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*CORRESPONDENCE

Hamzeh Izadi, ⊠ izadi@vru.ac.ir

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Chitin: a comparison between its main sources

Hamzeh Izadi¹*, Homa Asadi¹ and Marjan Bemani²

Department of Plant Protection, Faculty of Agriculture, Valie-e-Asr University of Rafsanjan, Rafsanjan, Iran, Center for Research and Education of Agriculture and Natural Resources of Kerman Province, Organization of Research, Education and Extension of Agriculture, Rafsanjan, Iran

Chitin is the second most abundant polysaccharide on Earth, after cellulose, and it is mainly obtained from the shells of crustaceans. While chitin is currently derived from shrimp shells, there is a growing interest in commercializing chitin sourced from insects. This review compares chitin from various sources, including crustaceans, insects, fungi, and mollusks, based on several factors: purity, molecular weight, crystallinity, cost, and sustainability. Although crustaceans yield higher amounts of chitin and exhibit better crystallinity, insects present significant advantages in terms of sustainability, ease of extraction, and lower impurity levels. Chitin derived from insects is emerging as a sustainable alternative due to its simpler extraction processes and reduced environmental impact. This work highlights the increasing potential of insects as a reliable source for sustainable chitin production.

KEYWORDS

insects, chitin, chitosan, crustacean, arthropod, demineralization

1 Introduction

Natural polymers like chitin, starch, cellulose, hemicellulose, and alginate are widely used in our daily lives to perform various essential functions (Silva et al., 2021; Muthukumaran et al., 2022). Polysaccharides and their derivatives, especially chitin and starch, attract considerable attention due to their extraordinary chemical properties. Starch and chitin are byproducts classified as modified polymers, derived biopolymers, and polysaccharide-containing materials (Crini, 2005). Chitin has the chemical formula (C8H13O5N)n. The structure of chitin is most similar to that of cellulose, while its function is most similar to that of keratin. There are minor structural differences between cellulose and chitin, but they are chemically identical. Chitin has an acetamide group at the C_2 position instead of a hydroxyl group within the glucose unit (Hamed et al., 2016).

Chitin is a naturally occurring polysaccharide found abundantly in nature. Microfibrils of chitin are structural components found in the extracellular matrix of various invertebrates, including sponge skeletal fibers, mollusk shells, nematodes eggshell and pharynx, crustacean shells, arthropod exoskeletons, and fungi cell walls, forming natural biocomposites (Roberts, 1992; Ehrlich et al., 2007; Chen and Peng, 2019; Liu et al., 2019; Yu et al., 2024). Chitin in fungi is primarily sourced from the mycelium of species like *Mucor rouxii, Aspergillus niger*, and *Lentinus edodes* (Huq et al., 2022). Chitin makes up the exoskeleton, the wings, the antennae, and the trachea (Mei et al., 2024).

Chitosan and chitin are polysaccharides composed of N-acetyl-D-glucosamine and D-glucosamine units. Chitosan is derived from chitin through several methods, primarily involving chemical and enzymatic processes. Chemical methods are the

predominant approach that involves the deproteination, demineralization, decolorization, and deacetylation of chitin (Gandhi et al., 2014; Ibitoye et al., 2018). Deacetylation is a process that eliminates acetyl groups from chitin's molecular chain, yielding chitosan. The degree of deacetylation (DD) determines the extent of chitin conversion to chitosan (Kumar et al., 2020). DD varies based on factors such as NaOH concentration, reaction temperature, and duration. Chitosan's DD can span from 56% to 99%, influenced by production techniques and species. Chitosan represents a partially deacetylated form of chitin, where a portion of N-acetyl-D-glucosamine transforms into D-glucosamine (Kumari and Kishor, 2020). Enzymatic methods utilize chitinases and chitosanases to transform chitin into chitosan. This method is increasingly attracting attention due to its ability to produce chitosan with a well-defined structure and a lower environmental impact compared to chemical methods (Kaczmarek et al., 2019; Pratiwi et al., 2023). In microbial methods, certain fungi, such as Pleurotus spp., can extract chitosan through fermentation processes. This approach is regarded as ecofriendly and can yield chitosan with notable antimicrobial properties (Johney et al., 2016). The efficiency, cost, and environmental impact of these methods differ significantly, with enzymatic and microbial approaches offering more sustainable alternatives to conventional chemical extraction. Chitin and chitosan are not present in mammals but can be degraded in the body by several proteases such as lysozyme, papain, and pepsin (Pangburn et al., 1982). Chitin and its derivatives, chitosan, are utilized in various fields such as water treatment, cosmetics, healthcare, and agrochemicals. Their biodegradable nature and non-toxic properties make them valuable for sustainable practices. They are employed in producing organic fertilizers that have been shown to enhance crop productivity. Chitin promotes plant growth by fostering beneficial microbial activity in the soil, enhancing nutrient availability and absorption by plants. This results in higher crop yields and improved plant health (Sharp, 2013; Malerba and Cerana, 2020a). Chitin and its derivatives enhance plants' ability to withstand both biotic and abiotic stresses by activating their defense mechanisms. This protection helps safeguard crops against pathogens and environmental challenges, ultimately leading to increased productivity (Malerba and Cerana, 2020b; Shahrajabian et al., 2021). Chitin functions as a bioadsorbent, facilitating the remediation of contaminated soils by extracting heavy metals and enhancing soil characteristics. This process not only boosts soil fertility but also promotes sustainable agricultural practices (Malerba and Cerana, 2020b; Anedo et al., 2024). A recent study has shown that fertilizers enriched with chitin are effective in controlling nematode populations, which pose a threat to crops. This approach decreases the reliance on chemical nematicides, providing an environmentally friendly alternative (Kisaakye et al., 2024). Additionally, chitin is utilized in various industrial processes, including creating edible films and as a thickening and stabilizing agent in foods and food emulsions. Insoluble fibers, such as chitin, are indigestible in the human gastrointestinal tract. They are essential for increasing stool bulk and softness, which helps promote regular bowel movements and reduce intestinal transit time. Moreover, chitin serves as a dietary fiber that enhances gut microbiota, playing a crucial role in gut health. This enrichment can lead to improved gut function and may help prevent conditions such as constipation and hemorrhoids (Rodriguez et al., 2020; Kipkoech, 2023). However, insects offer an efficient method for chitin production, requiring significantly less land, water, and feed, which reduces their environmental impact. Their shorter life cycles and rapid growth allow for frequent harvesting, enhancing production efficiency. Additionally, insect farming is generally more cost-effective due to these lower resource demands and quicker growth rates, further decreasing overall production costs. Insect farming often uses organic waste as feed, promoting waste reduction and resource recycling in line with circular economy principles (Zainol Abidin et al., 2020a; Triunfo et al., 2022; Rehman et al., 2023; Saenz-Mendoza et al., 2023). These environmental and economic benefits make insect-based chitin a promising alternative to traditional sources. This review aims to qualitatively and quantitatively compare the primary sources of chitin production, focusing on their chemical properties, production methods, environmental impacts, and applications across various fields, including agriculture, healthcare, and industrial processes. It seeks to highlight the advantages and limitations of traditional and alternative sources, such as insect farming and evaluates sustainable and eco-friendly methods of chitin extraction.

2 The importance of chitin in insects and other organisms

Chitin is widely found in animal and plant kingdoms and is a crucial constituent of the exoskeletons of insects, crustaceans, and other arthropods. Its primary function is to provide a protective barrier for the inner soft tissues. Chitin acts as a watertight barrier, preventing delicate tissues from drying out. This helps to ensure the survival of these organisms by providing hydration protection to their inner tissues (Roberts, 1992; Moussian, 2019). The cuticles of insects, crustaceans, and spiders are mostly composed of sclerotized chitin, which can have diverse appearances. Chitin is also found in the iridophores of the eyes, the epidermis of cephalopods and arthropods, as well as the cuticles of some vertebrates (Zhang et al., 2021). It is also present in the cell walls of fungi and some algae. The exoskeleton of oysters and spiders, particularly Theraphosidae also contains chitin (Machałowski et al., 2020). Insects usually have 30%-45% protein, 25%-40% lipids, and 10%-15% chitin in their exoskeletons. In contrast, crustacean shells consist of 20%-40% protein, 20%-50% calcium carbonate, and 15%-40% chitin. Nevertheless, certain insects may contain more than 40% chitin in their exoskeletons (Spranghers et al., 2017; Mohan et al., 2020; Basawa et al., 2023).

Chitin is a major component of the insect's external body, providing the necessary support and protection against various external factors, such as physical damage, predators, and environmental stresses. Therefore, chitin plays a significant role in insects' survival, growth, and interaction with their surroundings. Insects heavily rely on chitin to form their cuticles and peritrophic membranes. However, the importance of chitin in insects can be summarized as follows: (Figure 1).

- Chitin provides structural support to insects' exoskeletons, protecting internal organs and safeguarding them from physical damage, providing and maintaining rigidity and shape to their bodies.
- The chitinous exoskeleton provides physical, pathogenic, and predatory protection, aiding insects in surviving their environment.



