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A Comprehensive Study on Extraction and Characterization of Bioactive Pesticidal and Insecticidal Compound from Annona Squamosa (Custard Apple Seeds)

Shadab Shahanwaz Shaikh and Ankita Mhaske D. G. Tatkare Mahavidyalaya Mangaon, Raigad, Maharashtra, India

Abstract: The Insecticide and Pesticide is a substance which helps to kill the bugs and insects and helps to maintain the crop yield. There basically two types of pesticides are one is natural and second is synthetic. Due to industrialization and easily availability synthetic pesticides used more often. In this paper we will study about manufacturing of natural pesticide from custard apple seed. The manufacturing process based on distillation in which solvents are used acetone and hexane. Annona squamosa (custard apple or sugar apple), belonging to the Annonaceae family, is a small tree or shrub that grows natively in subtropical and tropical regions. Seeds of the custard apple have been employed in folk medicines because of the presence of bioactive chemicals/compounds such as alkaloids, flavonoids and phenolic compounds and acetogenins and cyclopeptides that are responsible for various biological activities. From investigations, it has been shown that the seeds of A. squamosa have considerable potential to be used as an antibacterial, hepatoprotective, antioxidant and antitumor/anticancer agent. Cyclosquamosin B, extracted from the custard apple seed, possesses vasorelaxant properties. Tocopherols and fatty acids, notably oleic acid and linoleic acid, are also found in the seed oil. Annona Squamosa seeds contain a high amount of annonaceous acetogenins compounds, which are potent mitochondrial complex I inhibitors and have high cytotoxicity. A survey primarily based on the nutritional, phytochemical and biological properties showed that A. squamosa seeds can be used for the discovery of novel products, including pharmaceutical drugs.

Keywords: Annona squamosa; Custard Apple Seed; Health Benefits; Bioactivities; Phytochemistry; Anticancer.

I. INTRODUCTION

Annona squamosa (Custard Apple or Sugar Apple). belongs to the Annonaceae family, a tropical fruit tree endemic to South and Central America, West Indies, Brazil, India, Egypt, Peru and Bermuda. In India, A. squamosa is widely cultivated in various states, including Assam, Uttar Pradesh, Bihar, Chhattisgarh, Maharashtra, Madhya Pradesh, Tamil Nadu, etc., for its edible fruit [1]. It is a small semi-deciduous branched tree or shrub that can reach up to 3–8 m in height and its fruits and seed by-product. Since ancient times, A. squamosa has been utilized in folk medicines and in various other applications involving food product development, e.g., the fruit pulp is used as a juice or as a flavoring agent. Custard apple industrial processing units generate large amounts of seeds, peels or seed coats [2]. The seeds are underutilized as the non-edible part of the fruit is discarded as waste, i.e., seed waste. The custard apple waste (seed) has a range of useful bioactive compounds [1,2,3]. Thus, seeds may potentially be extracted and may produce considerable income for the food processing industries. In India, the seeds have been used to make a hair tonic to remove headlice [1,3]. Ground seeds soaked in water have been used as an insecticide, a poison for fish, a strong eye irritant and a way to induce abortions [4,5,6]. Recent studies have shown that different parts of the plants, i.e., seeds, leaves, husks, peels and seed coats that are left after the main harvesting, are a rich source of phytochemicals and nutrients and can be utilized for novel product development, including usage in the food and pharmaceutical industries [1,7]. More than 400 active compounds have been isolated from A. squamosa [8]. In recent years, studies related to the

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pharmacological and phytochemical activities of A. squamosa seeds have confirmed that the major active chemical constituents are annonaceous acetogenins and cyclopeptides [9,10]. Annonaceous acetogenins, a class of polyketides, containing oxygenated functional groups including ketones, epoxides, hydroxyls, tetrahydropyrans and tetrahydrofurans, essentially found in the seeds, have been shown to have strong antibacterial, anti-ovulatory, antiinflammatory, antithyroidal and other properties [11,12,13]. In vivo studies show that seed extracts of A. squamosa are beneficial for treating liver cancer, prostate cancer, cervical cancer, pancreatic cancer, etc. [12,14]. The biological activities exhibited by Annona Squamosa seed extract are caused mainly because of phenolic compounds, alkaloids, peptides, amino acids, sterols, tannins, flavonoids, polysaccharides and tocopherols present in it [12,15,16]. Interestingly, annonaceous acetogenins extracted from custard apple seeds possess antitumor/anticancer activity [12,17]. These compounds proved to be cytotoxic against various cancer cell lines. For example, a volatile compound, namely bullatacin, isolated from the seed oil of A. squamosa is involved in antitumor activity [18]; the aqueous and organic seed extract of A. squamosa induced apoptosis of tumor cell death with the enhanced activity of caspase-3 and the down- regulation of antiapoptotic genes Bcl-2 and Bcl-xL when treated with organic seed extract and both seed extracts, respectively [11]; petroleum ether seed extract shows inhibition of keratinocyte (HaCaT cells) proliferation [19]. The seed extract of A. squamosa exhibits hepatoprotective activity, as it helps in lowering the increased levels of alkaline phosphatase (ALP), total bilirubin, serum glutamic pyruvic transaminase (SGPT) and serum glutamicoxaloacetic transaminase (SGOT) to normal levels [20].

