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Future Treatment of Cancer: Effectiveness of Integra, Herbal Extracts, And Chitosan Nanobiopolymer Integration in Cancer Therapy (a Narrative Review)

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Abstract

Cancer is a global issue affecting all ages and genders. Chemotherapy, while common, has significant physical and psychological side effects, prompting exploration of traditional herbal treatments. However, herbal products often suffer from poor solubility and bioavailability. To address this, chitosan nanobiopolymers are being investigated for their potential to enhance the effectiveness of herbal extracts. This review aims to assess the integration of chitosan nanobiopolymers with herbal extracts as anticancer agents and their manufacturing process, utilizing databases like PubMed, Google Scholar, DOAJ and NIH with keywords such as herbal medicine, cancer, nanobiopolymers, and chitosan. Studies show chitosan is an effective carrier for active substances in cancer therapy, offering improved solubility, stability, and bioavailability. Nanobiopolymers, ranging from 0-1000 nm, serve as carriers for active compounds in cancer treatment. This literature review is expected to contribute to the understanding of the potential integration of herbal extracts with chitosan, particularly in the production of chitosan nanobiopolymers as carriers in cancer treatment.

Keywords: Chitosan; cancer; herbal extract; nanobiopolymer

1. Introduction

Cancer is currently one of the most widespread diseases globally, with fatal impacts, and it can affect individuals of various age groups and genders, both females and males [1]. The Global Cancer Observatory 2022 offers a summary of the distribution of cases for the major cancer types. Breast cancer and lung cancer rank highest, with 2,296,840 and 2,480,675 instances, respectively, accounting for approximately 11.5% of the overall 12.4% of cancer cases. Colorectal cancer exhibits a significant prevalence of 1,926,425 cases (9.6%), followed by prostate cancer at 1,467,854 cases (7.3%), stomach cancer at 968,784 cases (4.8%), and liver cancer at 866,136 cases (4.3%). Other cancer types combined represent 49.9% of total cases, totaling 9,969,785 cases [2]. Chemotherapy is one of the most commonly used methods to kill cancer cells in the body. This process involves administering drugs specifically designed to stop the growth of or even kill these cancer cells. While chemotherapy can effectively combat cancer, it's crucial to acknowledge that it also carries several risks that require careful monitoring. Some of the risks and side effects of chemotherapy include digestive system disturbances from the oral cavity to the large intestine, alopecia, nausea, and spinal cord depression [3]. Chemotherapy also induces toxic effects on normal cells, leading to DNA damage [4], and it causes psychological effects such as anxiety in cancer patients [5]. This encourages the community to explore alternative treatments, including the use of traditional herbal remedies [6]. The Indonesian community turns to traditional medicine influenced by ancestral practices, considering it safer and believed to contribute to overall health [7]. Traditional herbal products have been tested and shown to contain a substantial amount of anticancer agents such as alkaloids, coumarins, polyphenols, flavonoids, quinone, terpenoids, and artesunate [8]. However, traditional herbal products used in treatment generally have limitations in terms of water solubility, which can significantly impact their low bioavailability [9]. The large molecular size of natural extract compounds can lead to low drug absorption, besides solubility issues, serving as a primary reason why some extracts are not utilized in clinical treatments [10]. Nanotechnology emerges as a solution to address these challenges by offering better solubility, stability, and bioavailability for active compounds or drugs used in cancer therapy. Nanoparticles with tiny sizes can enhance drug or phytochemical compound delivery to targets more efficiently [11], [12]. The use of natural ingredient extracts into drug delivery systems presents significant promise owing to their multi-active components, which may yield synergistic therapeutic effects. Nonetheless, issues pertaining to chemical stability, solubility, and bioavailability require resolution [13]. Enhanced drug delivery systems, such as chitosan-based nanoparticles, facilitate the encapsulation of active chemicals within the particles and their attachment to the polymer's surface.

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This dual feature facilitates enhanced regulation of release rates and improves targeting to specific cells or tissues, thereby minimizing systemic side effects and perhaps augmenting efficacy [14]. The technique of this drug delivery system entails encapsulating or affixing active chemicals to nanoparticles designed to release the medicine under particular physiological conditions or at designated places [15]. The assessment of these systems often includes the evaluation of particle size, polydispersity index, zeta potential, encapsulation efficiency, and release kinetics. Furthermore, the system's therapeutic efficacy and potential can be evaluated using in vitro studies utilizing cancer cells or in vivo experiments in cancer-induced animal models [16].