- Chitin acts as a watertight barrier against dehydration, preventing excessive water loss from the body.
- Insects undergo molting, a process by which they shed their exoskeleton to grow. Chitin, a flexible yet durable material, plays a crucial role in this process and can be shed and regenerated during each molt.
- Chitin provides the support and structure for insects with wings for efficient flight
- Chitin is present in sensory structures such as antennae and sensory hairs, where it aids in detecting environmental cues and communicating with the insect's surroundings.
- Chitin protects midgut epithelial cells and lines the foregut and hindgut, providing them with protection and support.

Overall, the unique properties of chitin make it an essential component in various biological contexts, and its metabolism is tightly regulated during growth and morphogenesis (Merzendorfer and Zimoch, 2003; Yu et al., 2024).

3 Sources of chitin

3.1 Crustacean

Crustaceans are a diverse group of arthropods that include shrimp, lobsters, crabs, and crayfish, among others. One of the defining features of crustaceans is their exoskeleton, which is composed primarily of chitin, a polysaccharide that provides structural support and protection. In crustaceans, chitin is integrated within a matrix of proteins and mainly carbonate minerals, which provide structural integrity and rigidity to the exoskeleton. The main sources of chitin are the shells of shrimp, crabs, and lobsters, which constitute a significant portion of seafood processing waste. These shells typically contain 6.0%–25.0% chitin by weight, making them a rich extraction source (Tables 1, 2). Utilizing crustacean waste for chitin extraction helps reduce environmental pollution and supports sustainable practices by transforming waste into valuable products. However, crustaceans are a vital source of chitin due to their structural makeup, biosynthetic processes, and the potential for sustainable extraction methods (Younes and Rinaudo, 2015; Zhang et al., 2021; Amiri et al., 2022; Verardi et al., 2023).

3.2 Insects

Chitin is a long-chain polymer of N-acetylglucosamine, derived from glucose, and serves as a key structural component in the exoskeletons of arthropods, including insects. Insects are a significant natural source of chitin, constituting about 1.2%-60.0% of the dry weight of their cuticles, which vary by species and developmental stage (Merzendorfer and Zimoch, 2003; Hahn et al., 2020). Chitin forms a robust outer layer of the exoskeleton and is essential to the peritrophic matrix that lines the gut, acting as a barrier against pathogens and aiding digestion (Tables 1, 2). This polysaccharide provides structural integrity and rigidity to the insect exoskeleton while serving as an attachment matrix for cuticular proteins. Insects, such as beetles, crickets, flies, locusts, and mealworms, are commonly harvested for chitin extraction. Compared to crustaceans like shrimp and crabs, insects are easier to farm and process, making them an environmentally sustainable and attractive source of chitin, particularly in sustainable agriculture and waste reduction contexts (Merzendorfer and Zimoch, 2003; Hahn et al., 2020; Muthukrishnan et al., 2020).

TABLE 1 A comparison between animal and non-animal sources of chitin.

Source	Example	Chitin content (%)	Chemical structure	Extraction	Applications
Animal	Crustaceans, Insects, Mollusks	1.2-60.0	High-purity chitin; often found in the form of chitinous shells	Demineralization and deproteinization	Biomedical, drug delivery, biodegradable material
Non-animal	Fungi (Mushrooms)	3.8–13.6	It often contains chitosan, which is readily soluble and has different properties	Residue extraction	Food industry, pharmaceuticals

Both animal and non-animal sources of chitin offer unique advantages and challenges. Animal sources like crustaceans provide high yields of chitin but raise sustainability concerns due to overfishing practices. In contrast, non-animal sources such as fungi present a more sustainable alternative but may yield lower quantities of chitin. The choice between these sources often depends on the intended application, availability, and environmental considerations (Casadidio et al., 2019; Lopez-Santamarina et al., 2020; Abo Elsoud et al., 2022; Rehman et al., 2023).

TABLE 2 Sources and properties of chitin.

Source	Tissue	Main form	Advantages	Disadvantages	Application	References
Fungi	Cell walls	α-Chitin	1-Environmentally friendly extraction processes, 2-Uniform and pure products, 3-Free from heavy metals 4- Easy to cultivate, 5- Unique molecular weight homogeneity, 6-Valued for chelating activity, biocompatibility, and antimicrobial properties	1 - Less abundant, 2- Specific cultivation conditions, 3 - May have variable chitin content depending on the life cycle and storage conditions		Abo Elsoud et al. (2022)
Crustaceans	Exoskeletons	α-Chitin	 Abundant and readily available, Well-established extraction methods, High molecular weight, and degree of acetylation 	1-Harsh demineralization and deproteinization processes 2-Chemical extraction methods have environmental drawbacks	Biomedical, Food Industry, Agriculture, Cosmetics	Casadidio et al. (2019), Amiri et al. (2022)
Insects	Exoskeletons	γ-Chitin	 Abundant and renewable resource, Easy to cultivate, High purity, 4- Diverse sources, Environmental benefits 	 Lower chitin content Complex extraction processes 		Triunfo et al. (2022), Rehman et al. (2023)
Mollusks	Radulae and beaks	β-Chitin	1-Structural integrity, 2- Specific applications, 3- Less contaminated	1- Lower availability 2- Complex extraction	-	

3.3 Fungus

Chitin is a versatile biopolymer found in various sources, each with distinct properties and applications (Figure 2) (Abo Elsoud and El Kady, 2019). Fungi are becoming an important source of chitin, with values reported of 8%–16% (w/w) chitin (plus chitosan) on a dry matter basis, mainly present in their cell walls. Notably, Zygomycetes fungi contain more chitin than other fungal groups.

This chitin is covalently bonded to glucans, complicating extraction while imparting unique properties for diverse applications (Cord-Landwehr and Moerschbacher, 2021; Huq et al., 2022). Fungal sources of chitin and chitosan have several advantages over other sources, such as unique molecular weight homogeneity, charge and viscosity distribution, and the absence of heavy metals (Bastiaens et al., 2019). Moreover, fungal chitin, sourced from food and biotechnology waste streams, provides a sustainable option



with consistent quality, unlike crustaceans (Cord-Landwehr and Moerschbacher, 2021). Fungal chitin is often preferred in biomedical applications due to its high purity and consistent quality. It's also non-toxic and biodegradable, making it suitable for various industries. Therefore, fungal chitin is preferred for vegan and cosmetic products due to its non-animal origin, appealing to consumers seeking cruelty-free options. Moreover, its availability is stable year-round, making it a reliable resource for industrial use (Cord-Landwehr and Moerschbacher, 2021; Hug et al., 2022). However, extracting chitin from fungal cell walls poses challenges, primarily due to the complex structure resulting from covalent bonds with glucans, which can lead to lower purity levels (Cord-Landwehr and Moerschbacher, 2021) Moreover, compared to other sources like crustacean shells, chitin from fungal sources, such as Ganoderma lucidum, may have lower thermal stability but can still be a viable alternative (Philibert et al., 2017). However, fungi are a promising source of chitin, providing opportunities for sustainable production and various applications across multiple industries (Tables 1, 2). Some studies have also explored the concurrent production of chitin from both shrimp shells and fungal sources, where the fungi can help deproteinate and demineralize the shrimp shells, providing a nitrogen source for their growth (Abo Elsoud and El-Kady, 2019).

3.4 Mollusca

Chitin is primarily known from crustaceans but is also present in mollusks, which include squids, cuttlefish, snails, clams, and chitons (Weiss, 2012; Chan et al., 2018). In mollusks, chitin is mainly found in shells, pens, and structural components, though in lower quantities than crustaceans. Molluscan chitin has a high degree of polymerization and crystallinity, providing excellent mechanical strength and biocompatibility (Tables 1, 2). It often exists alongside proteins and minerals, contributing to structural integrity, particularly in cephalopods like squids, where chitin forms specific associations with various proteins (Weiss, 2012; Younes and Rinaudo, 2015; Chan et al., 2018).

4 Physicochemical properties of chitin

The physicochemical characteristics of chitin found in insects and other organisms can vary slightly, which affects properties like mechanical strength and stiffness (Figure 3 and Table 3). Some traits that may exhibit differences include crystallinity, molecular weight, acetylation level, fiber structure, and crosslinking with other molecules (Wang et al., 2013; Zargar et al., 2015; Moussian, 2019; Khajavian et al., 2022). Chitin is a linear polymer of N-acetylglucosamine units linked by β -1,4 glycosidic bonds. It forms a robust and flexible matrix in insects and other organisms, offering structural support and protection. Chitin imparts mechanical strength and toughness to insect and crustacean exoskeletons, with properties influenced by crystallinity, molecular weight, and acetylation degree. Despite a similar basic structure, variations in chitin's physicochemical properties contribute to diverse functions and adaptations in these organisms (Merzendorfer and Zimoch, 2003; Erko et al., 2013; Moussian, 2013; Liu et al., 2019; Machałowski et al., 2020; Mohan et al., 2020; Zhang et al., 2021). However, the physicochemical, morphological, and structural characteristics of chitin and chitosan are influenced by the characteristics of the species, such as life stage, metamorphosis type, and genus (Mohan et al., 2020; Mohan et al., 2022). The main characteristics of chitin are (Figure 2).

