.

Interestingly, some scientists have attempted to use hEGFR- and CD25-targeted combined NIR-PIT to treat cancer.[25] The combined PIT group showed the greatest inhibition of tumor growth.

In addition to EGFR, other receptors such as HER2, CD25 can also be targeted in NIR-PIT experiments.

Yamaguchi et al[26] used a combination of NIR-PIT using the HER2 Affibody-IR700Dye conjugate and trastuzumab-IR700Dye conjugate to enhance the cytotoxic effect in HER2-positive breast cancer cells. They reported that their approach had potential to improve efficiency of the current NIR-PIT, especially for heterogeneous HER2-positive cancers.

Inagaki et al[27] evaluated the efficacy of PD-L1 targeted NIR-PIT (αPD-L1-PIT) in immunocompetent tumor mouse models. The results showed that αPD-L1-PIT was effective in a PD-L1 high-expression tumor model. Furthermore, αPD-L1-PIT induced an abscopal effect on distant tumors and long-term immunological memory.

In these studies, indices such as tumor volume, 50% survival rate, body weight and tumor size are usually used to evaluate the efficacy of NIR-PIT. However, we lack statistical analysis to directly observe its efficacy. By using statistical method, our study aims to conclude the EGFR-targeted studies in recent years and find out whether tumor volume and 50% survival rate are suitable for evaluating the efficacy of EGFR-targeted NIR-PIT or not. In order to observe the function directly destroying the tumor cell membrane, we decided to adopt studies of athymic nude mice so that we could exclude the impact of host's immune system. We hope that this analysis can provide reference of suitable indices for future NIR-PIT studies.

Method

This study was registered in PROSPERO(CRD42024613626). All related data and contents were known by all authors and agreed to be published.

Searching Strategy and Data Collection

Exploring strategy: (mice OR vivo study OR animal model) AND (NIR-PIT OR near-infrared photoimmunotherapy OR photoimmunotherapy) AND (EGFR OR EGFR receptor) AND (cancer OR tumor).

Inclusion criteria: ① Reports could obtain full text from 2014 to 2024. ② Reports using tumor volume as an index of outcome. ③ Reports using IR-700, NIR-PIT, and EGFR antigens.

The exclusion criteria were as follows: ① Reports could not obtain the full text. ② Reports that did not use the index above. ③ Reports that did not use IR-700, NIR-PIT, or EGFR antigens. ④ Reports could not obtain concrete data from articles.

Using the standards above, we acquired 20 articles that conformed to requests (Figure 2).

Index

Tumor volume[10,19,21,22,24,25,28–41] was adopted as the main index in this study (Table 1). In addition, other indices, such as the survival rate,[19,21,25,30,34–40] were collected if the articles had. After collecting the data of these indexes, we used SPSS 26.0 to analyze their characteristics. The time points were selected from the first day after using NIR-PIT to the last day recorded.