1.1 Proximate Composition of Custard Apple Seeds:-

The fresh fruits of A. squamosa are commonly eaten in various regions of India, but the seed oil has not been reported yet for edible purposes. The seed of the custard apple is mainly composed of a seed coat (32.4%) and a seed kernel (67.7%). On a dry weight basis, the investigation shows a 22.2% content of crude fatty oil in seed kernels. The method of gas chromatography-mass spectroscopy (GC/MS) was used for studying the methyl esters of custard apple seed's fatty oil to determine its chemical composition, and results showed a total of 11 fatty acids, among which linoleic acid (22.9%), oleic acid (47.4%), palmitic acid (12.1%) and stearic acid (13.6%) were present in higher amounts. 11eicosanoic acid (0.2%), dihydro sterculic acid (0.1%), eicosanoic acid (0.9%), heneicosanoic acid (2.3%) and margaric acid (0.2%) were all found in lesser amounts in the oil. [21] These 11 fatty acids together constitute nearly 99.8% of the oil. 17-methyloctadecanoic acid (0.1%) and palmitoleic acid (0.01%) were both identified in traces or in a minimum amount. The high quantity of unsaturated fatty acids (UFAs) was also determined in further investigations of the oil. About 70.3% of the oil was contributed by linoleic acid (22.9%) and oleic acid (47.4%); similarly, 25.7% of the oil consisted of palmitic acid (12.1%) and stearic acid (13.6%) [21]. Mariod et al., [22] have demonstrated the presence of leucine, isoleucine, glutamic acid, phenylalanine-tyrosine, aspartic acid, serine, alanine, methionine-cystine, histidine, arginine, glycine, valine, threonine and lysine in the amounts of 0.845, 0.464, 0.995, 0.671, 0.684, 0.299, 0.594, 0.106, 0.139, 0.704, 0.392, 0.642, 0.324 and 0.407 g/100g protein, respectively [22]. A proximate composition analysis of A. squamosa seeds demonstrated the presence of carbohydrate, fat, fiber, ash, protein and moisture at concentrations of 66.64, 29.21, 32.64, 1.90, 2.25 and 3.92 g/100g dry weight (DW) (%), respectively [23]. On the other hand, minerals (in mg/kg) such as K (56.47%) and Ca (46.90%) are present in higher amounts compared with P (33.30%), Mg (20.36%), Fe (6.74%), Cu (0.30%), Na (9.29%), Zn (0.43%) and Mn (0.25%).[22] By comparing the results of Shehata et al., [23] with other reference studies [24,25], it was concluded that the presence of a higher Ca content is important for healthy teeth and bones, while Fe is essential for preventing anemia. Minerals are also responsible for maintaining pH levels and blood pressure in the human body. [26]

II. LITERATURE REVIEW

2.1 Phytochemical Profile of Custard Apple Seeds:-

Phytochemical investigations reported cyclopeptides and annonaceous acetogenins as the chief constituents in the seeds of Annona squamosa. Different parts of A. squamosa contain several phytochemicals involving alkaloids, such as aporphine, norcorydine, roemerine, corydine, glaucine, anonaine and norisocorydine, in different parts of the plant. The seeds of Annona squamosa were found to contain acetogenins (polyketide), namely annotemoyin–1 and 2, cholesteryl,

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coumarinoligans, glucopyranoside, squamocin, and squamocins B-N. It was demonstrated that the custard apple seeds are toxic, but they are used as a biopesticide or an insecticide (its preparation may cause eye irritation that results in damage to the cornea). Custard apple seeds contain a high oil content and can be used for the production of soap and/or, if treated, can be used as an alternative to cooking oil. Seeds contain volatile substances such as 12,15-cis-squamostatin-A, bullatacin, β -caryophyllene, α - pinene, β -pinene, anonaine, camphene, spathulenol, germacrene, squamocin, duvariamicin-III, myrcene, liriodenine, annonacin and molvizarin.