A promising approach to the development of cancer therapy involves integrating nanotechnology with plant extracts. Nanotechnology has the potential to create efficient drug delivery systems and can be a solution to the limitations of herbal treatments [17]. On the other hand, plant extracts contain bioactive compounds with potential anticancer properties [18]. The delivery systems of herbal products can be optimized by using biopolymers as drug delivery agents [19]. Biopolymers are a type of biomolecule polymer formed by combining monomer units with covalent bonds, resulting in larger molecules. As biodegradable materials, biopolymers are produced by living organisms [20].

2. Result and Discussion

Herbal extract characteristics

Prior to examining the application of nanotechnology, it is crucial to comprehend the fundamental attributes of natural extracts utilized, particularly regarding stability, physicochemical features, and potential for alteration. The stability of natural extracts, particularly from medicinal plants, is essential for preserving their therapeutic value. This stability may be influenced by external factors such as oxygen exposure and temperature, which can expedite the decomposition of active chemicals and diminish biological efficacy [21]. Active chemicals in herbal extracts are generally susceptible to degradation under adverse environmental circumstances, hence requiring suitable storage or stabilization methods, such as freeze-drying, to preserve extract quality [22].

Furthermore, the physical and chemical characteristics of natural extracts affect their efficacy as medicinal agents. Numerous extracts exhibit low water solubility, which can impede absorption in the body and diminish their bioavailability. Herbal extracts comprise many bioactive chemicals that may interact, influencing their stability and biological activity [9]. The particle size of natural extracts influences cellular penetration and pharmacological efficacy, highlighting the necessity for additional modification [23]. Transforming natural extracts into nanoparticle form provides a viable option to overcome these limitations and augment their therapeutic efficacy. An essential method involves encapsulating extracts into nanoparticles, thereby enhancing the solubility, stability, and bioavailability of the active chemicals [24]. Nanotechnology enables the formulation of natural extracts for controlled release and targeted distribution to certain tissues, such as malignant tumors, therefore reducing adverse effects on healthy tissues [25]. Biocompatible polymers, such as chitosan, are frequently employed as nanoparticle matrices owing to their ability to be modified and their capacity to encapsulate or load active chemicals on their surfaces [14]. This method enhances the efficacy of nanoparticle-based natural extracts as targeted drug delivery systems, augmenting their potential for medicinal applications, particularly in anticancer therapy [10].

Anticancer coumpound in herbal extract

A multitude of chemical compounds derived from natural extracts have been acknowledged for their significant anticancer effects. Curcumin, extracted from turmeric (*Curcuma longa*), exhibits anticancer properties by suppressing cancer cell proliferation, promoting apoptosis, and diminishing angiogenesis [26]. Furthermore, resveratrol, present in grapes and many fruits, is recognized for its anticancer properties by influencing cellular biochemical pathways, particularly through the suppression of transcription factors that facilitate cancer cell proliferation [27]. Quercetin, a flavonoid found in numerous vegetables and fruits, exhibits cytotoxic effects against diverse cancer cell types by inducing apoptosis and inhibiting cancer cell proliferation through modulation of the cell cycle [28] . Alkaloids like vincristine, derived from the plant Catharanthus roseus, are frequently employed in cancer treatment by inhibiting cell division during the mitotic phase [29].

The flavonoid group, comprising quercetin and kaempferol, is acknowledged for its several anticancer properties, including the inhibition of inflammatory enzyme activity and the neutralization of free radicals, which diminishes oxidative stress that may facilitate cancer progression [30]. Terpenoids, such as limonene derived from citrus peels, demonstrate anticancer potential by inducing apoptosis [31]. Furthermore, flavonoid molecules such as hispidulin, present in various plants, exhibit cytotoxic effects on cancer cells by impeding their development and inducing apoptosis [32], Genistein, a flavonoid found in soy, influences cellular signaling pathways and functions as an antioxidant, offering protection against oxidative damage [33]. Comprehending the potential of these molecules facilitates the synthesis of efficient nanobiopolymer systems utilizing chitosan as a carrier, anticipated to improve bioavailability and efficacy in cancer treatment while minimizing adverse effects [34].

The use of biopolymers such as chitosan in the development of nano-carriers has emerged as a significant advancement in cancer treatment, specifically for safeguarding active chemicals and improving uptake by target cells [35]. Chitosan, a natural polymer, provides benefits of biocompatibility, biodegradability, and a positive charge that enhances interactions with cancer cells [36]. Chitosan in nano-carriers can encapsulate or coat active compounds via ionic or hydrophobic interactions, thereby preserving the stability of the compounds until they arrive to their targets [37]. Chitosan-based nanocarriers facilitate the regulation of particle dimensions and release kinetics, hence improving bioavailability and extending the half-life of active substances within the organism [14]. The assessment of these delivery methods is conducted via in vitro experiments on cancer cells to evaluate delivery efficiency and cytotoxicity, alongside in vivo tests on animal models to determine distribution

and therapeutic efficacy [38]. Consequently, chitosan not only improves the stability and transport of active chemicals but also facilitates regulated release aimed at cancer cells specifically, thereby minimizing systemic side effects and enhancing treatment efficiency [39].