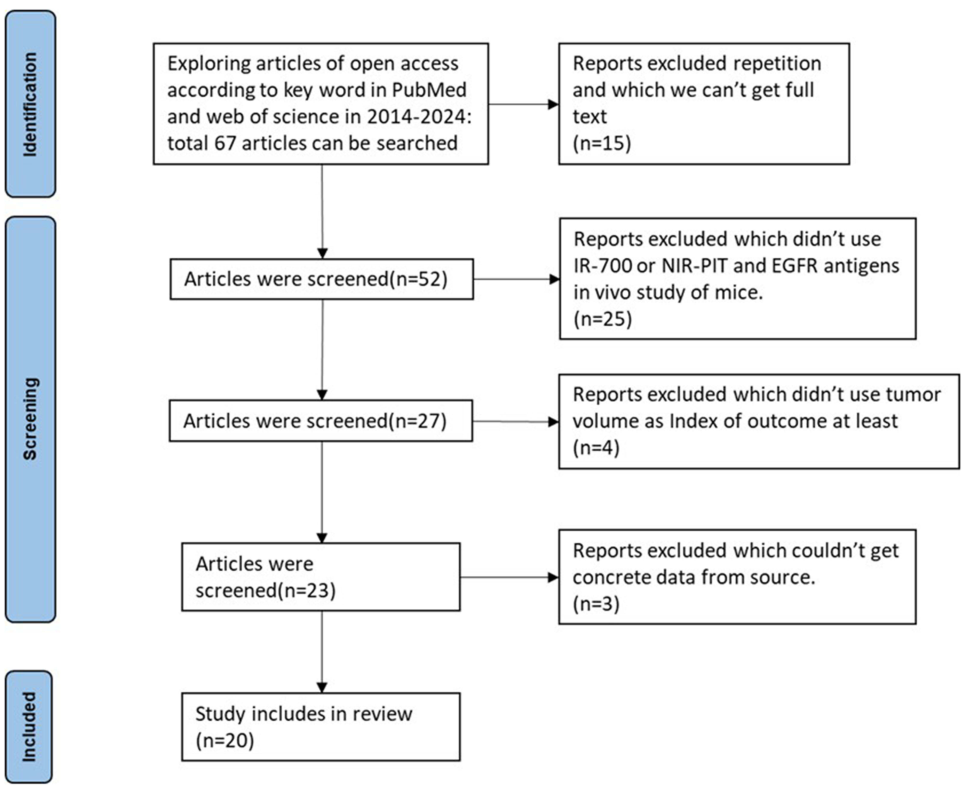

Figure 2 Flow chart of finding and selecting articles.
Note: The flow chart was completed according to PRISMA formula.

Table 1 Included Studies

Article Name	DOI	Year	APC	Mouse Model	Inoculated Situation	Index	Data Source
Burley, et al[28] 2018	10.1002/ijc.31246	2018	$Z_{EGFR:03115}$-IR700	Female NCr athymic mice (6 weeks)	Top right shoulder; Subcutaneous model	Tumor volume	Figure in article
Furumoto, et al[19] 2022	10.3390/cancers14164042	2022	Pan-IR700; Cet-IR700	Six- to eight-week-old female homozygote athymic nude mice	Right side of dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Hirata, et al[29] 2021	10.1111/cas.14780	2021	Pan-IR700	BALB/c Slc-nu/nu nude mice (6-week-old females)	Right side of dorsum; Subcutaneous model	Tumor volume Body weight	Figure in article
Kishimoto, et al[30] 2018	10.1016/j.freeradbiomed.2017.12.034	2018	Pan-IR700	Female athymic nude mice	Right hind legs; Subcutaneous model	Tumor volume Survival rate	Figure in article
Maczynska, et al[31] 2022	10.1186/s12916-021-02213-z	2022	$Z_{EGFR:03115}$-IR700	NCr athymic female mice (5–6 weeks)	The right shoulder; Subcutaneous model	Tumor volume	Figure in article
Nakajima, et al[32] 2018	10.18632/oncotarget.25068	2018	Pan-IR700	Balb/c Slc-nu/nu nude mice (6-weeks-old, females)	Right and left sides of dorsum; Subcutaneous model	Tumor volume Body weight	Figure in article
Nakamura, et al[33] 2017	10.1158/1535-7163.MCT-16-0663	2017	Pan-IR700	-	-	Tumor volume Survival rate	Figure in article
Sano, et al[34] 2014	10.1158/1535-7163.MCT-13-0633	2014	Pan-IR700	Six-eight-week-old female homozygote athymic nude mice	Right and left dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Suzuki, et al[22] 2024	10.1016/j.tranon.2024.102132	2024	Pan-IR700	6-week-old female mice (BALB/c-nu)	Left hind leg; Subcutaneous model	Tumor volume Body weight	Figure in article
Takao, et al[10] 2023	10.1111/cas.15965	2023	Cet-IR700	Female homozygote athymic nude mice, 6–8 weeks old	Right dorsum of mice; Subcutaneous model	Tumor volume	Figure in article
Yamaguchi, et al[24] 2024	10.3390/ijms25063233	2024	EGFR Affibody-IR700	Four-week-old female athymic mice (BALB/cSlc-nu/nu)	Dorsum; Subcutaneous model	Tumor volume	Figure in article
Maruoka (1), et al[35] 2017	10.1021/acs.molpharmaceut.7b00731	2017	Pan-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Maruoka (2), et al[36] 2018	10.1021/acs.molpharmaceut.8b00002	2018	Pan-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Nagaya (1), et al[21] 2015	10.1371/journal.pone.0136829	2015	Cet-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Right dorsumSubcutaneous model	Tumor volume Survival rate	Figure in article
Nagaya (2), et al[37] 2018	10.18632/oncotarget.24876	2018	Can225-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Right dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Nagaya (3), et al[38] 2018	10.1158/1535-7163.MCT-17-0851	2018	Pan-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Right dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Okada (1), et al[25] 2021	10.1016/j.ebiom.2021.103345	2021	Pan-IR700	Female C57BL/6 mice (6–8 weeks old)	Right side of dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Okada (2), et al[39] 2022	10.1007/s00262-021-03124-x	2022	Pan-IR700; Cet-IR700	Female C57BL/6 mice (6–8 weeks old)	Right side of dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Sato (1), et al[40] 2014	10.1016/j.molonc.2014.01.006	2014	Pan-IR700; Cet-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Right dorsum; Subcutaneous model	Tumor volume Survival rate	Figure in article
Sato (2), et al[41] 2016	10.18632/oncotarget.7365	2016	Pan-IR700	Female homozygote athymic nude mice (6–8 weeks old)	Right dorsum; Subcutaneous model	Tumor volume	Figure in article

