2

Review of Literature

2.1 General

A review of the literature supports carrying out of research work in a systematic manner, and it provides detailed information about selected work-related studies, which helps to get a novel research idea. In this section, the detailed reviews on plant-mediated metal, non-metallic nanoparticle synthesis and their various applications are discussed. The reviews on selected samples, hair removal techniques and different types of skin substitutes are also documented in this section.

2.2 Reviews on *Cyperus rotundus*, Pumice stone and *Hemigraphis alternata*

2.2.1 Reviews on *Cyperus rotundus*

Purple nutsedge is an Indian perennial worst weed all over the world **(Ameena *et al*., 2014)**. Due to the long propagation network, the high content of starch is stored in rhizomes **(Fischer *et al*.,1995)**. Flavonoids such as catechin, quercetin, kaempferol, and myricetin are widely used in Ayurvedic and Pharmaceutical industries. In tissue culture, the flavonoid content of the rhizome is 1.96 and 0.28 mg/g.d.w in 6 and 3 week old callus tissues. Leaves contain 0.58 and 0.48 mg/g.d.w in free and bound form respectively. In root contains 0.19 and 0.11 mg/g.d.w in free and bound form respectively **(Krishna and Renu, 2013)**.

Over 90 compounds from the essential oil composition were identified. The plant contains a pleasant and characteristic odour due to the aroma producing constituents (Octane compounds-limonene, α-terpineol, valencene and nootkatone; Monoterpenes-Trans-pinocarveol, pinocarvone, trans-verbenol, 1,8-cineole, myrtenal and verbenone; Sesquiterpenes-α-copaene, caryophyllene oxide and humulene oxide) present in the essential oil of *C. rotundus* **(Jirovetz *et al*., 2004)**.

Nidugala *et al*., 2015, reviewed the isolated compounds from *Cyperus rotundus* L. (**Table 1**). It is commonly known as traditional medicine for many diseases, inflammatory disorders and lymphoma cells cytotoxicity **(Licea *et al*., 2012)**. Hentriacontane possesses anti-inflammatory activity **(Kim *et al*., 2011)**. N-triacontane shows antimicrobial and anticancer activity **(Chakraborty *et al*., 2012)**.

The anticancer, anti-diabetic, antiangiogenic and inflammatory activity was observed due to gamma sitosterol **(Raman *et al*., 2012)**. The *C. rotundus* extract is an excellent cancer therapeutic agent **(Lawal and Oyedeji, 2009)**. The antimicrobial inhibition activity of oil (%) and phytoconstituents mainly depends on where the plant grows. It depends on the soil, rainfall, plants nutrition, environmental and climatic conditions, *etc*., **(Ali Abdella and Hajar, 2014)**.

Table 1. Compounds isolated from *Cyperus rotundus* tuber

S.No	Compounds	References
1	Cyperene, α-copaene, Trans-pinocarveol, Valerenal, Caryophyllene oxide, Nootkatone, Valencene	**Jirovetz *et al*., 2004**
2	Azulene, Cyperene, Humulen, Campholenic aldehyde, Terpineol, α-Selinene, β-Vatirenene, Isogermacrene D, Calacorene, Limonene, β- Selinene, zierone, Isolongifolene, α-Pinene, Myrtenal, Copaene	**Suneet and Tiwari, 2014**
3	Phytol, 1-(2-hydroxy-5-methylphenyl), 9, 12, 15-Octadecatrienoic acid, Ethyl ester, Hexadecanoic acid, 3,7,11,15-Tetramethyl-2 hexadecan-1-ol, n-Hexadecanoic acid, D-Allose, Ethanone, 4H-Pyron-4-one, Caryophyllene oxide	**Elezabeth and Arumugam, 2014**
4	Hentriacontane, Pentatriacontane, Heptadecane, Eicosane, Octacosane, Octadecane, α-Cyperone, Nonacosane	**Nidugala *et al*., 2015**
5	Caryophyllene oxide, β-pinene, α-selinene, α-Cyperone, Myrtenal	**Lawal and Oyedeji, 2009**
6	Camphenol, α-Pinene, Camphene, Carveol 2, Thymol, Myrtenal, D-Limonene, Copaene, α-Cyperone	**Ali Abdella and Hajar, 2014**
7	1,6-dimethyl-4-(1-methylethyl), Naphthalene, Caryophyllene	**Ilham *et al*., 2018**

Four chemotypes (H, K, M and O) of essential oils of *C. rotundus* obtained from different regions of the Asian continent have been identified is given in **Table 2 (Sivapalan, 2013)**. This is due to the geographical location of *C. rotundus*.