Nanopolymers and herbal extracts

With the advancement of technology, the use of nanotechnology in cancer treatment has become a significant research focus. The application of nanotechnology to plant extracts can provide two benefits: treatment effectiveness and a reduction in side effects [10]. Nanopolymers are one type of nano-technology that can be integrated with herbal extracts. Nanopolymers are particles with a size range of 1 to 1000 nm that can contain active compounds trapped within the polymer core or attached to the polymer core's surface[40]. Polymers used as excipients in nanoparticle production aim to improve drug solubility, control particle size, provide targeted delivery, and reduce drug or active substance toxicity [41]. Nanopolymers have more controlled and effective abilities to reach specific tissues or organs in the body. Their tiny size allows better penetration into specific targets. Additionally, nanopolymers' biocompatibility can improve body tolerance. The body can biologically break down the biodegradable properties of nanopolymers, thereby reducing the risk of accumulation in the body. Nanopolymers, due to their wide availability, have become a focus of research in the field of nanotechnology to date[42].

The right nanopolymer selection is critical in determining the success of integration with herbal extracts, making it a key factor in achieving effective delivery and the desired therapeutic effects. Nanopolymers used in nanoparticle production involve a combination of synthetic and biopolymeric polymers. Examples of synthetic polymers include polyvinyl alcohol (PVA), polyethylene glycol (PEG), polyacrylic acid (PMAA), polyvinylpyrrolidone (PVP), polycaprolactone (PCL), and polylactic acid (PLA). On the other hand, examples of biopolymers include chitin, alginate, hyaluronic acid, and cellulose [43]. Several types of herbs and foods used in cancer treatment therapy include soursop (*Annona muricata* L.), beetroot (*Beta vulgaris* L.), carrot (*Daucus carota* L.), wheatgrass (*Triticum aestivum* L.), papaya (*Carica papaya* L.), turmeric (*Curcuma longa* L.), aloe vera (*Aloe vera* (L). Burm.f.), cucumber (*Cucumis sativus* L.), lemon (*Citrus x limon* (L). Osbeck), noni (*Morinda citrifolia* L.), ginger (*Zingiber officinale Roscoe*), and garlic (*Allium sativum* L.) [6].

Nanobiopolymer chitosan, advantages and disadvantages

Chitosan is an intriguing and promising biopolymer Chitosan is an intriguing and promising biopolymer for integration with herbal extracts. Deacetylation of chitin molecules forms this derivative, which is the second most abundant polymer in nature after cellulose. The amphiphilic biopolymer chitosan is very important for biotechnology and pharmaceutical uses because it is biodegradable, biocompatible, biostable, and specific to its target [44]. The activity of chitosan involves amino, hydroxyl, and oxygen bridge functional groups at positions C-2, C-3, and C-6. Therefore, chitosan can undergo hydrolysis, biodegradation, and redox reactions [45]. The cationic charge on the chitosan polymer makes it work and let it interact with other polymers through multilayer structures or electrostatic complexes [46].

Chitosan is acknowledged as a biopolymer that has several benefits in cancer therapy, especially as a medication carrier in nanoparticle format. Its advantages include biocompatibility and biodegradability, rendering it safe for internal usage and rapidly decomposable into non-toxic substances [47]. Furthermore, chitosan possesses a positive charge at physiological pH, facilitating enhanced interaction with negatively charged cell membranes, hence improving penetration, absorption, and immune response inside the tumor microenvironment, synergistically augmenting anticancer efficacy [48]. In cancer therapy, chitosan can improve the bioavailability of hydrophobic active chemicals, hence enhancing their deadly effects on cancer cells [49].

Nonetheless, despite its numerous benefits, chitosan possesses some limitations that warrant consideration. Its stability is comparatively low under specific physiological settings, particularly in environments with very acidic or basic pH, which may diminish its efficacy as a drug carrier, resulting in unregulated drug release and reduced retention period of the drug in the body. Consequently, investigations focused on enhancing the stability of chitosan under diverse physiological situations are essential to optimize its prospective applications in cancer treatment. At elevated concentrations, chitosan may induce toxicity or adverse immunological responses, but this risk is generally minimal at therapeutic levels. The biocompatibility and biodegradability of chitosan reduce the risk of adverse responses and facilitate progressive breakdown in the body [47]. Therefore, suitable formulation and characterisation are essential to enhance the benefits of chitosan while mitigating its limitations, facilitating its safe and successful application in nanoparticle-mediated cancer therapy.