4.1 Crystallinity

Chitin is recognized for existing in multiple allomorphic forms, each characterized by varying degrees of crystallinity resulting from the hydrogen bonding interactions between its polymer chains. The degree of crystallinity (CrI) in chitin can fluctuate considerably, influenced by its source and the methods employed in its preparation (Table 4). Typically, CrI values range from 0.57 to 0.93, depending on the specific calculation method utilized (Casadidio et al., 2019). The crystallinity of chitin is a measure of the degree to which the polymer exhibits a regular, ordered structure (Soon et al., 2018). Crystalline regions within chitin provide strength and rigidity to structures like the exoskeleton, while amorphous regions contribute flexibility (Ogawa et al., 2011; Wang et al., 2013). Comparing the crystallinity of chitin from different sources involves examining the structural order and organization of its polymer chains across various biological origins. Several factors, including the source of the chitin, extraction methods, and processing conditions, influence its crystallinity. Chitin fibers associate with one another in three possible crystalline arrangements: α -chitin, β -chitin, or γ chitin. These arrangements further contribute to chitin's structural and functional diversity, with each allomorph exhibiting unique



TABLE 3	The physicocl	hemical prope	rties of chitin	derived from	various sources.

	Source	Crystallinity (%)	Molecular weight (kDa)	Purity (%)	Solubility behavior
Animal	Shrimp Shells	60-80	100-300	85–95	Insoluble in water; soluble in dilute acids
	Crab Shells	70–90	150-400	90–95	Insoluble in water; soluble in dilute acids
	Insects (e.g., Cockroaches)	50-70	50-200	80-90	Insoluble in water; more easily broken down by chitinase
	Silkworms	60-75	100-300	85-95	Insoluble in water; soluble in dilute acids
Non-animal	Mushrooms	40-60	20-150	70-85	Insoluble in water; limited solubility in organic solvents
	Fungi (General)	30-50	20-150	60-80	Insoluble in water; some solubility in organic solvents

Crystallinity: Chitin from crustaceans generally exhibits higher crystallinity than fungal sources, affecting its mechanical properties and solubility. The molecular weight of chitin varies significantly across sources, influencing its functional characteristics and applications. Due to established extraction methods, purity levels are generally high for chitin derived from animal sources, particularly crustaceans. All chitin sources are insoluble in water but exhibit varying solubility in acids and organic solvents, which is crucial for their processing and application.

properties and degrees of crystallinity (Figures 4–6) (Lv et al., 2023). Two-dimensional sheets of α -chitin crystals form laminae. These sheets consist of antiparallel chitin fibers associated with β -folded proteins. α -Chitin is a primary constituent of the arthropod cuticle, crustaceans' shell, and the fibrous extracellular matrix in sponges (Wang et al., 2013; Kumari and Kishor, 2020 January 1). β -Chitin found in hard biomaterials like the squid beak, β -chitin forms a tri-dimensional composite material with proteins. γ -Chitin is less well-studied and contributes to the physical barrier found in insect cocoons (Moussian, 2019). Chitin is a structural polymer that produces strong fibers both inside and outside cells. The fibers bond weakly with one another, increasing overall structural strength (Moussian, 2019; Hou et al., 2021). Moreover, chitin can be crosslinked with other molecules to enhance its strength, stability, and resistance to degradation (Jiao et al., 2023). For example, in fungal cell walls (e.g., *Candida albicans*), chitin forms strong bonds with glucans. Among arthropods and other invertebrates, chitin links with proteins either covalently or non-covalently (Arroyo et al., 2016; Wegrzynowska-Drzymalska et al., 2020; Wang et al., 2021). However, based on crystallinity, different sources of chitin can be categorized as: High crystallinity: Chitins derived from crustaceans such as crabs and shrimp typically demonstrate higher crystallinity.

Chitin structure	Degree of crystallinity (CrI)	Properties	References	
α-Chitin	0.72	Tightly packed, antiparallel, high crystallinity, strong hydrogen bonding, high mechanical strength, and low solubility	Cho et al. (2000), Nishiyama et al. (2011), Khasanah et al. (2015), Ramírez-Wong et al. (2016)	
β-Chitin	0.57	Loosely packed, parallel, lower crystallinity than α-chitin, more flexibility, higher water-binding capacity, and increased solubility		
γ-Chitin	Varies	Mixed arrangement (combination of α and β forms), intermediate properties between α - and β -chitin, rare in nature		

TABLE 4 The crystallinity properties of different chitin structures.

This is due to the highly ordered arrangement of chitin chains in their shells, which provides the necessary rigidity and strength. The crystallinity in crustaceans is influenced by the tight association with calcium carbonate and proteins, forming a complex matrix that enhances structural order (Wang et al., 2013; Pădurețu et al., 2019; Poerio et al., 2020; Hou et al., 2021; Jędrzejczak et al., 2024). Moderate to high crystallinity: Insect chitin also exhibits a relatively high degree of crystallinity, although it may be slightly lower than that of crustaceans. This is because insect cuticles are composed of chitin fibers embedded in a matrix of proteins, which may not be as densely packed as in crustaceans. The crystallinity can vary among different insect species and developmental stages. For example, some insects have softer, less crystalline exoskeletons in their larval stages compared to the highly crystalline cuticles in adults (Zhang et al., 2000; Sáenz-Mendoza et al., 2018; Henriques et al., 2020; Kumar et al., 2020; Poerio et al., 2020; Soetemans et al., 2020; Triunfo et al., 2022). Low crystallinity: Chitin extracted from fungi typically exhibits lower crystallinity compared to that from crustaceans and insects. In fungi, chitin is combined with other polysaccharides such as glucans and proteins, forming a less ordered matrix. Significant amorphous regions and a more heterogeneous composition in fungal cell walls contribute to the lower overall crystallinity (Blumenthal and Roseman, 1957; Velásquez and Pirela, 2016; Poerio et al., 2020).

Chitin's crystalline arrangements and its association with proteins define the mechanical properties of tissues and organisms, making it a versatile extracellular polysaccharide. The variation in crystallinity reflects differences in the biological roles and structural requirements of chitin in these organisms. Crustaceans need a highly ordered structure for their protective shells, insects require moderately crystalline exoskeletons for flexibility and protection, while fungi have less crystalline chitin due to their distinct structural and functional requirements (Kabalak et al., 2020; Iber et al., 2022).

4.2 Molecular weight

Comparing the molecular weight (MW) of chitin from different sources involves analyzing the polymer length of chitin chains. The molecular weight of chitin varies depending on the source, which affects its properties and applications. Chitin is a biopolymer made up of over 5,000 sugar units, with varying molecular weights (MW) depending on the number of monomeric units in the biopolymer, the source, and the deacetylation treatment (Yeul and Rayalu, 2013; Hahn et al., 2020). Here's how these sources compare based on molecular weight: High molecular weight: Chitin from crustaceans generally has a high molecular weight. This is because crustaceans produce long, continuous chitin chains, which contribute to the mechanical strength and rigidity of their exoskeletons (Reddy and Yang, 2015; El Knidri et al., 2018; Zhang et al., 2018; Kumari and Kishor, 2020; Hou et al., 2021). Moderate to high molecular weight: Insect chitin usually has a slightly lower molecular weight compared to crustacean chitin. However, it still tends to be relatively high due to the need for durable and flexible exoskeletons. The molecular weight of insect chitin can vary widely among different insect species and life stages (Sáenz-Mendoza et al., 2018; Kumar et al., 2020; Kumari and Kishor, 2020; Hou et al., 2021; Pedrazzani et al., 2024). Low molecular weight: Chitin from fungal sources often has a lower molecular weight than that from crustaceans and insects. Fungal chitin is synthesized in shorter chains, which are adequate for the structural roles they play within the cell walls. The molecular weight of fungal chitin generally ranges from about 50,000 to 300,000 Da (Hou et al., 2021).

Chitin from crustaceans generally has the highest molecular weight, followed by chitin from insects, with fungal chitin having the lowest molecular weight. This variation reflects the different biological requirements and structural roles of chitin in these organisms. Crustaceans require long, high molecular weight chitin chains for strong and rigid exoskeletons, insects need moderately high molecular weight chitin for flexible yet protective exoskeletons, and fungi have shorter chitin chains suitable for their cell wall structures.