Notes: Information of included studies.

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Tools

MATLAB and GRABIT were used to collect the data. By inputting the figure of articles, we obtained concrete data for the indices above. Data were collected using the following formula:

$$\text{Difference of Tumor volume per day} = (\text{tumor volume}_{\text{day2}} - \text{tumor volume}_{\text{day1}})/\text{days}$$

A total of 50 groups of data were included in the analysis. Twenty-five groups of them were assigned to the experimental groups. The other half of the patients were allocated to the control groups.

Quality Assessment

All articles were assessed using the SYRCLE tool,[42] which was revised from the Cochrane risk-bias tool. This process was completed by three researchers who participated in our study, and both conformed to the evaluation rules. According to the SYRCLE tool, all articles had at least 75% of the items at a low risk of bias. All included articles reported that all the mice in the experiments were randomly allocated into groups. In outcome of the experiments, all studies showed that experimental groups and control groups in their own research were similar at baseline. In the random allocation of housing, all articles stated that all steps proceeded according to related guidelines and conformed to experimental requests. Figure 3A and B show the evaluation of all articles regarding low, high, and unclear risks of bias.

Results

Through a series of procedures, we acquired the related outcomes of tumor volume and survival rate and completed the analysis.

Outcome of Tumor Volume

In the analysis of tumor volume, we adopted the data from the NIR-PIT groups (using NIR-PIT) and control groups (without APCs and NIR light). Moreover, to decrease the influence of the immunity and subcutaneous part of the tumor, we chose data from articles that used immunodeficient mice and subcutaneous models.