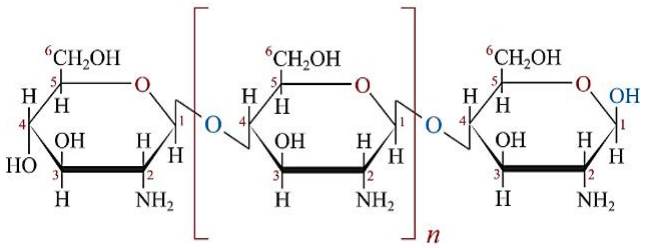

Fig. 1. Structure of chitosan [50]

The use of chitosan in cancer therapy

The utilization of chitosan as a foundational material for the creation of nanoparticles, nanocomposites, and drug-delivery polymers demonstrates promise in laboratory-based cancer treatment. The table below (Table 1) summarizes many studies investigating the role of chitosan in augmenting the efficacy of anticancer agents. Despite being based on in vitro or preclinical models, these research yield significant preliminary insights regarding the potential of chitosan in cancer therapy.

Table 1: Research on the use of chitosan in cancer therapy: in vitro and in vivo laboratory studies

No	Research	Year	Results
1	[51]	2023	Immobilization of olive leaf extract with chitosan nanoparticles results in a toxic reaction in MCF-7 breast cancer cells, with an IC50 of 810 µg/mL for the extract and a lowered IC50 of 540 µg/mL for the chitosan nanoparticles (OLE-CNPs). Lung cancer cells A549 exhibit an IC50 of 1000 µg/mL for the extract and an improved response with an IC50 of 540 µg/mL for the OLE-CNPs.
2	[52]	2020	Chitosan nanoparticles loaded with *Achillea magnifica* extract demonstrate antiproliferative activity against MCF-7 and MDA-MB-231 cells, with IC_{50} values of 0.300 mg/mL for MCF-7 and 0.320 mg/mL for MDA-MB-231, comparable to the chloroform extract, which exhibited IC_{50} values of 0.300 mg/mL for MCF-7 and 0.253 mg/mL for MDA-MB-231. The minimal enhancement in antiproliferative activity can be ascribed to the nanoparticles' low zeta potential, roughly 2.5 mV, signifying inadequate stability and a propensity for aggregation. This aggregation may diminish potency and obstruct the proper transport of active substances to cancer cells. Consequently, despite the development of nanoparticles, stability concerns may have led to comparable IC_{50} values to the chloroform extract, without exhibiting improved anticancer efficacy.
3	[53]	2020	Nano-ASLE, a chitosan nanoparticle formulation from *Annona squamosa* leaf extract, effectively inhibits HeLa cancer cell proliferation, as indicated by an IC_{50} value of 344.48 µg/mL. The research demonstrates that nano-ASLE markedly increases caspase-3 activity, yielding a mean expression score of 65.3 cells, so indicating that malignant cells are experiencing apoptosis via the mitochondrial pathway. The minimum recorded score of 45.3 cells further underscores the efficacy of nano-ASLE in promoting cell death.
4	[54]	2023	The extract of *Artemisia judaica*, when combined with chitosan nanoparticles (CNPsLE), exhibits considerable efficacy in combating cancer, evidenced by an IC_{50} value of 20.8 µg/mL against the human prostate cancer cell line (PC3), significantly lower than the IC_{50} of 76.09 µg/mL for the extract alone. CNPsLE demonstrates reduced minimum inhibitory concentration (MIC) values against many pathogens, underscoring its superior antibacterial efficacy relative to the extract alone.
5	[55]	2019	Chitosan nanoparticles integrated with pine bark extract (*Pinus merkusii*) have effectively obstructed the cell cycle and triggered apoptosis in cervical cancer cells, demonstrated by the nanoparticles' size of 394.3 nm and their cytotoxicity against HeLa cells, which exhibited an IC_{50} value of 384.10 µg/mL. Furthermore, these nanoparticles induced G0/G1 phase arrest and elevated the expression of p53 and caspase-9, hence reinforcing their function in facilitating apoptosis.
6	[56]	2023	Chitosan nanoparticles infused with *Plectranthus vettiveroides* root extract demonstrate cytotoxicity against oral cancer cells (KB cells), with toxicity contingent upon particle size and dosage. The findings indicated that chitosan nanoparticle (CNP) concentrations of 25 µg, 50 µg, 75 µg, 100 µg, and 125 µg markedly decreased the viability of KB cells. Nanoparticles infiltrate cells through multiple mechanisms, including macropinocytosis, clathrin-mediated transport, and endocytosis.
7	[57]	2016	Nanoparticles derived from *Selaginella doederleinii* leaf extract markedly impede the proliferation of cancer cells, particularly A549 cells, with an IC_{50} of 3% or 1020 µg/mL. Furthermore, these nanoparticles impede the proliferation of Chang cells (normal cells), exhibiting an IC_{50} of 4% or 1442 µg/mL. The optimal dose of nanoparticle extract that suppresses cancer cells while preserving normal cells is 0.5% or 167 µg/mL.
8	[58]	2017	Nanoparticles of curcumin loaded onto chitosan, alginate, and sodium tripolyphosphate exhibit cytotoxic activity and can enhance the expression of apoptosis genes in cervical cancer cells (HeLa cells).
9	[59]	2019	Chitosan nanoparticles infused with *Physalis alkekengi* demonstrate a notable cytotoxic effect, as evidenced by the MTT experiment, showing an escalation in cytotoxicity over 72 hours ($P < 0.05$). Moreover, these nanoparticles can elicit apoptosis at a rate of 71%, much beyond the 43% reported with the extract alone, when evaluated on HT-29 cells.
10	[60]	2023	Solid lipid nanoparticles of *Aloe perryi* extract coated with chitosan exhibit substantial cytotoxicity against A549 and MCF-7 cancer cells, with IC_{50} values of 11.42 ± 1.16 µg/mL for A549 cells and 8.25 ± 0.44 µg/mL for MCF-7 cells, signifying potent anticancer efficacy.
11	[61]	2021	Chitosan nanoparticles incorporating *Achillea goniocephala* extract demonstrate superior cytotoxic effectiveness relative to the unformulated extract. The cytotoxic efficacy of chitosan nanoparticles is markedly greater in nanoparticle form, despite their antioxidant activity being comparable to that of the extract. The IC_{50} value of *A. goniocephala* extract on MCF-7 cells was determined to be 56.780 µg, however the IC_{50} value of nanoparticles (NP) containing *A. goniocephala* extract was markedly lower at 40.860 µg, suggesting a more effective cytotoxic impact in nanoparticle form. In HT-29 cells, the IC_{50} value for *A. goniocephala* extract was