4.3 Purity

Comparing the purity of chitin from different sources involves assessing the level of contaminants or impurities present in the chitin extract. Purity can significantly impact the quality and suitability of chitin for various applications. Here's how these sources compare based on purity: High purity: Chitin extracted from crustacean shells can achieve high levels of purity with proper processing



techniques. However, achieving absolute purity can be challenging due to the presence of other biomolecules, such as proteins, lipids, and minerals, which are often tightly associated with chitin in the exoskeleton. Common contaminants in crustacean chitin include proteins like chitin-binding proteins and residual pigments from the shells (Zhang et al., 2018; Panchal and Desai, 2022). Moderate purity: Chitin from insect exoskeletons can also be purified to relatively high levels, but it may contain more impurities compared to crustacean chitin. Similar to crustaceans, insect chitin is associated with proteins and other biomolecules. Insect chitin may contain proteins, pigments, and other components present in the cuticle matrix (Panchal and Desai, 2022). Low purity: Chitin extracted from fungal cell walls generally has lower purity compared to crustacean and insect chitin. Fungal cell walls contain a complex mixture of polysaccharides, proteins, and other cell wall components, which can be challenging to separate from chitin. Common contaminants in fungal chitin include glucans, proteins, lipids, and other cell wall constituents (Panchal and Desai, 2022).

Achieving high-purity chitin requires careful extraction and purification processes to minimize contaminants and impurities. Crustacean chitin can be purified to relatively high levels due to the availability of well-established extraction methods, while insect chitin may contain slightly more impurities. Fungal chitin, on the other hand, is often associated with a more complex mixture of cell wall components, making it more challenging to obtain pure chitin extracts (Panchal and Desai, 2022).

4.4 Solubility

The ratio of N-acetyl-D-glucosamine to β -linked Dglucosamine units determines the molecular weight and deacetylation degree (DD0 of chitosan. Higher DD generally leads to increased water solubility and improved biological activities (Amor et al., 2023). Chitin molecules can be partially or fully acetylated, impacting the polymer's properties. Chitin is usually

more acetylated in insects than in other arthropods, affecting solubility and enzymatic degradation. While insoluble in most solvents, chitin can dissolve in strong acids or bases. Its solubility in insects and arthropods depends on acetylation degree and exoskeleton components. Chitin is composed of repeating Nacetylglucosamine (GlcNAc) units. The degree of acetylation (DA) refers to the proportion of acetyl groups attached to the GlcNAc units. In chitin, the DA is typically around 0.90, indicating the presence of certain amine groups (though some deacetylation can occur during extraction, resulting in about 5%-15% amine groups in chitin). Acetylation level is a crucial factor that determines the properties of chitin. The degree of acetylation (DA) measures the proportion of acetyl groups in chitin and affects its physicochemical properties, such as solubility, chain conformation, and reactivity (Mouyna et al., 2020). Chitins may have different levels of acetylation depending on their sources such as fungi, insects, crustaceans, or mollusks, which may affect their potential uses (Brigode et al., 2020). Low deacetylation degree: Chitin from crustacean shells typically has a low DD, ranging from 10%-15%, when deacetylated using conventional chemical methods like treatment with concentrated alkali solutions. The highly crystalline structure of crustacean chitin makes it resistant to deacetylation (Zhao et al., 2010; Poerio et al., 2020; Novikov et al., 2023). Moderate deacetylation degree: Chitin from insect exoskeletons, like those of silkworms or bees, can reach a DD of around 50%-70% through chemical methods. The degree of deacetylation depends on the specific insect source and the deacetylation conditions. For example, chitosan extracted from Blaps lethifera, Pimelia fernandezlopezi, and Musca domestica showed different DD values (Zhao et al., 2010; Poerio et al., 2020; Amor et al., 2023). High deacetylation degree: Chitin derived from fungal cell walls, such as those from M. rouxii or A. niger, can achieve a higher DD of around 70%-90% through chemical deacetylation. Fungal chitin has a less crystalline structure compared to crustacean chitin, making it more susceptible to deacetylation (Zhao et al., 2010).



(C) Chitin 3 was obtained from the darkling beetle, Zophobas morio larvae, over a scanning angle range of 5°–45° at 30 kV and 30 mA. This data is reprinted with permission (5950520533931) from (Soon et al., 2018).

As the degree of deacetylation increases, chitin becomes more soluble and transforms into chitosan, which exhibits greater solubility in aqueous solutions, particularly under acidic conditions. The solubility of chitosan can be tailored by controlling the degree of deacetylation during its production. Fungal chitin generally achieves the highest DD through chemical deacetylation, followed by insect cuticles and crustacean shells. Enzymatic deacetylation offers the potential for higher and more controlled DD, but its efficiency varies based on the chitin source and process parameters (Zhao et al., 2010; Poerio et al., 2020; Novikov et al., 2023).

5 Minerals

Besides chitin and proteins, the arthropod cuticles and crustacean shells also contain various minerals that make them



strong and rigid. These minerals include calcium carbonate, calcium phosphate, and magnesium carbonate. (Erko et al., 2013; Moussian, 2013). The minerals found in arthropod cuticles and crustacean shells are crucial for their mechanical properties and structure. They add strength and rigidity to the cuticle, which acts as a protective shield for the animal's body against any damage. Some animals have heavily mineralized cuticles, which makes them appear as if they have a hard, shell-like exterior. This is commonly observed in crustaceans like crabs and lobsters (Plotnick, 1990; Erko et al., 2013). The main sources of chitin differ in their demineralization requirements during the extraction process. The demineralization step is crucial for crustacean shells due to their high mineral content, while it is less critical for insect cuticles and not required for fungal chitin. The use of dilute HCl is the most common method for demineralization across all sources, but the specific conditions, such as acid concentration, temperature, and duration, may vary depending on the source (Younes and Rinaudo, 2015; Ibram et al., 2019; Abidin et al., 2020; Pellis et al., 2022).

Here's a comparison of the common chitin sources based on the demineralization step (Zhao et al., 2010; Ibram et al., 2019; Vicente et al., 2021; Pellis et al., 2022; Novikov et al., 2023): Crustacean shells: Chitin from crustacean shells requires a demineralization step to remove the high mineral content, primarily

calcium carbonate. Insect cuticles: Insects have fewer minerals than the shells of crustaceans (Khayrova et al., 2021). However, chitin from insect exoskeletons also requires a demineralization step, but the mineral content is generally lower than in crustacean shells. As a result, chitin and chitosan extracted from insects may have different physicochemical attributes than those extracted from crabs and shrimp (Saenz-Mendoza et al., 2023). Fungal chitin: Chitin derived from fungal cell walls does not require a demineralization step, as fungi generally have a lower mineral content than crustacean shells. Unlike other invertebrates (such as sponges, mollusks, or crustaceans), the cuticle of spiders lacks minerals such as CaCO3, which allows for the elimination of the demineralization step during chitin separation (Machałowski et al., 2020). The origin of chitin affects not only its crystallinity and purity but also its polymer chain arrangement and thus its properties (Rinaudo, 2006; Kumari and Kishor, 2020). The dosage of the main mineral elements present in the exoskeleton of different chitin sources shows that the content varies with the species and thus the processing conditions (Tolaimate et al., 2003). Two separate studies have shown differences in the structure of chitin obtained from different species of insects. In the first study (Waśko et al., 2016), chitin isolated from adults of Hermetia illucens was found to have a relatively smooth surface and was composed of parallel distributed fibers. In comparison, the larval chitin had a more complex structure, with a convex shape composed of repeated honeycomb-like units. In the second study (Erdogan and Kaya, 2016a), chitin obtained from adults and nymphs of Dociostaurus maroccanus was found to be composed of long nanofibers with large nanopores. High similarity was found in the properties of chitin and chitosan from nymphs and adults of the species. In experiments conducted on the potato beetles, Leptinotarsa decemlineata, it was found that chitin extracted from the adults and larvae of the species had a similar structure to nanofibers but differed in the number of pores. A study investigated chitin structure in four grasshopper species using various analytical techniques. Differences in chitin quantity and surface structure were observed between male and female grasshoppers. Despite gender variations, elemental analysis, thermal properties, and crystalline index values for chitin were similar. Enzymatic digestion showed no significant differences in digestion rates between chitins from both sexes and commercial chitin (Kaya et al., 2015e).

The deproteinization process for chitin extraction involves the removal of protein content from the raw material. The deproteinization method should be used to extract chitin from different sources. Deproteinization of chitin involves the use of alkaline solutions, primarily NaOH, to eliminate the protein content present in the raw material (Zainol Abidin et al., 2020b).

6 Different forms of chitin in insects and other arthropods

There are three different crystalline allomorphs of chitin found in nature which include α -chitin, β -chitin, and γ -chitin (Erdogan and Kaya, 2016a; Kaya et al., 2017a; Hou et al., 2021). α -chitin is the predominant chitin in insects and other arthropods (Figure 4). It has a highly ordered crystalline structure, with chitin molecules arranged in parallel chains. α -chitin is recognized for its high tensile strength and rigidity (Hou et al., 2021). It is the most consistent polymorphic form of chitin in nature and is present in crustaceans such as shrimps, lobsters, and crabs, as well as in the chitinous cuticles of insects. It can also be found in fungal cell walls (Ifuku et al., 2009; Hassainia et al., 2018). In a study, the structure of α -chitin has been determined through X-ray diffraction, revealing an orthorhombic unit cell with specific dimensions and a space group of P212121. The chains form hydrogen-bonded sheets and have a statistical mixture of side-chain orientations. The structure contains two types of amide groups with different hydrogen bonding, explaining the lack of swelling in water due to extensive intermolecular hydrogen bonding (Minke and Blackwell, 1978).