We compared the difference between the control and NIR-PIT groups and found that the value of tumor volume per day in the NIR-PIT group was lower than that in the control group (Table 2 and Figure 4A). Next, we evaluated the normality of the two groups through SPSS 26.0. All the results conformed to a skewed distribution (Figure 4B). We used the Mann–Whitney *U*-test to analyze the data and found that the index of tumor volume per day in the NIR-PIT group

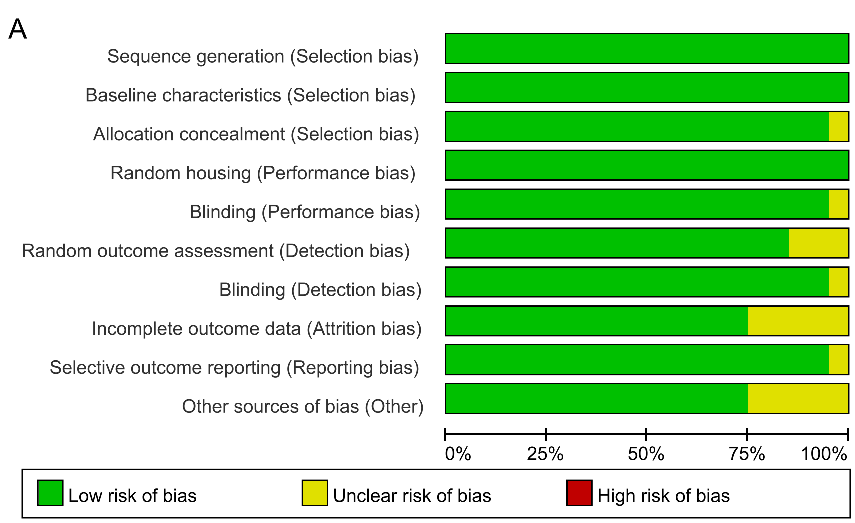


Figure 3 Continued.