Table 2. Categorized compounds of essential oils of *C. rotundus* in different region of Asia

Types	Country	Compounds	Percentage (%)
H	Japan	α – cyperone	36.6
		ß-selinene	18.5
		Cyperol	7.4
		Caryophyllene	6.2
M	China (Vietnam and Hong Kong)	α -cyperone	30.7
		Cyperotundone	19.4
		ß-selinene	17.8
		Cyperene	7.2
		Cyperol	5.6
O	Japan, Taiwan, Thailand, Hawaii, Philippines	Cyperene	30.8
		Cyperotundone	13.1
		ß-elemene	5.2
	Hawaiian	Cyperene	20.7
		Cyperotundone	25
K	Hawaii	Cyperene	28.7
		Cyperotundone	8.8
		Patchoulenyl acetate	8
		Sugeonyl acetate	6.9

2.2.1.1 Pharmacological application of *Cyperus rotundus*

The extract of *Cyperus rotundus* produces a considerable antidepressant activity in rats than fluoxetine (positive drug) in a dose-dependent manner **(Hao Gui *et al*., 2017)**. Also, the hexane extract of the tuber portion of *Cyperus rotundus* showed efficient mosquito repellence at a low dose. The highest dose of tuber extract (10%), suggesting complete repellence (100%). It can be used as a mosquito repellent against mosquito bites **(Singh *et al*., 2009)**. *Cyperus rotundus* rhizomes ethanol extract significantly reduced the level of triglyceride in test animals. *C. rotundus* rhizomes extract exhibits antioxidant activity by lipid-peroxidation **(Jeyasheela *et al*., 2011)**.

C. rotundus rhizomes is an analgesic herb for treating child stomach ache and used as folk medicine products in Turkey. Sesquiterpenoid hydrocarbons are obtained from South India, Turkey, Chine and Tunisian. Sesquiterpenoid (α-Copaene and Cyperene) content in *C. rotundus* tuber may be helps in allelopathic activity **(Ilham *et al*., 2018)**.

Cyperus rotundus (CR) oil is an efficient and healthy hair growth reducing agent. Flavonoids in oil on androgenic hair have anti-androgenic activity. The combination of CR oil and the Alexandrite laser showed no side effects and effective for reducing the growth of hair (white and black) **(Ghada, 2014)**. The Egyptian *Cyperus rotundus* essential oil is an active agent in axillary hair and hirsutism removal without affecting serum testosterone. *C. rotundus* essential oil was compared with 0.9% saline on androgenic hair. The existence of androgen (Androgenic hair) depends entirely on sexual hair growth **(Evans *et al*., 1995)**. This oil significantly decreases the growth of white hair, and it is an easy, cheap (1 dollar/month), and harmless treatment (No cases were reported on infection, irritation and itching). The existence of flavonoids in CR oil promotes anti-androgenic activity. It is an effective treatment method for moderate levels of androgenic hair **(El-kaream, 2012)**.

Cyperus rotundus rhizomes ethanol extract is used in the treatment of epilepsy. The presence of flavonoids in this extract shows anticonvulsant activity **(Shivakumar *et al*., 2009)**. Ethanol extract of *Cyperus rotundus* showed significant dose-dependent anti-angiogenic and antioxidant activity due to the phytoconstituents present in extract **(Amjed *et al*., 2018)**.

Antioxidant activity and antimicrobial activity were tested with whole essential oil and its fractions. The whole essential oil is very effective than its fractions, and it is highly inhibiting *Staphylococcus aureus*. The fractionate of sesquiterpene components exhibits moderate antioxidant and antimicrobial activity. The activity is due to the synergy among different oil constituents **(Ismahen *et al*., 2014)**. The ethanolic extract showed antidiabetic activity in STZ induced Swiss mice. *Cyperus rotundus* has improved the weight of mice and it showed antidiabetic activity by reducing SGPT, SGOT, cholesterol, and triglyceride levels **(Pradeep *et al*., 2019)**.

The ethanol extract of *Cyperus rotundus* L. rhizomes was tested against joint pain. The test was carried out in male albino rats and the penetration of compounds through the mouse skin was noticed in a liquid chromatogram. A decrease in joint pain with 7% extract concentration was observed **(Rahim *et al*., 2016)**. The anti-ulcer activity was observed 41.2% at the dosage of crude extract of *Cyperus rotundus* (500 mg/kg). The crude extract showed an anti-inflammatory and lesser muscle relaxant effect **(Ahmad *et al*., 2014)**.

The growth, acid production, adhesion, and water-insoluble glucan synthesis of *S. mutans* is affected by *Cyperus rotundus* tuber extract. In a dose-dependent manner, the acid production and growth of *S. mutans* decreased, and the complete growth inhibition was noticed in 4 mg/mL **(Yu Hyeon *et al*., 2007)**. *Cyperus rotundus* exhibited antimicrobial inhibition on *Escherichia coli*, *Candida albicans*, *Pseudomonas aeruginosa* and *Bacillus subtilis* **(Wangila, 2017)**. The mixture of zerumbone and 1,8-cineole compounds of rhizome showed repellent activity in male *B. germanica* within 24h than an individual compound **(Chang *et al*., 2017)**.