β-chitin is a less common form of chitin found in certain species, such as some crustaceans and mollusks. It has a different crystal structure compared to a-chitin, with the chains of chitin molecules arranged in an antiparallel fashion. This form of chitin is less rigid than α-chitin. It is characterized by loosely packed parallel chains with weak intermolecular interactions, making it more soluble and droplet-like than the α form. Additionally, the β form of chitin has been derived from squids (Pillai et al., 2009; Hamed et al., 2016; Hossin et al., 2021; Hou et al., 2021). y-chitin is a type of chitin that is found in certain types of algae and fungi. It has a unique crystal structure that distinguishes it from the more wellknown α -chitin and β -chitin. This is a less common form of chitin that has a disordered structure with random orientations of Nacetylglucosamine units. However, further research is required to fully comprehend its characteristics and prevalence in insects and other arthropods (Hou et al., 2021). A schematic shape of three forms is depicted in Figure 4. There is a mix of α and β forms in γ chitin. The presence of γ -chitin is lower than that of α and β forms, and it has been examined in only a few studies. It is noteworthy that a chitin can convert to the β form, but the reverse is not true (Velásquez and Pirela, 2016; Hou et al., 2021).

Both α and β chitins keep a powerful network dominated by intrachain hydrogen bonds between the groups of C=O···NH and C==O…OH within an interval of 0.47 nm. In α form conformation, supernumerary inter-chain hydrogen bonds connect the hydroxymethyl groups while this type of interaction is not observed in the β conformation (Roy et al., 2017). These three types differ mainly in the degree of hydration, the size of the unit cell, and the number of chitin chains in each cell (Merzendorfer and Zimoch, 2003; Yu et al., 2024). On the other hand, the properties of pure chitin depend on their molecular weight, degree of acetylation, purity, and dispersion index. For example, in a study, the chitin of crab shells and mealworms was determined by X-ray diffraction and FTIR analysis. In this research, a form of chitin was observed in four crystalline reflections, shown at 9.4°, 19.3°, 20.8°, and 23.3° in the chitin of crab shell, and at 9.44°, 19.3°, 20.7°, and 23.3° in the chitin of mealworm by crystalline structure. a-form showed doublet at amide I band in crab shell and mealworm chitin (Kaur and Dhillon, 2015). In another study, chitin was extracted from various marine crustacean shells using chemical methods, showing a chitin content of 17.50%-23.75% on a dry weight basis. The chitin had low molecular weight and was similar to commercial chitin based on various analyses. The extracted chitin exhibited α form characteristics, with a crystalline index value ranging from 80.3% to 80.8%. Analysis showed nanofiber and nanopore structures (Mohan et al., 2021). Chitin polymorphs α , β , and γ were isolated and analyzed for their crystalline structures and characteristics using

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various spectroscopic and analytical techniques. Molecular weights, spectral features, X-ray diffraction patterns, and thermal properties were determined for each chitin type. Differences in crystalline reflections, thermal decomposition temperatures, and activation energies were observed, highlighting the distinct properties of achitin, β -chitin, and γ -chitin. The study also demonstrated the temperature sensitivity differences among the chitin polymorphs based on their activation energies (Jang et al., 2004). Chitin can have different crystalline organizations and interact with chitin-binding proteins to create higher-order structures. This combination defines the mechanical properties of tissues and organisms (Moussian, 2019). Analysis through techniques like ATR-FTIR, NMR spectroscopy, and WAXS confirmed Bombyx mori as a promising source of α -chitin (Jędrzejczak et al., 2024). Overall, chitin, an abundant biopolymer, exhibits diverse structural conformations through natural processes like biopolymerization and self-assembly, leading to unique physical effects and mechanical properties (Hou et al., 2021).

7 Comparison of chitin content in insects and other organisms

Chitin is an amino polysaccharide found in yeast cells, arthropod cuticles, fungal cell walls, and crustacean shells (Rehman et al., 2023; Sanjanwala et al., 2024). When comparing these sources of chitin, several factors come into play: Purity, as the purity of chitin extracted from different sources can vary. Some sources may require more extensive purification processes to remove impurities such as proteins and minerals; Yield: The quantity of chitin extracted from a specific source varies depending on the species, size, and processing methods employed. Some sources may contain higher amounts of chitin compared to others. The quality of chitin can also vary depending on factors such as the degree of deacetylation (which affects its solubility and other properties) and the presence of contaminants; Cost and availability: The cost and availability of chitin from various sources can vary depending on factors such as geographical location, seasonal variability, and demand.

The choice of chitin source depends on factors such as intended applications, availability, cost considerations, and sustainability concerns. Researchers and industries are exploring various sources and methods for chitin extraction to meet the increasing demand for this versatile biopolymer. If we are ranking sources based solely on the amount of chitin that can be obtained, it generally follows this order (Merzendorfer and Zimoch, 2003; Ma et al., 2020; Khayrova et al., 2021; Iber et al., 2022; Lv et al., 2023; Thakur et al., 2023): Crustacean shells: Crustaceans have relatively large exoskeletons compared to insects and fungi. Therefore, they typically yield the highest amount of chitin per individual organism; Insect exoskeletons: Although insects have smaller bodies than crustaceans, they often exist in larger populations and have a higher overall biomass. Therefore, insect exoskeletons can still yield a significant amount of chitin, especially if sourced from species that are abundant or cultivated in large numbers; Fungal cell walls: Fungi generally have smaller biomass than insects and crustaceans. Additionally, not all fungi produce chitin in significant quantities. Therefore, while fungal cell walls can be a source of chitin, the overall yield may be lower compared to insect and crustacean sources. So, based solely on the amount of chitin that can be obtained from each source, the ranking would be crustacean shells[>] insect exoskeletons[>] fungal cell walls.

One way to compare the chitin content of various organisms is through chemical analysis of the chitin found in their exoskeletons or cell walls. This involves isolating the chitin from the tissues and quantifying it using techniques like Fourier Transform Infrared Spectroscopy (FTIR) or High-Performance Liquid Chromatography (HPLC) (Qin et al., 2019; Ghosh and Dhepe, 2021; Tsurkan et al., 2021). Another approach is to assess the thickness or density of chitinous structures in different organisms, as these traits can indicate the amount of chitin present. For instance, insects with thicker exoskeletons may have higher chitin content than those with thinner exoskeletons. Furthermore, genetic studies can help compare chitin content across organisms by examining the genes related to chitin synthesis and degradation. This allows researchers to understand how chitin production and turnover are regulated in various species. Combining chemical, morphological, and genetic analyses can comprehensively compare chitin content in different organisms, providing insights into the role of chitin in their biology (Kabalak et al., 2020; Khayrova et al., 2021; Lv et al., 2023). The chitin content in the shells of commercial organisms like shrimps, crabs, and crayfish is approximately 20%. In contrast, certain insect species have a chitin content of around 15%. Additionally, the chitin content in some aquatic insect species ranges from 10% to 20% (Table 5) (Susana Cortizo et al., 2008; Sagheer et al., 2009; Wang et al., 2013).

8 Impact of biotic and abiotic factors on chitin content and properties

Chitin content, purity, and structural properties differ among species, developmental stages, sexes, and body parts. Additionally, abiotic factors like processing methods and environmental and cultivation conditions influence the quality and quantity of chitin (Weiss, 2012; Chan et al., 2018; Hahn et al., 2020). Table 5 presents chitin yields across various insect species and other organisms. The data, organized by species, order, class, and yield, facilitates comparisons of chitin levels. This information is valuable for applications that utilize chitin due to its versatility in pharmaceuticals, biotechnology, agriculture, and materials science. Certain insects, such as *B. eri* (45%) and *H. illucens* puparium (59.9%), exhibit notably high chitin yields. These species may be prioritized for chitin extraction due to their superior yields, making them more efficient sources.

8.1 Animal *versus* non-animal sources of chitin

Animal and non-animal sources of chitin offer distinct content ranges, purity levels, and extraction requirements, impacting their applications and sustainability. Animal-derived chitin, present at higher concentrations (8%–64%) in shell structures, undergoes rigorous demineralization and deproteinization processes to remove non-chitin substances (Table 1). This high-purity chitin is widely applied in biomedical fields, such as drug delivery systems

TABLE 5 The amount of chitin in different species of insects and other organisms.