No	Research	Year	Results
			64.460 µg, whereas it was 45.482 µg for the NP containing *A. goniocephala* extract, thereby illustrating the superior cytotoxic activity of chitosan nanoparticles loaded with A. goniocephala extract.
12	[62]	2023	The study's results indicated the efficacy of *Selaginella doederleinii* extract as an anti-cancer drug, illustrated by its moderate cytotoxicity with an IC_{50} of 215 µg/mL against the MCF-7 cell line. The creation of effervescent nanoparticles from the extract was successfully accomplished, with Formula I exhibiting a pH of 6.55, granule dissolution in under 5 minutes, and a water content below 4%. This formulation enhances the bioavailability of the extract and utilizes the anticancer properties revealed by molecular docking studies, indicating that hexadecanoic acid from the extract demonstrates superior binding to the TP53 protein, potentially modulating its function in inhibiting cancer cell proliferation.
13	[63]	2019	Nano-ASLE, a chitosan nanoparticle produced from the leaf extract of *Annona squamosa*, exhibits the capacity to suppress the proliferation of human colon cancer (WiDr) cells, showcasing significant cytotoxicity with an IC_{50} value of 292.39 µg/mL. Research indicates that nano-ASLE can enhance the activity of caspase-3, signaling the demise of malignant cells via the mitochondrial pathway, resulting in cell cycle arrest at the G2/M phase and triggering apoptosis in WiDr cells.

Development of nanobiopolymers involving chitosan and herbal extracts by ionic gelation method

The development of chitosan nanobiopolymer with herbal extract can be achieved through the employment of the ionic gelation method. As a polysaccharide, chitosan's cationic characteristics facilitate interaction with negatively charged or polyanionic compounds, resulting in the formation of nanoparticles. The primary objective of utilizing this approach is to minimize or eliminate the use of organic solvents, high temperatures, and excessively vigorous stirring [37]. An example of this is the creation of conjugated nanoparticles incorporating chitosan and areca nuts. The preparation of betel nut (*Areca catechu*) nanoparticles involves several steps. Firstly, prepare a chitosan solution by mixing 0.1 and 0.2 grams of chitosan with 100 mL of a 1% acetic acid solution in distilled water. This solution is stirred using a magnetic stirrer at a speed of 1500 rpm. Subsequently, in the second step, a sodium tripolyphosphate solution is prepared by weighing between 0.1 and 0.2 grams of sodium tripolyphosphate and adding 100 mL of distilled water. This solution is also stirred using a magnetic stirrer. The third step involves preparing a Tween 80 solution by dissolving 0.5 mL of Tween 80 in 100 mL of distilled water, which is then mixed using a magnetic stirrer. In the final step, following the nanoparticle formation method, 0.5 grams of extract are mixed with the chitosan solution at various concentrations and stirred using a magnetic stirrer at a speed of 2500 rpm for 30 minutes. The sodium tripolyphosphate solution is then slowly added with continuous stirring for 30 minutes, followed by the addition of Tween 80 until a nanoparticle suspension is formed [64]. The formulation of chitosan nanoparticles with areca nut extract is provided in Table 2.