The rhizomes essential oil exhibited antimicrobial inhibition on *Bacillus subtilis*, *Bacillus pumilus*, *Pseudomonas aeruginosa*, *Shigella flexneri*, *Aspergillus niger* and *Candida albicans*. Benzene extract of rhizome showed antibacterial activity against *S. flexneri*, *P. aeruginosa*, *B. subtilis* and it also showed antifungal activity against *C. albicans* and *A. niger*. The $CHCl_3$ extract revealed antibacterial inhibition on *B. pumilus* only **(Vijender *et al*., 2018)**. Silver nanoparticles were biosynthesized using the tuber portion of *Cyperus rotundus* in 30 min. In UV-Visible spectroscopy, the SPR peak was observed at 400 nm. DLS pattern reveals the Zeta average diameter of 122.3 nm with a polydispersity index (PDI) of 0.439. The nanoparticles revealed antibacterial inhibition on *Bacillus thuringiensis* and *Staphylococcus aureus* **(Mounika *et al*., 2017)**.

2.2.2 Reviews on pharmacological activities of *Hemigraphis alternata*

Hemigraphis alternata is a tropical and medicinal species that belongs to Acanthaceae. It is an indoor and outdoor perennial plant and is commonly called an Aluminium plant, Red flame Ivy **(Nair and Nair, 2018)**. The ethanolic, chloroform and benzene extract contain essential metabolites. The dry leaves contain 4.5% moisture and 12.5% ash. The moisture loss and ash value determined the quality and purity of the drug. The plant sample contains carbohydrates (28.12 mg), protein (5.6 mg) and starch (25.30 mg). It can potentially treat a fresh wound, inflammations, skin-related complaints, and ulcers **(Reshma *et al*., 2018)**.

Silver nanoparticles were prepared using the aqueous extract of *Hemigraphis alternata* and the study also suggests the phytoconstituents of leaves to be useful in the medical and cosmetics industries **(Priyanka *et al*., 2016)**. The silver nanoparticle formation using the leaves of *Hemigraphis colorata* aqueous extract is reported to be due to the reduction of silver salt by the electron-donating ability of higher phenolic contents in the extract. The prepared nanoparticles showed significant anti-inflammatory activity **(Thomas and Mathew, 2019)**.

The extract of *Hemigraphis alternata* was incorporated in the chitosan scaffold. Synthesized scaffold showed antibacterial activity against *E. coli* (45-folds colony reduction) and *S. aureus* (25-fold colony reduction). The cell viability showed 90% on Human Dermal Fibroblast cells than chitosan. The prepared scaffold has high hemostatic and antibacterial activity, and it can be effectively applied for infectious wounds **(Annapoorna *et al*., 2013)**.

The wound healing activity of *Hemigraphis alternata* was tested on fibroblasts and endothelial cells. The proliferation and migration of fibroblast were tested using aqueous and ethanol extract of *Hemigraphis alternata* by MTT and scratch-wound healing assay. The aqueous extract (2.5 and 3.5 mg/mL) enhanced the proliferation in serum-containing and serum-free culture conditions **(Boby and Prabha, 2011)**. Wound healing and anti-inflammatory activity were studied using various inhibition assays **(Joo, 2016)**.

Aqueous extracts (50% and 25%) of leaves of *Hemigraphis alternata* was treated on onion root tips. The extract treated root tips showed lower Mitotic index frequency and Chromosome aberrations than control. In addition, it showed Mitodepressive inhibition effect on *Allium cepa* cell division, and it may be used to detect tumour formations and finds use in cancer treatments **(Hilal, 2019)**.

Anti-inflammatory activity on mice showed a significant reduction of ear weight and granuloma formation at 400 mg/kg for MHAL and 200 and 400 mg/kg for EAHAL. At 400 mg/kg dose of EAHAL showed the highest anti-inflammatory activity (35.15±11.78% and 34.76±11.30%) and it showed the highest percentage inhibition (88.21±2.23% and 54.00±2.38%) at 400 mg/kg dose of EAHAL in anti-nociceptive experiments. Thus, both MHAL and EAHAL extracts showed their potential in anti-nociceptive, anti-inflammatory and anti-diarrhoeal activities **(Rahman *et al*., 2019)**.

Hemigraphis alternata was tested to remove the efficiency of five volatile indoor pollutants (benzene, toluene, octane, trichloroethylene and a-pinene). The efficiency was monitored by gas chromatography-mass spectroscopy for 3h and 6h. Glass container was used as a substrate. The plant had the highest removal efficiencies for all pollutants, and the removal efficiency depends on the indoor air quality and multiple species **(Dong *et al*., 2009)**.