Class	Order	Species Chitin yield		References	
		Cryptotympana pustulata	36.6%	Sajomsang and Gonil (2010)	
		Rhodnius prolixus	14.1 µg/insect	Henriques et al. (2020)	
		Coridius nepalensis	44.0%	Sharbidre et al. (2021)	
		Coreus marginatus	14.5%		
		Lygaeus equestris	11.1%	Kaya et al. (2016a)	
		Pyrrhocoris apterus	10.6%		
	Hemiptera	Ranatra linearis 15.0%–16.0%		Kaya et al. (2014a)	
		Cicadatra platyptera	8.8		
		Cicada lodosi	5.0		
		Cicada mordoganensis	6.5		
		Cicadatra atra	6.7	Mol et al. (2018)	
		Cicadatra hyaline	5.5		
		Cicadivetta tibialis	5.8		
	Hymenoptera	Vespa crabro	8.3%		
		V. orientalis	6.4%	Kaya et al. (2015a)	
		V. germanica	11.9%		
Insecta		V. velutina	11.7%	Feás et al. (2020)	
		Bombus terrestris	6.9%	Majtán et al. (2007)	
		B. lapidaries	9.3%	Kava et al (2016b)	
		Formica clara	7.8%	Raya et al. (20100)	
		Apis mellifera	6.8%-13.2%	Kaya et al. (2015f)	
	Lepidoptera	Bombyx eri	45.0%	Zainol Abidin et al. (2020b)	
		Galleria mellonella	6.2%	Sáenz-Mendoza et al. (2018)	
		Ephestia kuehniella	9.5%-10.5%	Mehranian et al. (2017)	
		Argynnis pandora	8.0%-22.0%	Kaya et al. (2015c)	
		Anoplotrupes stercorosus	20.1%		
		Blaps tibialis	25.2%	Kaya et al. (2016b)	
		Cetonia aurata	18.2%		
	Coleontera	Geotrupes stercorarius	20.4%		
	Coleoptera	Cosmopolites sordidus	11.8%	Ssekatawa et al. (2021)	
		Holotrichia parallel	15.0%	Liu et al. (2012)	
		Catharsius molossus	24%	Ma et al. (2015)	
		Allomyrina dichotomy	10.5%-14.2%	Shin et al. (2019)	

(Continued on the following page)

Class	Order	Species	Chitin yield	References	
		Leptinotarsa decemlineata (Adults)	20.0%		
		L. decemlineata (Larvae)	7.0%	Kaya et al. (2014b)	
		Melolontha melolontha	13.0%-14.0%	Kaya et al. (2014c)	
		Calosoma rugosa	5.0%	Marei et al. (2016)	
		Cosmopolites sordidus	1.2%	Ssekatawa et al. (2021)	
		Tenebrio molitor	3.9%-8.4%		
		Zophobas morio	3.9%-8.4%	Shin et al. (2019)	
		Eupolyphaga sinensis	11.6%	Jiang et al. (2023)	
		Blaberus giganteus	21.2%-26.9%	Kaya et al. (2017b)	
	Blattaria	Blaptica dubia	3.8%-5.6%	Machado et al. (2024)	
		Blattella germanica	5.4%-6.2%	Deceries at al. (2010)	
		Periplaneta americana	8.4%-15.0%	Basseri et al. (2019)	
		Schistocerca gregaria	12.2%	Marei et al. (2016)	
			2.4% FW	V_{im} at al. (2017)	
		Gryllus bimaculatus	10.9% DW	Kiii et di. (2017)	
			5.1%	Chae et al. (2018)	
		Brachytrupes portentous	4.3%-7.1%	Ibitoye et al. (2018)	
		Dociostaurus maroccanus	12.0%-14.0%	Erdogan and Kaya (2016b)	
		Gryllotalpa gryllotalpa	9.8%-101%	Kabalak et al. (2020)	
		Acheta domesticus	5.7%-7.3%	Malm and Liceaga (2021)	
		Brachytrupes portentosus	4.3%-7.1%	Ibitoye et al. (2018)	
	Orthontoro	Ailopus simulatrix	5.3%	-	
		Ailopus strepens	7.4%		
		Duroniella fracta	5.7%		
		Duroniella laticornis	6.5%	Kaya et al. (2015d)	
		Oedipoda miniata	8.1%		
		Oedipoda caerulescens	8.9%		
		Pyrgomorpha cognata	6.6%		
		Celes variabilis			
		Decticus verrucivorus	A 704 11 804	Kava et al $(2015a)$	
		Melanogryllus desertus	4./%-11.8%	кауа et al. (2015e)	
		Paracyptera labiate			

TABLE 5 (Continued) The amount of chitin in different species of insects and other organisms.

(Continued on the following page)

Class Order Chitin yield References Calliptamus barbarus 20.5% Kaya et al. (2015b) Oedaleus decorus 16.5% Chrysomya megacephala 26.2% Abidin et al. (2020) 7.9% Drosophila melanogaster Kaya et al. (2016b) Calliphora vicina 8.1% Kaya et al. (2016c) Musca domestica Mohan et al. (2020) 8.0 Aedes aegypti $24.6\ \mu\text{g/insect}$ Henriques et al. (2020) Lutzomyia longipalpis 7.6 µg/insect Hermetia illucens (Larvae) 7.8-9.5 H. illucens (Prepupae) 9.1-10.9 H. illucens (Pupae) 10.3-10.7 Soetemans et al. (2020) Diptera H. illucens (Shedding) 23.7-31.1 H. illucens (Cocoon) 22.4-23.8 H. illucens (Flies) 5.6-8.4 10.0-13.0 H. illucens (Larvae) H. illucens (Pupal exuviae) 23.0-31.0 Pedrazzani et al. (2024) H. illucens (Adults) 6.0-9.0 59.90% H. illucens (puparium) Xiong et al. (2023) H. illucens (adult) 47.3% H. illucens Triunfo et al. (2022) 12.4% Various crustaceans 14%-25% Acosta et al. (1993) Crustacea Penaeus kerathurus 20.0% Hajji et al. (2014) 6.0%-7.0% Kaya et al. (2014a) Isopoda Oniscus asellus Malacostraca Amphipoda Gammarus argues 11.0%-12.0% Kaya et al. (2013) Farfantepenaeus aztecus 21.5% Abdou et al. (2008) Decapoda Litopenaeus vannamei 19.6% Liu et al. (2020) Aspergillus niger 10%-15% Teng et al. (2001) Eurotiomycetes Fungi Aspergillus flavus 13.2%-13.6% Blumenthal and Roseman (1957) Sordariomycetes Glomerella cingulata 3.8%-6.3% 8.0%-8.5% Geolycosa vultuosa Arachnida Aranea Abdou et al. (2008) 6.5%-7.0% Hogna radiata Mollusca Cephalopoda Loligo brevipenna 35.5 Soetemans et al. (2020)

TABLE 5 (Continued) The amount of chitin in different species of insects and other organisms.

FW, Fresh weight; DW, Dry weight. The bold values indicate the highest chitin yields.

and biodegradable materials, where quality and functionality are paramount. In contrast, non-animal sources, such as fungi, yield lower chitin content (8%-16%) and produce chitosan, a soluble chitin derivative with unique chemical properties. Fungal chitin extraction is less intensive, focusing primarily on residue removal, making it an eco-friendly alternative to animal chitin. The selection of a chitin source depends on specific application needs, sustainability considerations, and resource availability. While animal-derived chitin offers higher yields, concerns about environmental impact, such as overfishing, may limit its use. Fungal chitin, though yielding less chitin per source, is a more sustainable option, frequently employed in food and pharmaceutical industries due to its distinct biochemical profile. Thus, each chitin source has advantages and limitations, influencing its suitability across diverse industries. This revision provides improved clarity, consistency, and balanced comparison of the two chitin sources, emphasizing their unique benefits and environmental considerations.