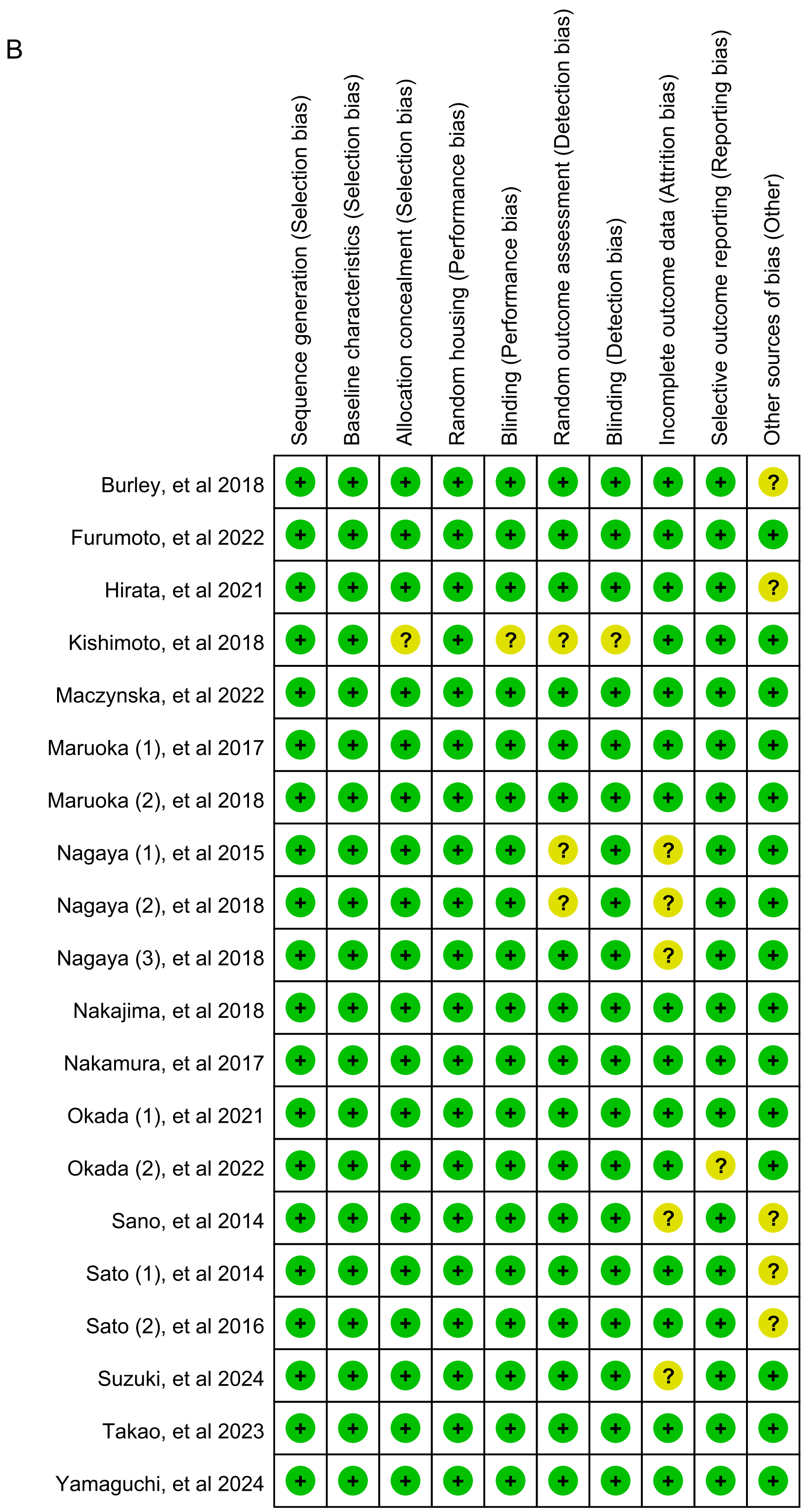


Figure 3 The results of quality assessment for included studies. (**A**) Total situation about risk of bias. (**B**) Concrete situation about risk of bias.

Table 2 Data of Tumor Volume

Article	Difference of Tumor Volume per Day (mm^3/d)		D-Value
	Control	NIR-PIT	
Burley, et al[28]	44.15	12.33	31.82
Furumoto, et al[19]	80.47	25.30	55.71
	30.66	10.91	19.75
Hirata, et al[29]	26.86	0.11	26.75
Kishimoto, et al[30]	119.60	18.92	100.68
Maczynska, et al[31]	95.12	24.43	70.69
Nakajima, et al[32]	55.79	9.00	46.79
Sano, et al[34]	70.68	13.25	57.43
Suzuki, et al[22]	50.65	25.05	25.60
Takao, et al[10]	17.90	5.15	12.75
	54.43	6.95	47.48
Yamaguchi, et al[24]	33.51	9.51	24.00
Maruoka(1),et al[35]	120.14	44.05	76.09
Maruoka(2),et al[36]	130.36	47.84	82.52
Nagaya (1), et al[21]	75.63	38.52	37.11
	30.78	13.94	16.84
	35.66	16.43	19.23
	34.11	9.96	24.15
Nagaya (2), et al[37]	41.93	17.96	23.97
Nagaya (3), et al[38]	24.00	6.50	17.50
Okada (1), et al[25]	32.71	10.13	22.58
Okada (2), et al[39]	24.57	14.04	10.53
Sato (1), et al[40]	285.52	15.02	270.5
Sato (2), et al[41]	165.86	50.20	115.66

Notes: D-value=Tumor volume per day (NIR-PIT) – Tumor volume per day (control).

had apparent differentiation from that of the control group (Figure 4C–E). Therefore, we have at least a 95% accuracy to realize that the outcome of tumor volume is statistically significant. In this analysis, we realized that NIR-PIT could prevent tumor growth.