The ethanolic extract contains carbohydrate, protein, alkaloid, flavonoid, saponin, terpenoid and tannin. The antioxidant activity of this extract (ascorbic acid-standard) showed based on a dose-dependent manner. The antibacterial activity of this extract showed against *Staphylococcus aureus* (38 mm) and *Proteus mirabilis* (34 mm) and antifungal activity against *Penicillium notatum* and *Candida albicans*. The petroleum ether extracts did not inhibit all bacteria and fungi tested **(Agneeswari and Jansi, 2019)**. Synthesized silver nanoparticles (20-50 nm) showed antibacterial activity against *E. coli*, *Bacillus species*, *Pseudomonas aeruginosa*, *S. aureus*, *Enterococcus species*, *Moraxella* species, *Klebsiella pneumoniae*, and *Serratia plymuthica*. The whole plant was used to stop external bleeding **(Neenu *et al*., 2018)**.

Table 3. List of identified phytoconstituents in *Hemigraphis alternata*

S. No	Constituents	Solvents	References
1	Alkaloids	Ethanol	**Reshma *et al*., 2018, Agneeswari and Jansi, 2019**
		Chloroform	**Reshma *et al*., 2018**
2	Flavonoids	Ethanol	**Reshma *et al*., 2018, Rahman *et al*., 2019, Agneeswari and Jansi, 2019**
		Methanol	**Rahman *et al*., 2019**
3	Sterols	Ethanol, Chloroform, Benzene	**Reshma *et al*., 2018**
		Hot water	**Rangheetha *et al*., 2016**
4	Tannins	Ethanol	**Reshma *et al*., 2018, Rahman *et al*., 2019, Agneeswari and Jansi, 2019**
		Chloroform, Benzene	**Reshma *et al*., 2018**
		Cold water, Hot water	**Rangheetha *et al*., 2016**
		Methanol	**Rahman *et al*., 2019**
5	Proteins	Ethanol	**Reshma *et al*., 2018, Agneeswari and Jansi, 2019**
		Chloroform, Benzene	**Reshma *et al*., 2018**
		Hot water	**Rangheetha *et al*., 2016**
6	Saponins	Ethanol	**Reshma *et al*., 2018, Agneeswari and Jansi, 2019**
7	Glycosides	Ethanol	**Reshma *et al*., 2018, Rahman *et al*., 2019**
		Chloroform, Benzene	**Reshma *et al*., 2018**
		Aqueous	**Priyanka *et al*., 2016**
		Methanol	**Rahman *et al*., 2019**
8	Amino acids	Chloroform	**Reshma *et al*., 2018**
		Hot water	**Rangheetha *et al*., 2016**
9	Terpenoids	Ethanol, Methanol	**Rahman *et al*., 2019**
		Benzene	**Reshma *et al*., 2018**
10	Gum	Benzene	**Reshma *et al*., 2018**
11	Phenols	Hot water	**Rangheetha *et al*., 2016**
12	Fat, oil	Ethanol, Methanol	**Rahman *et al*., 2019**

2.2.3 Reviews on pumice stone

Pumice is a type of water floating stone, and it is a hardened foam of lava that comes out from a volcano. The temperature (1600^{o}C) and pressure are very high inside the volcano. The volcanic lava form as a lightweight stone after it meets the cool air (25^{o}C) **(Das, 2017)**. It can also be done by mixing lava with hot water, followed by applying rapid cooling and depressurization **(Ersoy *et al*., 2010)**.

The pumice stone is utilized as a coarse aggregate. This lightweight concrete reduces the self-weight and helps to construct larger precast units. It reduces the mass of construction materials and improves the thermal properties and sound insulation capacity. Acid resistance properties of pumice maintain the adequate strength of concrete and it behaves like conventional concrete. This pumice can be utilized with M30 grade concrete and it used to build various parts of the building like lintels, partition walls and sunshades. The valuable properties of pumice lead to the construction of unaffected earthquake buildings **(Muralitharan and Ramasamy, 2015)**.

The M20 concrete strength increased due to the addition of pumice aggregate. Using pumice (40%) and fibre (0.5%) improves the strength for M20 concrete was achieved. The density of concrete is found to be decreased when the pumice content (%) increases **(Sivalinga *et al*., 2013)**. This showed good strength and sound absorption capacity **(Fauzi *et al*., 2016)**. The washing of garments (100% denim cotton) has achieved using enzyme and pumice stone mixture **(Sarkar *et al*., 2014a)**. It acts as a lightweight aggregate used for lightweight concrete **(Tanveer *et al*., 2016; Manzoor *et al*., 2018; Caiza *et al*., 2018; Meyyappan *et al*., 2019; Sangeetha *et al*., 2020)**. The porous structure is formed due to the release of gas from the molten lava **(Bhavana and Rambabu, 2017)**. Pumice stone act as a new adsorbent to remove impurities from the aqueous solution. This adsorption is dependent on pH (pH 3) of the solution. The increases in sample pH reduce the absorption **(Ulker and Sarioglu, 2014)**.