8.2 Chitin content range by taxonomy

The dataset spans species across multiple taxonomic classes, including insects, fungi, crustaceans, and mollusks, allowing for a comprehensive cross-taxonomic comparison of chitin yields. Such diversity provides insights into environmentally and economically viable choices, as some species may be more sustainable or costeffective to cultivate and harvest. On a broad taxonomic scale, insects (Class Insecta) exhibit notable variability in chitin yield percentages, distinguishing them from other groups like fungi and crustaceans. Crustacean species such as Farfantepenaeus aztecus (21.5% chitin content) and specific life stages of the black soldier fly (H. illucens) show high chitin levels, making them strong candidates for highyield chitin extraction. Among insects, the orders Lepidoptera and Diptera are particularly promising, with species like B. eri (Lepidoptera), C. megacephala, and H. illucens (Diptera) achieving chitin yields as high as 59.9% in certain developmental stages, such as the puparium of H. illucens. This trend suggests that insects within these orders may serve as highly efficient sources for chitin harvesting. In addition, substantial variability exists within certain insect orders, such as Orthoptera, where chitin yields range from 1.2% in Cosmopolites sordidus to approximately 26.2% in Calliptamus barbarus, highlighting the extensive yield variation within a single order. Furthermore, the physicochemical properties of chitin differ significantly across taxa and are partially specific to each order. These differences manifest in surface morphology, which can vary even among species within the same order, suggesting that taxonomic and developmental factors influence chitin properties. This comprehensive taxonomic analysis underscores the potential for selecting species not only based on yield but also considering the physicochemical attributes of chitin, which may impact its functionality across different applications (Kaya et al., 2016a). However, findings indicate a correlation between chitin content, crystalline index, and taxonomical relationships. In this line, chitin was isolated from 16 arthropod species (13 insects and 3 arachnids) in a study. Among Insecta, chitin content was highest in Coleoptera (18.2%-25.2%), followed by Hemiptera, Odonata, Hymenoptera, Diptera, and Blattodea. Additionally, Coleoptera demonstrated higher crystalline index values compared to other orders (Kaya et al., 2016b). Among crustaceans, the composition of shrimp shells varies according to species, seasonal changes, and geographical locations. The components of shrimp shell waste consist of 10%-25% chitin, 13%-50% protein, 15%-70% minerals (Babu et al., 2008), and low-fat value (Cira et al., 2002). A study characterized the functional properties and microstructure of chitin extracted from the house cricket, Acheta domesticus, field cricket, Gryllus bimaculatus, and black soldier fly, H. illucens cocoons compared to commercial shrimp chitin. Various properties like solubility, emulsion capacity, and purity were determined. G. bimaculatus chitin showed high fat absorption and emulsion capacity, while H. illucens chitin had the highest deacetylation level. The highest values for purity were found in commercial chitin and G. bimaculatus, followed by H. illucens. Overall, insect chitin exhibited similar characteristics to commercial chitin, suggesting its suitability for industrial applications (Ndiritu et al., 2023).

8.3 Cross-classification comparison: insects *versus* other organisms

Insects vs. Crustaceans: The chitin yield in crustaceans, such as F. aztecus (21.5%) and Litopenaeus vannamei (19.6%), is relatively high and comparable to that of many high-yield insect species. Chitin structures from two European species, the common cockchafer, Melolontha melolontha, and Crustacea, Oniscus asellus, were isolated and compared. Both underwent similar isolation procedures using HCl and NaOH. Physicochemical properties of the chitins were analyzed, showing higher chitin content, purer composition, and more pores in M. melolontha compared to O. asellus (Kaya et al., 2014a). Various analyses confirmed similarities between the chitins, but differences in nanofiber adherence and pore structures were observed. M. melolontha was deemed a more appealing chitin source due to its purity (Kaya et al., 2014b). A study demonstrated that Holotrichia parallel (Coleoptera: Scarabaeidae) chitin was compared with commercial chitin from shrimp using various analytical techniques such as infrared spectroscopy, X-ray diffraction, scanning electron microscopy, and elemental analysis. Chitin from both sources exhibited comparable chemical structures and physicochemical properties (Liu et al., 2012). These indicate that crustaceans could serve as a viable alternative for chitin extraction, particularly in regions where they are abundant.

Mollusks and Fungi: The cephalopod *Loligo brevipenna* exhibits a fresh weight yield of 35.5%, which is significant; however, it is not directly comparable to the dry weight percentages reported for other organisms. In fungi, *A. niger* contains a chitin content of approximately 10%–15%, suggesting that fungi could serve as an alternative, although they may require more specialized processing methods.

8.4 Variation among body parts

Chitin extracted from various body parts of insects shows differences in content, surface morphologies, and degradation temperatures. The study investigated the correlation between chitin properties and honeybee body parts—head, thorax, abdomen, legs, and wings. The highest chitin content was found in the legs, while the lowest was in the thorax. Different surface morphologies were observed across body parts, with the abdomen showing the most variation. Maximum degradation temperatures varied among body parts, with the head chitin having the lowest value (Kaya et al., 2015f). A study investigates the physicochemical properties of chitin extracted from the butterfly species' wings and other body parts, *Argynnis pandora*. The findings revealed that the chitin content in the wings (22.0%) is significantly higher than that in other body parts (8.0%). Additionally, the wings exhibited a single surface morphology, while four distinct morphologies were observed in the other body parts. (Kaya et al., 2015c). However, the findings suggest a strong connection between the surface morphology of chitin and the specific body part from which it is isolated.

8.5 Variation among developmental stages and sexes

In certain species, such as the black soldier fly, H. illucens, chitin content varies significantly across different life stages (Larvae: 10.0%-13.0%, Prepupae: 9.1%-10.9%, Pupae: 10.3%-10.7%, Puparium: 59.9%, Adult: 47.3%) (Triunfo et al., 2022) (Henriques et al., 2020). The cocoon and puparium stages exhibit higher chitin percentages compared to adults. This variation suggests the potential for optimizing extraction processes by targeting specific life stages. Harvesting chitin at specific developmental stages, particularly from the puparium, could yield higher extraction rates, enhancing efficiency for large-scale chitin production. Chitin was extracted from different life stages of the mealworm beetle, Tenebrio molitor, and Oryctes rhinoceros with varying yields (Shin et al., 2019). Chitin and chitosan extracted from Colorado potato beetle, L. decemlineata were analyzed, revealing significant differences between larvae and adults. Adult beetles had a chitin content of 20% based on dry weight, while larvae had only 7.0%. Additionally, adults produced higher yields of chitosan, with 72.0% compared to 67.0% in larvae (Kaya et al., 2014c). Chitin was extracted from different life stages of the mealworm beetles (T. molitor, Zophobas morio) and the rhinoceros beetle (Allomyrina dichotoma) through processes of deproteinization and demineralization. The yields were as follows: 4.6%, 8.4%, and 3.9% from the larvae, adult, and superworm of the mealworm beetle, respectively; and 10.5%, 12.7%, and 14.2% from the larvae, pupa, and adult stages of the rhinoceros beetle (Shin et al., 2019). The chitin content of adult (14.0%) and nymph (12.0%) D. maroccanus, measured using the same method, was similar. The crystalline index values of chitin from adult and nymph grasshoppers were 71% and 74%, respectively (Erdogan and Kaya, 2016b). A study investigated the changes in the physicochemical properties of chitin in the insect Vespa crabro throughout its developmental stages: larva, pupa, and adult. The chitin content increased progressively with growth, reaching levels of 2.1%, 6.2%, and 10.3% at each stage, with a significant threefold increase occurring between the larva and pupa stages. Notably, the surface characteristics of chitin changed considerably as the insect matured (Kaya et al., 2016c)

In four grasshopper species, *Celes variabilis*, *Decticus verrucivorus*, *Melanogryllus desertus*, and *Paracyptera labiate*, males had higher chitin content than females, with the dry weight of chitin varying between species from 4.7% to 11.8% (Kaya et al., 2015e). A

recent study examined how gender affects the physicochemical properties of chitin isolates sourced from various body parts of the coleopteran species, Melolontha sp. The results revealed significant differences in chitin content, crystallinity, thermal stability, and surface morphology between samples extracted from male and female specimens across eight body parts, including antennae, head, and legs. Furthermore, interactions between the chitin samples and bovine serum albumin indicated potential differences in their applications based on the gender of the specimens (Kaya et al., 2016d). Testing a new method utilizing calcofluor fluorescent staining followed by microplate readings on disease vector species like Lutzomyia longipalpis, Aedes aegypti, and Rhodnius prolixus revealed significant variations in chitin levels, influenced by species, sex, and instar. For instance, in L. longipalpis females, the chitin content is approximately 12.0 µg per insect, while individual males have about 8.0 µg of chitin (Henriques et al., 2020).

8.6 Extraction methods

It should be noted that the chitin content of different sources may vary depending on the extraction and bleaching techniques employed. The chitin extraction process can significantly influence the final chitin content from a particular source. Extraction of chitin can utilize a range of methods, such as biological, chemical, and physical techniques. The primary methods employed in industry and medical fields are biological and chemical. However, these methods differ in their ability to target and eliminate unwanted components like proteins and minerals with varying degrees of effectiveness. Harsh chemicals may degrade some chitin along with impurities, resulting in reduced yield. Conversely, milder techniques could leave behind more proteins or minerals, thereby diluting the chitin content. Additionally, certain methods, particularly those involving high temperatures or strong chemicals, have the potential to degrade the chitin itself, leading to a diminished yield of usable chitin. Using different extraction methods revealed variations in chitin levels across developmental stages of the black soldier fly. Additionally, bleaching led to a notable decrease in chitin content at various developmental stages of the black soldier flies (Soetemans et al., 2020; Hahn et al., 2022).