In a study, the identification of phytochemicals present in the seed extract of A. squamosa was carried out via Fouriertransform infrared (FTIR) analysis. The result of the investigation shows the presence of alkenes, imine, oxime, quinone or conjugated ketone, nitro compounds, amides, nitroso compounds, sulfone, aromatics, sulphate ester, alkyl halides, phosphine, ethers, phosphonate, trimethylsilyl, amine oxide, phosphor amide, carboxylic acids, thiocarbonyl esters, phosphine oxide, phosphate, organosilicon, phosphite esters and amines in the methanolic seed extract of Annona squamosa.

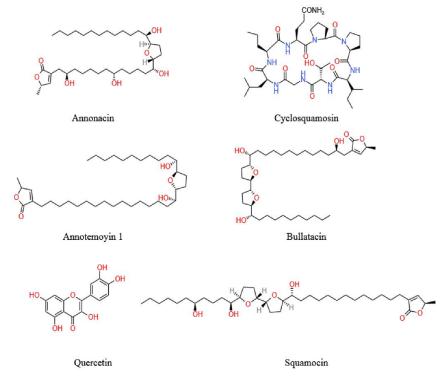


Figure 1. Structure of important compounds found in seeds of A. squamosa.

2.2 Antidiabetic Activity:-

Diabetes is one of the common endocrine disorders, characterized by altered carbohydrate, insulin and protein metabolism as a consequence of pancreatic insulin deficiency or insulin dysfunction. According to the reports of the World Health Organization (WHO), 80–90% of people above 40 years old are more prone to non-insulin-dependent diabetes mellitus. In a study, the ethanolic and methanolic seed extract of A. squamosa was administered to alloxan-induced diabetic rats (150 mg/kg body weight (BW)) to check its effect on blood glucose levels in diabetic rats. The ethanolic (dose: 200 mg/kg) and the methanolic (dose: 200 mg/kg) seed extract of A. squamosa exhibited significant dose-dependent 43.96% and 45.99% antihyperglycemic activity, respectively. From the results, it was observed that the ethanolic extract was less effective than the standard used (glibenclamide) for hyperglycemic activity. Compounds such as saponins, flavonoids, acetogenins, phenolic compounds and alkaloids are known to be active antidiabetic agents. The antidiabetic property of the methanolic and ethanolic seed extract of the apple may be due to the presence of more than

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one antihyperglycemic agent mentioned above. Further, the action mechanism will be explained by pharmacological and biological studies that help in presenting the seeds of Annona Squamosa as therapeutic agents in antidiabetic research. However, there are very limited studies investigating the antihyperglycemic effect of Annona Squamosa seeds. It has been concluded that there is a need to conduct investigations in the same field.

2.3 Anti-Inflammatory

Inflammation is the human body's defensive system, regulated by pro- and anti-inflammatory mediators (chemokines, cytokines, etc.). Many factors may cause inflammation in the human body, including exposure to allergens, physical trauma, thermal or chemical stimuli and microbial infection. However, inflammatory disorders such as chronic asthma, rheumatoid arthritis, inflammatory bowel disease and multiple sclerosis may be caused by internal or external factors that disrupt anti-inflammatory mediators. For instance, the seed extract of Annona squamosa causes a decrease in IL-6 and TNF- α levels in the lipopolysaccharide (LPS)- stimulated macrophage J774A.1 cell line. It was shown that two parallel synthesized cyclic cyclopeptides extracted from the seeds of the custard apple, i.e., cyclosquamosin D and cyclopeptide B, were demonstrated to have anti-inflammatory actions by inhibiting the generation/production of IL-6 and TNF- α . From the results, it was concluded that cyclopeptides have strong anti-inflammatory effects, reducing the levels of IL-6 and TNF- α in the bloodstream (~25%), with an IC50 value of 1.22 and 9.2 μ M, which is significantly higher than observed with natural products or natural cyclic peptides. Similarly, in another study, cyclosquamosin D inhibited the generation of pro-inflammatory cytokines in Pam3Cys- stimulated and LPS-stimulated J774A.1 macrophages in a dose-dependent manner. For TNF- α a 60–20%, whereas for IL-6 a 50–10% reduction was observed at a dosage of 5–50 µg/mL, respectively. From the findings of different studies, it was deduced that cyclic peptides extracted from custard apple seeds might be utilized as an anti-inflammatory agent, though further investigation is needed for their anti-inflammatory effect.