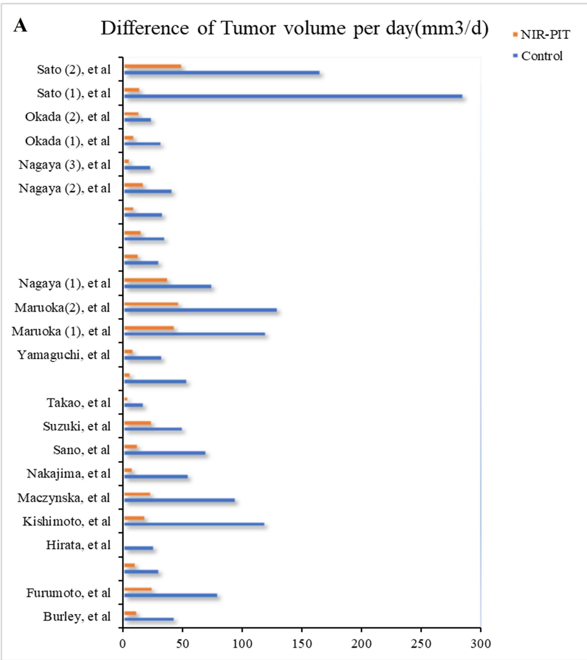
Outcome of Survival Rate

In addition to the tumor volume, we also collected data on the 50% Survival Rate (Table 3). The 50% survival rate in the NIR-PIT group was higher than that of the control group. We performed a normality test and found that the data conformed to a skewed distribution (Figure 5A). The Mann–Whitney *U*-test showed that there was no difference in the survival rate between the two groups (Figure 5B and C). Therefore, we could not realize that outcome of 50% survival rate had statistical meanings. In this analysis, we could not conclude that NIR-PIT could enhance the days of 50% survival rate of the experimental mice.

Discussion

Some studies on NIR-PIT in animal experiments on cancer were conducted and achieved advanced outcomes.

NIR-PIT can not only be used to kill tumor cells directly but also function in the TME. Maruoka et al[43] found thatNIR-PIT targeting the combination of TME and tumor cells showed significant tumor growth inhibition and prolonged survival. Akai et al[44] scientists found that FAP-targeted NIR-PIT induced specific cell death in cancer-associated fibroblasts (CAFs) without damaging the adjacent normal cells. FAP-targeted NIR-PIT in mice showed apparent tumor regression in the CAF high-expressing tumor model accompanied by an increase in CD8+ tumor-infiltrating lymphocytes (TILs).



B

		Kolmogorov-Smirnov[a]			Shapiro-Wilk		
	method	Statistic	df	Sig.	Statistic	df	Sig.
tumor volume	control	.218	24	.005	.750	24	.000
	NIR-PIT	.198	24	.016	.856	24	.003

a. Lilliefors Significance Correction

C

	Null Hypothesis	Test	Sig.	Decision
1	The distribution of tumor volume is the same across categories of method.	Independent-Samples Mann-Whitney U Test	.000	Reject the null hypothesis.

Asymptotic significances are displayed. The significance level is .050.

D

Total N	48
Mann-Whitney U	55.000
Wilcoxon W	355.000
Test Statistic	55.000
Standard Error	48.497
Standardized Test Statistic	-4.804
Asymptotic Sig.(2-sided test)	.000

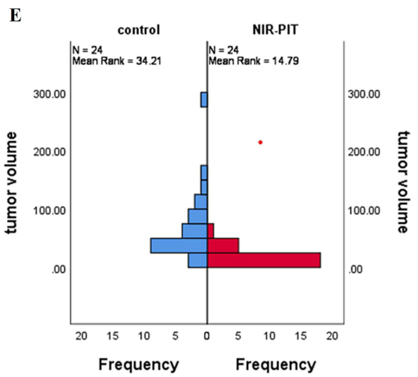


Figure 4 (**A**) Difference of Tumor volume per day (mm^3/d) between NIR-PIT groups and control groups in each study. (**B**) Normality tests of tumor volume between the groups. (P<0.05). (**C**) Hypothesis Test Summary of tumor volume (P<0.05). (**D**) Summary of independent-Samples Mann–Whitney *U*-Test. (**E**) Frequency distribution of tumor volume in Independent-Samples Mann–Whitney *U*-Test.