Table 2: Formulation of chitosan nanoparticles with *Areca catechu* extract [64]

Ingredient	Formula 1	Formula 2	Formula 3	Formula 4
Chitosan	0.1% (18 ml)	0.2% (18 ml)	0.1% (18 ml)	0.2% (18 ml)
TPP (Na-tripolyphosphate)	0.1% (9 ml)	0.2% (9 ml)	0.2% (9 ml)	0.1% (9 ml)
Tween 80	0.5% (3 ml)	0.5% (3 ml)	0.5% (3 ml)	0.5% (3 ml)
Extract	0.5 gram	0.5 gram	0.5 gram	0.5 gram

The ionic gelation process is widely used for producing extract nanoparticles. One example is the synthesis of chitosan nanoparticles utilizing *Annona squamosa* extracts. The preparation of *Annona squamosa* leaf extract-loaded chitosan nanoparticles (nano-ASLE) involves several steps. Firstly, a chitosan solution is prepared by dissolving chitosan in glacial acetic acid, while a solution of sodium tripolyphosphate is also prepared. Next, the chitosan solution is mixed with the *Annona squamosa* leaf extract, and the sodium tripolyphosphate solution is slowly added while stirring. Subsequently, the pH of the solution is adjusted using a 0.1 M NaOH solution, and the mixture is stirred for 2 hours at room temperature using a magnetic stirrer. After that, the mixture is centrifuged to separate the nanoparticles. The resulting supernatant is collected and dried to obtain the nano-ASLE, which is ready for use [63]. The formula for producing chitosan nanoparticles with *Annona squamosa* extract is presented in Table 3.

Table 3: Formulation of chitosan nanoparticles with *Annona squamosa* extract [63]

Ingredient	Concentration or amount
Annona squamosa leaves	1 gram
Distilled water	50 ml
Chitosan	100 ml (0.1% w/v)
Glacial acetic acid	0.25% v/v
Sodium tripolyphosphate	350 ml (0.84% w/v)
Sodium hydroxide (NaOH) solution	0.1 M

In the third case, an extract is used to create nanocapsules; the extract from noni (*Morinda citrifolia* L.) is the subject of particular attention. First, 1.5 grams of chitosan gel are dissolved in 100 milliliters of 1% acetic acid. Until the solution reaches clarity, homogenization with a magnetic stirrer is used to control particle size for sixty minutes. Then 0.1% Tween 80 is added, along with Noni extract. In the last stage, spray-drying combined with homogenization at 1500 rpm is used to dry the mixture and produce nanoparticles that are between 27 and 59 nm in size (Fig. 2.). An assessment of the nanocapsules at a dose of 250 mg/kg body weight shows that they can prevent the proliferation of fibrosarcoma cancer cells by 54.75% [65].

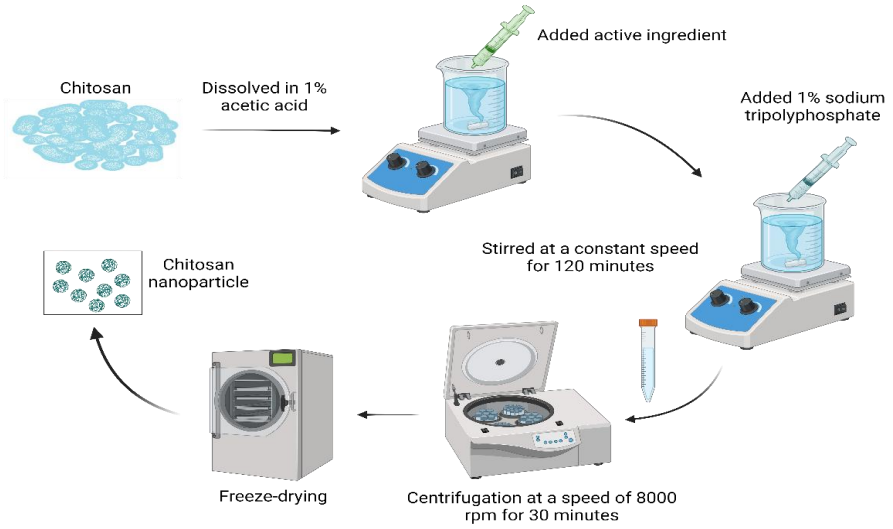

Fig. 2. Illustration of chitosan nanobiopolymers making process using ionic gelation method