Nanoscale zero-valent iron (nZVI) was prepared using a pumice stone, and it showed efficiency to remove copper from water. It possesses a high surface area, and it increases the copper absorption (90%) than the untreated one. It also can be used to remove Ni, Zn, Cd, *etc*. **(Bilgehan and Mesut, 2016)**. Pumice stone used as a useful coating material in Ni-composites. The texture and wear resistance are improved for Ni–pumice composite compared to direct Ni coating. The pumice aided Ni composite improves the microhardness, wear and oxidation resistance. It can act as an alternative coating material for trochoids of Wankel engines **(Aruna *et al*., 2014)**.

Over 2000 years pumice used in various medicinal applications with the combination of various herbs. It is served a part in decoction (tea) for gall bladder cancer, anxiety disorders, coughs and urinary problems. Pumice act as dentifrices, cleansers (particularly of the skin, cornea), scar (cicatrizing agents) and used in eye ointments. Pumice powders are inducing sneezing problems, but it too helps in body hair removal by rubbing and resistance for pharmaceutical ingredients **(Christopher, 2012)**. Pentachlorophenol is a toxic phenolic compound that is effectively removed by copper treated zeolite and pumice. The highest removal efficiency were obtained using pumice (92%), and zeolite modified copper (96%) depends on pH, dose and time. This composite is used as a catalytic stabilizer and removal of water and wastewater contaminates were recommend due to their low cost and penetration efficiency **(Ali Reza *et al*., 2018)**. Deactivation of bacteria (*E-coli* from river water) and degradation of various organic dyes were successfully demonstrated using pumice stone coated TiO_2. This is an economically cheaper and efficient method in the treatment of bacteria and pollutants from drinking water **(Subrahmanyam *et al*., 2008)**.

Methylene Blue dye was removed using pumice stone from aqueous solution. The absorption was increased by increasing initial dye concentration (pH 10) and contact time **(Derakhshan *et al*., 2013)**. Acid Black dye was removed by pumice stone with high sorption capacity (72.46 mg/g). The removing capacity increases due to increase the contact time, initial dye concentration and acidic pH. The mechanism of adsorption depends on the molecular weight and sulphonate groups in the chemical structure of the adsorbent. Regeneration of the pumice also achieved (83%) **(Mohammad *et al*., 2013)** and the azo dye **(Veliyev *et al*., 2006)** and acid Red 14 **(Samarghandi *et al*., 2012)** was removed from wastewater also achieved.

Mainly the pumice stone is used by women for cleaning their feet (by removing calluses) and preventing snagging of nylon hose. It is used for removing hyperkeratotic tissue by the combination of keratolytic agents. Pumice stone with soap and water is beneficial in removing stains from the hands **(Howard and Boston, 1980)**. The adsorption of water hardening cations of Ca^{+2} and Mg^{+2} was achieved using modified pumice stone (Natural and alkaline). The removal efficiency of modified pumice stone absorbent 94% and 73% were obtained for calcium and magnesium, respectively. The raw pumice stone helps in the absorption of 48% and 83% for magnesium and calcium, respectively. Increase the adsorbent mass and contact time leads to increases in the efficiency of cations removal (**Sepehr *et al*., 2013)**.

The cream formulations containing pumice stone with other emulsifiers showed right consistency and high stability with time. It can be used in skincare products **(Estanqueiro *et al*., 2014)**. Cadmium was removed by using iron nanoparticles coated pumice from wastewater **(Shokoohi *et al*., 2016)**. Acid Red 14 (3.1 mg/g) and Acid Red 18 (29.7 mg/g) were effectively removed and the pumice absorbent showed 86% and 89% regeneration, respectively **(Reza *et al*., 2012)**.

2.3 Reviews on green synthesis of gold nanoparticles

Nanomaterials are versatile significance in all fields of science and technology. The high surface area of nanoparticles makes it unique with different properties that distinguish it from the bulk material. Nanomaterials are prepared by several means. Owing to the need for more economical methods and less-time consuming methods, the search for different types of synthesis or novel methods of synthesis is on the forefront. This review encompasses the various methods using plant sources for the synthesis of nanomaterials available in the literature. This review consists of gold nanoparticle synthesis of more than 75 plants, and biomaterial was reviewed on their synthesis, colour, size, shape, and proposed applications are listed in **Table 4**.