2.4 Toxicity of Annona squamosa Seeds

Annona Squamosa seeds have been employed in the traditional medical system since time immemorial for skin exfoliation and elimination of headlice. The seeds of the custard apple include biologically active substances such as polyphenols, alkaloids, acetogenins and cyclohexapeptides. The pharmacological properties of certain acetogenins, including wound healing capabilities, anti-lice, mosquitocidal characteristics, anticancer, antifungal and antioxidant properties have been investigated in the past few years. Custard apple seeds are toxic mainly due to the presence of a high amount of annonaceous acetogenins (neurotoxins) that are stated to cause irritation in mucosa and the eye and vomiting (taken orally). Safety concerns are stated concerning the use of the plant in dietary supplements and is cited in the poisonous plants database of the Agence Francaise de Securite Sanitaire des Aliments (AFSSA) and the American Food and Drug Administration (FDA). Others have claimed that when crushed seeds come into contact with the eyes, they may cause conjunctival irritation that ultimately leads to eye ulcers. When a toxic extract of the custard apple seed was tested on rat eyes, it resulted in a conjunctival infection and delayed damage to the corneal epithelium. Some examples show that this plant's seeds induce severe symptoms of toxic keratitis after inadvertent contact with the custard apple seed. For instance, a patient utilized the oil combined with A. squamosa seed powder for the control of headlice. Results demonstrated that the patient exhibited considerable conjunctival congestion, blepharospasm, coarse punctate epithelial diffuse erosions and dense stroma and no anterior chamber reaction was seen in either of the patient's eyes. Individuals in a different study experienced a similar pattern after being exposed to custard apple seeds for 8 to 12 h. Slit-lamp examination revealed widespread erosions with coarse punctate epithelium in both eyes. Furthermore, it is concluded that the seeds or the seed extract/powder of the custard apple are highly toxic and an irritant, causing conjunctivitis and corneal epithelium damage with a high risk of secondary infection. However, people need to learn about the toxicity of the seeds of the custard apple, which requires further investigations to acquire the knowledge of its safety aspects and dosage.

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2.5 Anticancer/Antitumor Activity

Cancer is a genetic disorder, caused by the mutations that happen to take control of genes in our body and control how cell functions, grow, multiply and die. The reports of Cancer Research UK estimate that there are more than eight million cancer-related deaths worldwide per year and this may increase in the future. In a study conducted by Chen et al., the in vivo and in vitro antitumor activity of acetogenins isolated from the custard apple seed oil was investigated against human tumor cell lines. Two major acetogenins isolated from the seed oil of A. squamosa, i.e., 12, 15-cissquamostatin-A (47.98 mg/g) and bullatacin (256.18 mg/g), were detected and quantified by high-performance liquid chromatography (HPLC). The result of the study indicates that seed oil shows considerable antitumor properties against A-549, Hela, MCF-7 and HepG2, especially for Hep G2 (IC50: 0.36 mg/mL) and MCF-7 (IC50: 0.25 mg/mL) cells in vitro. Furthermore, the oral treatment of custard apple seed oil also prevents growth of H22 tumor cell lines in mice, with a reduction rate of 69.55% with no post-treatment side effects, suggesting that the seeds of the custard apple may be used as a potent ingredient for the production of anticancer drugs. Similarly, the seed oil (dose: 0.5–1.0 mL/Kg) of the custard apple shows considerable antitumor activity in H22 xenograft-bearing mice, with an inhibitory rate of 53.54% against the development of H22 cell lines. It was found that seed oil shows antitumor effects by inhibiting the interleukin-6/Jak/Stat3 signaling pathway by reducing the production of interleukin-6, Janus kinase and activators of transcription (p-Stat3) and phosphorylated signal transducer expressions. The findings of the investigation revealed that the ASSO-NPs group (15 mg/kg) has the highest tumor growth inhibitory rate of 69.8%, significantly greater than the free seed oil group (135 mg/kg, 52.7%, p < 0.05) in a 4T1 tumor-bearing mice model [64]. In addition to antitumor activity, it was noticed that there was no significant change in the weight of the mice, indicating that ASSO-NPs have good safety.