Table 3 Data of 50% Survival Rate

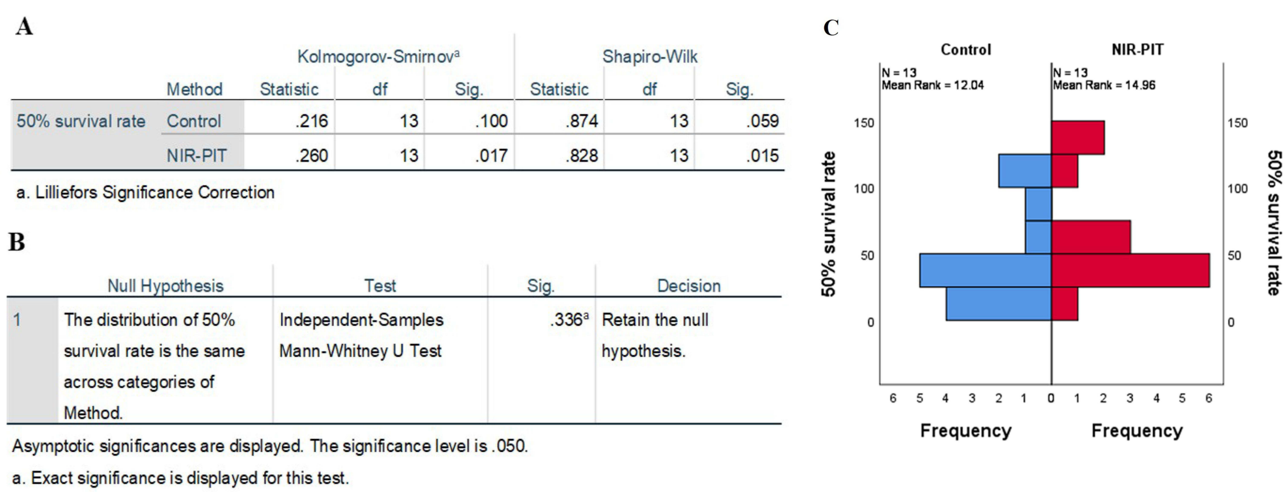
Article	Days of 50% Survival Rate (d)	
	Control	NIR-PIT
Furumoto, et al[19]	31	46
	46	53
Kishimoto, et al[30]	22	30
Sano, et al[34]	12	14
Maruoka (1), et al[35]	23	30
Maruoka (2), et al[36]	23	32
Nagaya (1), et al[21]	60	63
	104	146
	101	142
Nagaya (2), et al[37]	46	60
Nagaya (3), et al[38]	82	105
Okada (1), et al[25]	34	44
Okada (2), et al[39]	37	41

Notes: Data of days of 50% Survival Rate (d).

NIR-PIT also has treatment efficacy in spontaneously occurring transgenic models (Table 1). Nakamura et al[33] found that the growth of spontaneously occurring tumors could also be prevented by NIR-PIT. These results could be readily translated to human conditions compared to the outcomes of the subcutaneous model. However, because this type of study is still rare, we cannot collect sufficient data to perform the analysis at present.

According to the outcome of tumor volume, the results had statistical meaning, which meant that NIR-PIT groups showed statistically significant differences compared to the control groups. This result conformed to our prediction before starting the investigation, which showed that NIR-PIT could prevent tumor tissue growth.

We also analyzed the survival rates in this study. However, in the analysis of survival rate, the outcome of 50% survival rate did not have the statistical meaning, and we could not realize that it had statistically significant difference between the NIR-PIT and control groups. This outcome differed from that of the tumor volume. We originally analyzed this position and thought that this might be due to the following reasons: ① the number of samples was insufficient. ② Other indices of survival rate, such as 75% and 100% survival rates, might be more suitable in this study than the 50% survival rate. An off-target toxicity is also possible for this outcome. Drug itself may also be toxicity to the cancer cell.