Table 4: Reviews on synthesis of gold nanoparticle using green sources by various methods

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
Sonication									
1	Citrus fruits	Fruit	Juice	20 min	Purple	45-75	Spherical	Drug delivery	**Dhanemozhi and Chitra, 2015**
2	*Amaranthus dubius*	Leaves	Aqueous	15 min	Violet	<70	Cubic	Nutraceuticals	**Firdhouse and Lalitha, 2014a**
3	Calothrix algae	Whole	-	3 days	Pink	30-120	Spherical and triangle	Catalytic conversion of 4-NP to 4-AP	**Kumar *et al*., 2016**
4	*Pithophora oedogonia*	Whole	Aqueous	1h	Purple	32	Spherical	Carbendazim detection in real soil samples	**Li Lei and Zhang, 2016**
5	*Piper betel*	Leaves	Aqueous	-	Pink	-	-	Medical and Pharmaceutical	**Mallikarjuna *et al*., 2013**
6	*Andrographis paniculata*	Leaves	Ethanol	2 min	Ruby red	5-75	Spherical	Cancer activity against HeLa and MCF-7	**Babu *et al*., 2012**
Microwave heating									
7	*Camellia sinensis* (Green tea)	Leaves	Aqueous	1 min	Red	17	Spherical	Biomedical	**Ahmmad *et al*., 2010**
8	*Trigonella foenum graecum*	Seed	Aqueous	30s	Purple-red	13.71	Spherical, triangular, pentagonal, rod	Vaccine storage indicator	**Fragoon *et al*., 2016**
						1.85	Hexagonal		
9	*Naregamia alata*	Leaves	Aqueous	-	Ruby red	17	Uneven	Antioxidant	**Francis *et al*., 2018**
								Antibacterial activity against 6 pathogens	
								Methylene blue and rhodamine degradation	
10	Papaya	Leaves	Aqueous	60s	Red	15	Spherical	Catalytic activity on p-nitrophenol (4-NP)	**Sunkari *et al*., 2017**
								Antimicrobial activity	
11	*Hibiscus rosasinensis*	Leaves	Aqueous	30s	Ruby red	16–30	Spherical	Biomedical and sensor applications	**Yasmin *et al*., 2014**
12	Heart leaf moon	Seed	Aqueous-ethanol	1 min	Ruby red	12	Polygonal, cylindrical, prism-shape	Biomedical	**Roy *et al*., 2017**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
Room temperature									
13	Palm oil	Fronds	Aqueous	1 min	Ruby red	66,79	Triangular, hexagonal	Biomedical	**Adamu *et al*., 2018**
14	*Delonix elata*	Leaves	Aqueous	30 min	Pink	24	Spherical	Antibacterial activity against *Staphylococcus aureus, Streptococcus pyogenes, Pseudomonas aeruginosa* and *Proteus vulgaris*	**Akilandeswari and Sathya, 2017**
15	*Peltophorum pterocarpum*	Flower	Aqueous	30 min	Pink	5-50	Spherical	Optics and solar cell	**Balamurugan *et al*., 2016**
16	*Bougainvillea glabra*	Leaves	Methanol	20h	Ruby red	-	-	-	**Rose *et al*., 2014**
17	*Corallina officinalis*	Red seaweed	Aqueous	-	Red	14.6	Spherical	Cytotoxicity against MCF-7 cell lines	**Yassin and Mostafa, 2014**
18	*Prosopis juliflora*	Leaves	Aqueous	15 min	Ruby red	22	Spherical	Antimicrobial inhibition on *E. coli* and *B. subtilis*	**Rao *et al*., 2017**
19	*Aloe barbadensis miller*	Whole	Aqueous	1h	Violet	-	Spherical	-	**Muralikrishna *et al*., 2014**
20	*Hypericum hookerianum*	Aerial	Ethanol	30 min	Ruby red	33-60	-	Antiparkinson effect	**Subakanmani *et al*., 2015**
21	*Zataria multiflora*	Leaves	Aqueous	30 min	Red	10-42	Pentagonal, triangle	Apoptosis for HeLa cells	**Javad *et al*., 2016**
22	*Bauhinia variegate*	Leaves	Aqueous	4h	-	50	Triangular, hexagonal, polygonal	Medicine, drug delivery and diagnostics	**Kumar and Sudesh, 2011**
23	*Callistemon citrinus*	Leaves	Aqueous	12h	Purple	8-15	Triangle, spherical	Antioxidant, antimicrobial and cytotoxic activities	**Saad *et al*., 2017**
24	*Aegle marmelos*	Leaves	Aqueous	-	-	-	Triangle	Antibacterial activities Cytotoxic activities	**Arunachalam *et al*., 2013**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
25	*Averrhoa bilimbi* Linn	Fruit	Aqueous	-	-	75-150	Hexagonal	Biological activities	**Rimal *et al*., 2013**