III. APPLICATIONS OF CUSTARD APPLE SEEDS

3.1 Nutritional Composition:-

Annona squamosa is a lowland tropical shrub that possesses a high pharmaceutical potential for treating cardiac ailments, thyroid-related disorders, diabetes, and cancer. Phytochemical analysis of ASL extracts revealed the presence of numerous phytochemicals, such as proteins, carbohydrates, saponins, alkaloids, flavonoids, phenolics, and glycosides]. A study conducted at four different sites in Egypt showed the highest protein content in ASLs compared to seeds and fruit. ASLs from Menofia showed a protein content of 13.47 mg/g on a fresh weight (FW) basis, whereas ASLs from Mansoura demonstrated the highest protein content of 17.26 mg/g FW. ASLs from Alexandria (3.52 mg/g FW) and Giza (6.80 mg/g FW) showed the lowest protein content. Thus, a higher protein content in ASL. extracts can harness the nutritional value of the food for use by humans as well as animals. Proteins and amino acids were also present in high quantities in the methanolic and aqueous extracts of ASLs.

3.2 Nutritive value of custard apple:-

Custard apples are usually consumed as dessert fruit. The nutritive value of custard apple per 100 g of pulp is presented in table 1. The fruit is rich in starch when firm but increases markedly in sugar as it softens. The main sugars are glucose and fructose (80-90%). The calorific value is high (300-450 kJ per 100 g) and is almost doublethat of peach, orange and apple. Custard apple is full of vitamin C anti-oxidants, which helps to combat many diseases and also enhances the immune system. It is abundant source of dietary fibre, Vitamin A. Vitamin C. Antioxidant, Potassium, magnesium and also contains calcium. Vitamin B6, Copper and Low fat levels, excellent source of fron. It has high calorific value, able to provide sustained energy and delicious in nature. It has about 3.1% of fibre in the edible portion.

3.3 Medicinal value of custard apple:-

The health and medicinal benefits of the Custard apple fruit are numerous and it appears to possess potent bioactive principles in most of its plant parts (fruit, seed and leaves). The various chemical constituents isolated from leaves, stems, and roots of the plant include anonaine, aporphine, coryeline, isocorydine, norcorydine, and glaucine For centuries, Ayurvedic practitioners in India haveextensively use various parts of the sugar apple (Annona squamosa) tree for the management of diabetes. Essential oils, pinenes have also been described and extracted from custard apple. One

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clas of chemicals which sets custard apple apart from other fruit species is the presence of acetogenins The acetogenins are unique to the Annonaceous family, which are very long chain fatty acids, and only found in Annonaceous species. The in vitro and in vivo studies appearto have considerable anti-cancer properties and anti- hypertensive properties The anti-cancer properties of custard apple appear to be mainly due to a class of compounds called acetogenins which are specific to Annonaceaous species. Acetogenins have been tested in vitro against 60 types of cancer cells, including breast, prostate and colon. Compared with paclitaxel a standard anti-cancer drug. pullatacin, an acetogenin, was 300 times as potent even at in vivo test system. Custard apple has been used for treatment of malaria. Three known aporphine alkaloids were solared apple thebark. Structures of compounds were identified as N-Nitrosoxylopine, Roemerolidine and Dugurvalline. All the three anti-malarial alkaloids exhibit moderate activity against chloroquine sensitive strain (D10) and a chloroquine resistan strain (Dd2) of Plasmodium falciparum.

3.4 Pharmacological Properties:-

Recently, custard apple seeds have emerged as a potential ingredient for the development of supplementary foods because of its significant nutraceutical and phytochemical composition. However, the development of by-products by integrating the bioactive compounds from the custard apple seeds are well-endowed with novel pharmacological properties. These properties of A. squamosa or the custard apple seed have been extensively studied for the antimicrobial, antidiabetic, anti-inflammatory, anticancer, antitumor, antioxidant, hepatoprotective, antiproliferative, antiheadlice, antihelminthic and antilarval activities.