A

	Method	Kolmogorov-Smirnov[a] Statistic	df	Sig.	Shapiro-Wilk Statistic	df	Sig.
50% survival rate	Control	.216	13	.100	.874	13	.059
	NIR-PIT	.260	13	.017	.828	13	.015

a. Lilliefors Significance Correction

B

	Null Hypothesis	Test	Sig.	Decision
1	The distribution of 50% survival rate is the same across categories of Method.	Independent-Samples Mann-Whitney U Test	.336[a]	Retain the null hypothesis.

Asymptotic significances are displayed. The significance level is .050.

a. Exact significance is displayed for this test.

Figure 5 (**A**) Normality tests of 50% survival rate between NIR-PIT and control groups (P<0.05). (**B**) Hypothesis Test Summary of 50% survival rate (P>0.05). (**C**) Frequency distribution of 50% survival rate in Independent-Samples Mann–Whitney *U*-Test.

However, because all outcomes of survival rate did not show the difference between the drug-only group and control group in the studies of Table 1, we did not consider and analyze this reason thoroughly.

This study had some limitations. First, we investigated the two indices of tumor volume and survival rate in the NIR-PIT experiment. Other indices, such as body weight, tumor weight, and fluorescence intensity, might also be related to NIR-PIT. If we found their relationships, we could more fully analyze the differences between the NIR-PIT and control groups. Second, this analysis only analyzed the position of NIR-PIT in various cancers, because the number of studies on one tumor was insufficient. If we could collect adequate data for cancer diseases, such as colon cancer and rectal cancer, it might be better to certify the function of NIR-PIT, which was effective in killing tumor cells.

In addition to the tumor volume and survival rate, other indices were also used in NIR-PIT experiments.

In a study of EGFR-related bone metastases,[45] Luciferase Activity was used to differentiate efficacy between NIR-PIT groups and the other two groups. The value in the NIR-PIT group was significantly lower than that in the other groups.

Body weight is also a general index used in the NIR-PIT study because it directly reflects the weight of mice directly.[29,32] Based on body weight, we could initially determine the condition of mice bearing the tumor, such as whether the tumor plundered the nutrition of the host or not. However, because there are too many factors that can affect body weight, whether body weight can be a good index in the NIR-PIT study still needs to be confirmed.

Fluorescence intensity[39] and tumor weight[46] are also good indices for NIR-PIT studies. Although the fluorescence intensity sometimes cannot reflect the change in NIR-PIT before and after, it can show the accumulation of antibody-IR700 in tumor tissue, which indicates that the drugs can target the receptor. Tumor weight is an index that can show the differentiation of tumors directly between the NIR-PIT and control groups.

Therefore, based on the outcome of this analysis, we originally realized that tumor volume may be a suitable index for NIR-PIT experiments. As for 50% survival rate, we need to determine the real reasons for its outcome and collect more samples to identify whether it is suitable Other indices such as Luciferase Activity and Fluorescence intensity maybe are the better compared with tumor volume and survival rate because they can directly reflect the situations that NIR-PIT kills the cancer cell directly and photosensitizers combine with receptors on tumor cell membrane. Moreover, host immune system also plays an important role in NIR-PIT therapy. It is necessary to evaluate indices including tumor volume and survival rate in immunocompetent animal models. If the results of them are the same as Subcutaneous models we used in this analysis, they may be used as vital indices in clinical evaluation. If we can fully use these indices in suitable studies, we believe that they will offer an important conference not only for NIR-PIT research but also for clinical treatment in the future.

Conclusion

In this study, the outcome of tumor volume has statistical meaning. As for the 50% survival rate, we cannot realize that the result of it has statistical meaning. Compared with 50% survival rate, tumor volume may be the better index for evaluating the efficacy of NIR-PIT.

Data Sharing Statement

All dataset could be acquired in the article.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

All authors declare no competing interests in this work.

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