26	*Momordica cochinchinensis*	Rhizome	Aqueous	24h	Ruby red	16	Spherical, Triangle, hexagonal	Antioxidant, anti-bacterial activities	**Lakshmanan *et al*., 2016**
27	*Camellia sinensis*	Leaves	Aqueous	15 min	Purple-Red	40-70	Hexagonal	-	**Geraldes *et al*., 2016**
	Juniperus communis			90 min		20-30	Circular, hexagonal		
	Green coconut	Coconut water		30 min		30-70	Circular, triangular, hexagonal, rod		
28	*Turbinaria conoides*	Leaves	Aqueous	30 min	Pinkish ruby red	60	Square, rectangle, cubic, triangle	Antibacterial activity (3 pathogens)	**Kumar *et al*., 2013**
29	*Lavandula angustifolia*	Leaves	Aqueous	4h	Pink	34–300	Poly disperse, quasi-spherical, Triangular	antioxidant activity	**Brajesh *et al*., 2016**
30	*Gnidia glauca*	Flower	Aqueous	20 min	Ruby red	50	Spherical	Chemo catalytic activity	**Ghosh *et al*., 2012**
31	*Bacopa monniera*	Whole	Ethanol	1h	Ruby red	15-35	Spherical	Antibacterial activity (4 pathogens)	**Mahitha *et al*., 2013**
32	*Garcinia cambogia*	Leaves	Aqueous	24h	Violet	-	-	Free radical scavenging activity	**Nithya and Jayachitra, 2016**
33	Apple juice	Juice	Aqueous	30 min	Red-purple	11.1	Undesired	Sensitive colorimetric detection of cysteine	**Bagci *et al*., 2015**
34	*Amentotaxus assamica*	Leaves	Aqueous	5 min	Wine red	23	Polygonal	Catalytic activity of borohydride to different isomeric nitrophenols	**Phukan *et al*., 2016**
35	*Tinospora cordifolia*	Stem	Aqueous	24h	Purple	16-30	Spherical	-	**Anuradha and Abbasi, 2014**
						25-75	Triangular, hexagonal, pentagonal		
36	*Mimosa pudica*	Flower	Aqueous	-	Ruby red	~24	Spherical	Catalytic activity of 4-nitrophenol	**Mapala and Pattabi, 2017**
37	*Callophyllum inophyllum*	Leaves	Aqueous	-	-	27.52	Spherical	-	**Yudha *et al*., 2015**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
38	*Lantana camara* Linn	Leaves	Aqueous	5 min	Pinkish red	6–7	Spherical	Reduction of 4-nitrophenol to 4-aminophenol	**Shib *et al*., 2015**
39	*Dillenia indica*	Fruit	Aqueous	15 min	Ruby red	5–50	Spherical	Cytotoxicity, drug delivery and molecular imaging	**Setti *et al*., 2016**
40	*Senna siamea*	Seed	Aqueous	3h	Pink	100	Hexagonal	Antibacterial activity	**Reddy *et al*., 2013**
41	*Lagerstroemia speciosa*	Leaves	Aqueous	30 min	Ruby pink	40-200	Hexagonal, triangular, spherical	Waste water treatment	**Bharat *et al*., 2017**
42	Mushroom	Fungus	Aqueous	24h	Red	-	-	Antioxidant	**Madhanraj *et al*., 2017**
43	*Azima tetracantha* Lam	Leaves	Aqueous	10 min	Pink	80	Spherical	Antibacterial activity (against 4 pathogens) Antifungal activity (against 4 pathogens)	**Hariharan *et al*., 2016**
44	*Couroupita guianensis*	Flower	Aqueous	10 min	Purple	37.2	Spherical	Antibacterial activity against *Bacillus cereus*	**Kirbha and Alagumuthu, 2014**
45	*Ficus benghalensis*	Leaves	Aqueous	5 min	Pink	-	Spherical	Antibacterial activity (against 4 pathogens)	**Francis *et al*., 2014**
46	*Garcinia mangostana*	Peel	Aqueous	3 min	Purple	32.96	Spherical, hexagonal, triangular	Biomedical	**Lee Kar *et al*., 2016**
47	*Terminalia catappa*	Leaves	Aqueous	5 min	Red wine	18-44	Spherical	-	**Muhammad *et al*., 2014**
48	*Allium cepa*	-	Aqueous	-	Cherry yellowish red	~100	Spherical, cubic	Cell viability against MCF-7 cells	**Parida *et al*., 2011**
49	*Costus igneus*	Leaves	Ethanol	15 min	Ruby red	54-62	Spherical	Antibacterial activity (against 6 pathogens)	**Velumani, 2015**
50	*Camellia sinensis* (green tea) *Syzygium aromaticum*	Leaves	Aqueous	<30s 30–60 min	Wine red	-	-	-	**Priya and Priya, 2014**
51	Black tea	Leaves	Ethanol	15 min	Purple	2.5-27.5	-	-	**Banoee *et al*., 2010**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
52	*Silybum marianum*	Seed	Aqueous	15 min	Magenta	64	Spherical and anisotropic	Biological and medical applications	**Gopalakrishnan and Raghu, 2014**