The mineral content is a crucial factor influencing the efficiency and yield of chitin extraction from different natural sources. The mineral content in chitin-containing materials varies significantly depending on the source. For example, crab shells typically contain 20%-30% chitin, 30%-40% protein, and 30%-50% minerals (mainly calcium carbonate). However, this composition can differ based on factors such as species, season, and even the body part of the organism. Calcium carbonate is a common mineral found in chitin sources, but other minerals like sodium, potassium, and magnesium might also be present in smaller amounts. Insect cuticles have lower levels of inorganic material than crustacean shells, making their demineralization treatment more convenient and economical with milder chemicals. Animals with lower mineral content, such as insects, tend to have higher chitin yields and require milder extraction conditions compared to highly mineralized crustacean sources. The mineral content in insects can vary, with some sources like cicadas having around 10% minerals (Berezina, 2016; Abidin et al., 2020; Poerio et al., 2020; Terkula Iber et al., 2022; Triunfo et al., 2022).

9 Insects as a source of chitin

The amount of chitin in different species is presented in Table 5. Certain insects have less chitin than shrimp exoskeletons, but this is not universally true. For instance, B. eri, Coridius nepalensis, and P. cicadae have notably higher chitin levels at 45.0%, 44.0%, and 36.6% respectively (Figure 7). These amounts surpass the chitin content of shrimp, a significant source of commercial chitin and chitosan. It should be noted that in a fermentation process, the chitin yield of *H*. illucens puparium and adults was substantially higher (Xiong et al., 2023). Edible insects boast rich biodiversity and significant chitin content, positioning them as an underutilized asset (Ndiritu et al., 2023). Of the studied insects, the chitin content was highest in B. eri, followed by two hemipteran species (C. nepalensis, P. cicadae), and H. illucens. However, the black soldier fly, H. illucens, is an edible insect that can be easily raised under various conditions and is considered a potent source of chitin, even at the commercial level (Eggink and Dalsgaard, 2023; Pedrazzani et al., 2024). In this line, a study explored the potential of the black soldier fly (BSF) as a chitin source compared to shrimp shells. Different extraction methods were tested, and BSF chitin was found more challenging to extract and purify due to strong protein binding. The purity of BSF chitin ranged from 47.6% to 79.9%, lower than shrimp chitin at 88.3% (Pedrazzani et al., 2024). In another study, chitin was extracted from different developmental stages of the BSF, with varying chitin content. The physicochemical evaluation revealed that BSF chitin was a-chitin, similar to shrimp chitin (Soetemans et al., 2020). Research explores using black soldier fly puparium and adults with a continuous fermentation method employing specific bacteria for chitin extraction. Results show successful extraction of α -chitin with varying rates and yields based on the developmental stage of the flies. The study suggests that simultaneous continuous fermentation could offer a novel biological approach for chitin extraction from black soldier flies (Xiong et al., 2023). Comparing this research's results with others indicates the significant effect of the extraction method on chitin yield. However, the increasing demand for sustainable alternatives to crustaceans as a source of chitin and chitosan highlights bioconverter insects like H. illucens as potential sources. A study (Triunfo et al., 2022) emphasized the suitability of pupal exuviae for high chitin yields. Analysis shows that insect-derived polymers are similar to commercial ones, making them suitable for industrial and biomedical applications. The chitin from H. illucens is noted for its fibrillary nature, conducive to producing fibrous materials, while chitosan exhibits properties that could support antimicrobial applications (Triunfo et al., 2022). Researchers have found that the larvae of B. eri are a promising source of chitin (Huet et al., 2020). A study compared the chitin extracted from B. eri larvae to shrimp chitin. While both exhibited similar characteristics in terms of crystallographic structures and thermal stability, they differed in crystallinity and morphological structures. Furthermore, insect chitins treated with room-temperature ionic liquid showed higher digestibility when subjected to enzymatic degradation, making them a more promising source for N-acetyl-Dglucosamine production (Huet et al., 2020). Another study focused

on extracting chitin from the edible insect, C. nepalensis and compared it with commercial shrimp chitin. Results showed a chitin yield of 44.0% from C. nepalensis with a degree of acetylation of 57. 7%. The extracted chitin displays a mixture of chitin and chitosan, differing in crystalline structure and surface morphology from shrimp chitin. This research highlights the potential of C. nepalensis as a novel biomaterial source (Sharbidre et al., 2021). Research on the impact of various food wastes on H. illucens larvae revealed no significant differences in chitin and chitosan composition based on feeding substrates. The study underscores the importance of understanding how feeding substrates relate to valuable by-products, such as chitosan, in industrial insect production (Soetemans et al., 2020; Eggink and Dalsgaard, 2023; Xiong et al., 2023; Adamaki-Sotiraki et al., 2024; Pedrazzani et al., 2024). However, when choosing the best sources of chitin for extraction, several factors such as the amount of chitin, the purity, and the availability of its sources need to be considered. Although some insects can provide the highest amount of chitin, crustacean shells (such as those of crabs, shrimp, and krill) have been the traditional commercial sources due to their moderate but consistent yield of chitin. These shells are usually readily available from seafood processing industries. However, insects offer a potentially more sustainable option with a consistent supply, making them a promising source of high yield and purity of chitin. That being said, crustacean shells remain a good option due to their established processing techniques and easily available supply.

Insect chitin degrades more easily than shrimp chitin when treated with 6 N HCl and the enzyme chitinase. N-deacetylation of insect chitin is also simpler than that of crustaceous chitin. In a study, the enzymatic reaction of beetle and shrimp chitins was conducted heterogeneously in an acetate buffer solution. The hydrolysis rates of beetle and shrimp chitins by chitinase vary, with beetle chitin being hydrolyzed faster than shrimp chitin, suggesting that cockroach chitin has a higher affinity for chitinase than shrimp chitin (Zhang et al., 2000). The catechol compounds in insect cuticles inhibit chitin crystallinity, making the amorphous structure of insect chitin more easily destructible. N-deacetylation of insect chitin is simpler compared to crustaceous chitin, with approximately 94.0% of N-acetyl groups removed in a single treatment using 40% NaOH for 4 h at 110°C. Silkworm chitin, when treated with 2 N HCl under similar conditions, had 55% of its N-acetyl groups removed. Beetle chitin exhibited greater chitinase affinity than shrimp chitin (Zhang et al., 2000). The deacetylation process results in a copolymer of N-acetyl-glucosamines and glucosamines. When the resulting copolymer contains more than 50% N-acetyl-glucosamine units, it is typically referred to as chitin. For chitin polymers, the percentage of N-acetyl-glucosamine units is termed the degree of acetylation (DA) and can vary from 50% to 100% (Sivashankari and Prabaharan, 2017). A higher degree of acetylation reduces the solubility of chitin. Shrimp shells usually contain chitin with a degree of acetylation exceeding 50% (Sivashankari and Prabaharan, 2017; Yanat et al., 2023). This high acetylation level contributes to its lower solubility. Insects, however, offer chitin with superior characteristics. Studies indicate that chitin sourced from insects has a more favorable balance of deacetylation and acetylation than aquatic crustaceans (Kumar et al., 2020). While crustacean shells, such as those from shrimp, have long been the traditional and



reliable source of chitin due to established processing methods and consistent yields, insects-particularly species like H. illucens, B. eri, and C. nepalensis-are emerging as promising alternative sources. Insects offer several advantages, including higher chitin content in certain species, greater biodegradability, and potentially more efficient extraction methods, making them a sustainable and underutilized resource for chitin production. Furthermore, the distinct characteristics of insect chitin, such as its more favorable balance of deacetylation, suggest it may provide unique benefits in industrial and biomedical applications. However, factors such as purity, yield, and the availability of these insect sources must be considered when selecting the optimal chitin provider, as crustaceans still maintain an advantage in specific commercial contexts. Nevertheless, the growing demand for eco-friendly and sustainable alternatives positions insects as a viable future chitin and chitosan production source.

10 Conclusion

Chitin, the second most abundant polysaccharide on Earth, is a crucial resource with diverse applications in agriculture, medicine, and industry. Its sources, crustaceans, insects, fungi, and mollusks, each have distinct advantages and limitations. Crustaceans provide high chitin yield and superior crystallinity but present environmental and economic challenges due to their mineral content and seasonal availability. In contrast, insects serve as a sustainable alternative, offering high-purity chitin with lower environmental impact and minimal resource requirements, while also contributing to organic waste recycling. Fungal chitin, despite its lower purity and yield, holds promise for biomedical applications due to its consistent availability and cruelty-free sourcing. Mollusks provide unique mechanical and biocompatible properties, though in limited quantities. The physicochemical properties such as crystallinity, molecular weight, and degree of deacetylation vary among these sources, affecting their functionality for various applications. These insights emphasize the need for eco-friendly, cost-effective extraction methods tailored to each source to enhance chitin's utility. Additionally, the potential of insect-based chitin aligns with circular economy principles, advocating for its wider adoption. Future research should aim to refine extraction processes, improve chitin yield and purity, and investigate novel applications to fully utilize this biopolymer in a resource-constrained environment.

Author contributions

Conceptualization, HI: Data curation, Investigation, Methodology, Resources, Software, Validation, Visualization, Writing-original draft, Writing-review editing. and HA: Investigation, Resources, Writing-original draft. MB: Writing-review and editing.

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Conflict of interest

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Supplementary material

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