IV. CONCLUSION

Custard Apple (Annona Squamosa) has gained popularity due to a recent increase in investigations/studies conducted on the health benefit and the bioactivities of different parts of the plant such as the seed parts, the bark, the leaves, the fruits, etc. A. squamosa has been employed in indigenous folk medicine worldwide and likely used in the food industry, as 50– 80% of the fruit is edible. The pulp is used as a flavoring agent in ice cream and contains vitamin B1 (thiamine), dietary fiber, potassium and sodium in considerable amounts. The seeds of Annona Squamosa are a rich source of phytochemicals such as polyketides, annonaceous acetogenins (neurotoxins), cyclopeptides, carbohydrates, proteins, lipids, oleic acid and linoleic acid. Based on in vivo and in vitro experiments Annona Squamosa seed extracts were found to be effective in various bioactivities such as antitumor, antimicrobial, antifungal, antidiabetic, hepatoprotective, anticancer and antifertility. The production of natural insecticides from custard apple seeds holds great potential to revolutionize agriculture by offering sustainable, eco-friendly, and cost-effective pest management solutions. Future advancements in technology, policy support, and market demand can further enhance its applicability and impact globally. A few studies were available on the phytochemical profile and the molecular mechanism of various bioactivities of A. squamosa seeds.

However, more pharmacological studies need to be performed to determine the nutraceutical and food supplement potential of the seeds. Based on the studies available, Annona Squamosa seeds may likely be used as an ingredient in the nutraceutical and food/nutrition industry, especially as anticancer drugs and antitumoral dietary supplements, benefiting human health. We can conclude that the natural pesticide derived from custard apple seed oil is effective, cost- effective, and easy to manage. It recovers to about 19- 20% while using hexane as a solvent. If we use acetone as a solvent then we get oil yield is about 12-15 % yield of oil. Hexane recovered during extraction is around 80-85% as well as acetone recovered during extraction is around 55-60%. Without putting forth a lot of work, this pesticide substance may be made easily available to everyone in India. This raw material will be very inexpensive, lowering the overall processing and solvent recovery costs.

In conclusion, the natural pesticide formulated from custard apple seed oil is an effective and environmentally friendly solution for pest control. The use of natural oils like acetogenins from custard apple seeds offers a sustainable alternative to chemical pesticides, which can have harmful environmental and health effects. Further studies could explore the long-term effectiveness of the pesticide and its impact on non-target organisms, as well as investigate the optimization of extraction and formulation methods to maximize efficacy and reduce costs.

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V. FUTURE SCOPE

The production of natural insecticides and pesticides from custard apple seeds offers promising potential in sustainable agriculture and pest management. Below are the key areas of future scope:

5.1. Commercialization and Market Expansion:-

Growing Demand for Organic Farming: With the increasing demand for organic produce, natural insecticides can cater to farmers looking for eco-friendly alternatives. Development of Biopesticide Brands: Large-scale production can lead to branded, eco-friendly pesticide products, catering to both domestic and international markets. Cost-effective Solution: Utilizing custard apple seeds, often considered waste, reduces raw material costs, making the product more accessible to farmers.

5.2. Research and Development:-

Optimization of Extraction Techniques: Advanced methods like supercritical fluid extraction can improve the yield and potency of active compounds. Formulation Improvements: Researchers can develop more stable, long-lasting formulations suitable for diverse crops and climates. Toxicity Studies: Comprehensive studies can help ensure the safety of these products for humans, non-target organisms, and the environment. Active Compound Identification: Further research can isolate and enhance the insecticidal properties of specific bioactive compounds like acetogenins.

5.3. Integration with Modern Agriculture:-

Precision Agriculture: Combining natural pesticides with modern technologies (e.g., drones for spraying) can increase efficiency and reduce waste. Compatibility with Other Practices: Testing and integrating custard apple seed pesticides with Integrated Pest Management (IPM) programs can offer holistic pest control solutions.

5.4. Environmental Impact:-

Reduction of Chemical Pesticides: Scaling up production can significantly reduce reliance on harmful synthetic chemicals, promoting sustainable agriculture. Biodegradability: Unlike synthetic pesticides, these products decompose naturally, reducing soil and water contamination. Preservation of Beneficial Insects: Natural pesticides often have selective toxicity, sparing pollinators and other beneficial organisms.

5.5. Waste Management and Circular Economy:-

Utilization of Agricultural Waste: Custard apple seeds, typically discarded as waste, can be repurposed, promoting a zero-waste approach. Economic Opportunities: The process creates new avenues for employment and entrepreneurship in rural and urban areas.

5.6. Policy and Government Support:-

Governments and environmental organizations can promote the use of bio-pesticides through subsidies, awareness campaigns, and certifications for natural products.

5.7. Global Collaboration:-

Partnerships between research institutes, agricultural organizations, and private industries can drive innovation and expand the use of custard apple-based insecticides to international markets.

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