Assessment of chitosan and herbal extract incorporation in pre-clinical trials

Chitosan is widely considered a non-toxic and biocompatible substance, rendering it a desirable option for numerous medicinal uses. Numerous studies have demonstrated that chitosan and its derivatives, including chitosan-based nanogels, exhibit no harmful effects on human cells. In several investigations, chitosan-based nanogels did not influence cell growth nor cause nitrogen oxide production in cell cultures. Despite studies about the potential genotoxicity of nanogels at specific dosages, there is no data indicating substantial harmful effects on human cells. Moreover, chitosan has been utilized in several medicinal applications, such as medication transport and hemostasis, hence reinforcing its safety and non-toxic characteristics. It is essential to perform a comprehensive risk assessment for each given application, particularly when nanomaterials are utilized, to guarantee safety and biocompatibility within their intended context of use [66].

The amalgamation of herbal extracts with chitosan for nanoparticle synthesis has become a prominent research focus in nanotechnology, demonstrating numerous benefits, particularly in medication delivery systems. Numerous studies have demonstrated the efficacy of this combination, emphasizing both an improved therapeutic profile and advantageous safety features associated with its application. A notable study demonstrated that chitosan nanoparticles infused with garlic (*Allium sativum*) extract displayed an impressive in vitro drug release rate of 90.65% [67]. Besides its drug release capability, the safety of this amalgamation of herbal extracts and chitosan has been thoroughly assessed across diverse cell types. Investigations on fibroblasts indicated that a gel formulation containing 1.2% *Camellia sinensis* and 1% chitosan was deemed safe, facilitating substantial fibroblast growth in Wistar rats [68].

Chitosan and *Arrabidaea chica* extract (AcE) packed in nanoparticles exhibit an excellent safety profile according to biocompatibility assessments conducted on human fibroblast cells. This work evaluated chitosan nanoparticles (NP) infused with *A. chica* extract (AcE-NP) with the MTT assay, which assesses cell viability as a measure of cytotoxicity. The findings indicate that cell viability (CV) was maintained at or above 80% across all evaluated concentrations, suggesting that AcE-NP did not generate detrimental cytotoxic byproducts. At low concentrations (0.001–0.125 mg/mL), both NP and AcE-NP preserved cell viability near 100%, indicating that encapsulating the extract in nanoparticles mitigates the potential harmful effects of the extract on fibroblast cells. Moreover, the pro-oxidant effects that may arise with flavonoid components at elevated concentrations, as noted at 0.5 mg/mL with the free AcE extract, were successfully mitigated via encapsulating in chitosan nanoparticles. At this elevated concentration, free AcE exhibited a viability reduction of up to 20%, whereas AcE-NP demonstrated more stable viability and even promoted cell proliferation at specific doses. This study substantiates the assertion that chitosan nanoparticles might improve the biocompatibility of herbal extracts, rendering them a safe and efficacious alternative for therapeutic applications, particularly in pharmaceutical systems necessitating modest yet effective dosages [69].

Furthermore, additional research has demonstrated the safety and efficacy potential of integrating chitosan nanoparticles with the root extract of *Leonotis nepetifolia* as an anticancer treatment. The produced chitosan nanoparticles shown considerable cytotoxic efficacy against many cancer cell lines, including cervical cancer (HeLa), breast cancer (MCF-7), and glioma (MO59J), particularly when used in conjunction with both normal and altered root extracts of *L. nepetifolia*. This

combination exhibited reduced IC_{50} values (0.608 mg/mL for HeLa, 0.306 mg/mL for MCF-7, and 0.112 mg/mL for MO59J), signifying a more potent anticancer impact than the individual application of nanoparticles or extracts. Moreover, among the concentrations evaluated, these nanoparticles exhibited no harmful effects on normal human fibroblast cells (Hs68), so strengthening their selective safety profile concerning healthy cells [70].

A separate study assessed the protective effects of Thymus serpyllum (TS) extract and TS-loaded nanoparticles against hydrogen peroxide-induced cytotoxicity in mesenchymal stromal cells (MSC) in vitro. The gas chromatography-mass spectrometry (GC-MS) analysis validated the spectrum of active constituents in the extract. Of the three extracts evaluated, the hexane extract demonstrated notable free radical scavenging efficacy. Treatment of MSC with H_2O_2 markedly elevated intracellular cell death; however, pre-treatment with TS extract and TS-loaded nanoparticles at a concentration of 200 µg/mL effectively mitigated the rise of cytochrome c (Cyt-c) and MMP13 generated by H_2O_2, while improving the survival rate of MSC. The alterations in cytokines caused by H_2O_2 (0.1 mM) were diminished through pre-treatment and co-treatment with the extract and nanoparticles at two distinct time intervals ($p < 0.05$). H_2O_2 elevated the rate of apoptosis, but treatment with TS-loaded nanoparticles markedly reduced the percentage of apoptosis ($p < 0.05$) [71].

Chitosan, a natural polymer, exhibits a favorable safety profile in several medicinal applications, including its use as a foundational material for nanoparticles. Studies demonstrate that chitosan nanoparticles exhibit negligible harmful effects when delivered intravenously, with no major hemodynamic alterations seen. The study revealed that chitosan nanoparticles, characterized by their modest size and slight positive charge, demonstrated negligible cytotoxic effects, along with minimal antiplatelet and anticoagulant activities. Moreover, despite a transient delay in the weight growth of the monitored rats, no indications of discomfort or distress were evident during the observation period. These data confirm that chitosan and its derivatives, including nanoparticles, exhibit substantial tolerance and can be deemed safe for therapeutic purposes, hence endorsing their utilization in drug delivery systems and other treatments [72]. These findings collectively underscore the potential applications of herbal extracts combined with chitosan for the development of effective and safe nanomaterial carriers in medicinal formulations.

3. Experimental

This article is a narrative review that seeks to consolidate existing knowledge on herbal medicine in cancer treatment, specifically emphasizing the function of nanobiopolymers in anticancer applications. Data were obtained from searches on PubMed, Google Scholar, DOAJ, and NIH, using keywords including "herbal medicine and cancer," "nanobiopolymer and anticancer," "nanopolymer and anticancer," "chitosan and herbal extract and anticancer," as well as supplementary terms such as "chitosan biopolymer and drug delivery," "chitosan nanoparticle safety," "characteristics of natural extracts," "active compounds of extracts and anticancer activity," and "advantages and disadvantages of chitosan."

The reviewed publications predominantly encompass the timeframe from 2013 to 2024, including several pertinent sources from preceding years. The inclusion criteria were publications that were completely accessible (beyond abstracts), copyright-protected, freely available, and authored in either Indonesian or English. Exclusion criteria encompassed studies that failed to align with the designated publication timeframe, articles without peer review (including preliminary reports or conference abstracts), and investigations centered on delivery systems that did not employ chitosan polymers. The number articles used were described in Fig. 3.

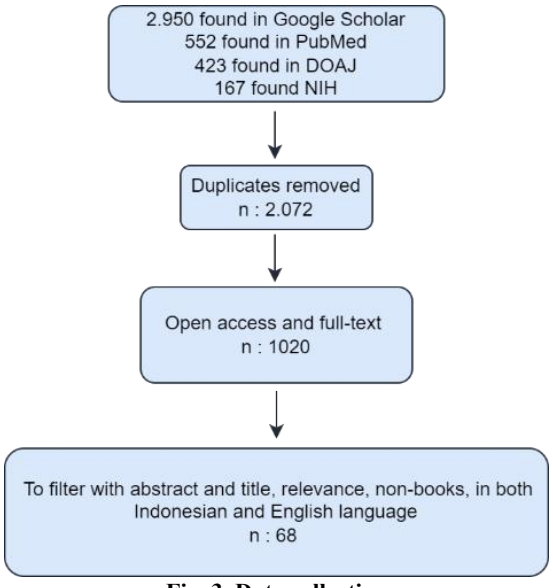

Fig. 3. Data collection

4. Conclusion

Cancer remains a prevalent disease affecting individuals of all ages and genders worldwide. Despite the widespread use of chemotherapy, its unpleasant side effects have led many individuals to seek alternatives, such as traditional herbal medicine. However, herbal products often suffer from low bioavailability and poor solubility. The application of chitosan nanobiopolymers shows promise in enhancing the potency of herbal extracts as anticancer agents by improving the solubility, stability, and bioavailability of active compounds in cancer therapy. This review highlights the significant potential of chitosan nanobiopolymers as carriers in cancer treatment, paving the way for further exploration. Future research should focus on optimizing chitosan nanoparticle formulations for specific types of cancer, understanding long-term safety, and conducting clinical trials to assess therapeutic efficacy in patients. With continued development, chitosan nanobiopolymers hold potential for clinical applications that could revolutionize cancer treatment by integrating natural and nanotechnology-based approaches.

5. Conflicts of interest

There are no conflicts to declare.

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