53	*Punica granatum*	Juice	Aqueous	24h	Red	23-36	Triangle, pentagonal, hexagonal, spherical	Borohydride reduction of 4 nitrophenol	**Shib and Braja, 2014**
54	*Acacia nilotica*	Leaves	Aqueous	5 min	Red	6-12	Spherical	Catalyst for the reduction of 4-nitrophenol to 4-aminophenol	**Majumdar *et al*., 2013**
55	*Shewanella algae*	Whole	Aqueous	60 min	Purple	9.6	Spherical	-	**Ogi *et al*., 2010**
						100-200	Nanoplate		
56	*Galaxaura elongate*	Whole	Aqueous	2-5 min	Red	3.85-77.13	Rod, triangular, hexagonal	Gram-negative bacterial inhibition activity	**Neveen *et al*., 2013**
				Varying pH					
57	*Momordica charantia* 30 ^{0}C	Fruit	Aqueous	pH 4 (24 h)	Wine red	-	-	Drug delivery	**Pandey *et al*., 2012**
				pH 6					
				pH 8					
				pH 10		10-100	Spherical		
	100 ^{0}C			pH 4 (5 s)		-	-		
				pH 6					
				pH 8					
				pH 10		30-100	Spherical		
58	*Neurospora crassa* 60^0 C	Fungus	Aqueous	pH 3 (3 h)	Purple	10-200	Triangles, hexagons, pentagon	Sensor technology	**Quester *et al*., 2013**
				pH 5.5		6-23	Quasi-spheres		
				pH 10		3-12			
59	*Shorea tumbuggaia*	Bark	Aqueous	20 min	Ruby red	20	Spherical	Cytotoxic activity against SW579 cell lines	**Li Yuan *et al*., 2017**
60	*Capsicum baccatum*	Fruit	Aqueous	1h	Purple-pink	8–35	Spherical	Photocatalytic activity	**Brajesh *et al*., 2015**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
61	*Mentha piperita*	Essential oil	-	24h	Ruby red colour	60	Dot round shape	Antifungal activity against *A. niger*, *A. flavus, C. tropicalis* and *C. albicans*	**Thanighaiarassu *et al*., 2014**
62	*Ziziphus ziziphus*	Leaves	Aqueous	-	Ruby-red	30-50	Triangular and hexagonal	Antibacterial activity	**Alaa *et al*., 2018**
Incubatory shaker									
63	*Ocimum sanctum*	Leaves	Chloroform	6h	Purple and red	>200	Circular discs with rough edges	Large scale production of AuNP	**Lee *et al*., 2016a**
			Hexane			1-50	Spherical		
			Aqueous			50-300	Anisotropic platelets		
			n-butanol			-	Au aggregates		
64	*Zingiber officinale*	Rhizomes	Aqueous	12h	Ruby red	5–35	Spherical	-	**Bahig *et al*., 2016**
Water bath heating									
65	*Magnolia kobus*	Leaves	Aqueous	1 min	Ruby red	100-300	Spherical	Environmental, biotechnological, electrochemical and biomedical applications	**Li Ying *et al*., 2012**
Stirring method									
66	*Moringa oleifera*	Pods	Aqueous	24h	Brown	40-80	Irregular	Hepatoprotective activity	**Belliraj *et al*., 2015**
67	*Solanus lycopersicum*	Fruit	Aqueous	1h	Violet	416	-	-	**Pattanayak and Nayak, 2014**
68	*Erythrina variegate*	Leaves	Aqueous	10 min	Ruby red	2-20	Spherical	-	**Rani *et al*., 2016**
69	*Leucas aspera*	Leaves	Aqueous	4h	Dark brown	17	Spherical	Antibacterial activity against *S. epidermidis* and *E. coli*	**Prabu *et al*., 2016**
								Anticancer activity against HepG2 cells	
70	Orange	Peel	Aqueous	1 min	Purple	-	-	-	**Castro *et al*., 2013**
71	*Papaver somniferum*	Pod	Methanol	4h	Yellow brown	77	Spherical	Drug delivery and gene therapy	**Muhammad *et al*., 2017**
72	*Pergularia daemia*	Leaves	Aqueous	10 min	Brown	3-15	Spherical	Bacterial inhibition activity	**SenthiKumar *et al*., 2017**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
Sunlight									
73	Manuka Honey	-	Aqueous	-	Brownish -black	100	Spherical	Bacterial inhibition activity (*E. faecalis, S. aureus* and *E. coli*)	**Meenakshi and Jangir, 2016**
74	*Pisonia* grandis	Leaves	Aqueous	10-20 min	Pink	60	Spherical	Drug carrier	**Devendiran *et al*., 2014**
UV Light									
75	*Polyscias scutellaria*	Leaves	Aqueous	2h	Pink	5–20	Spherical	Catalytic activity to reduce methylene blue	**Yulizar *et al*., 2017**
76	*Vetiveria zizanioides*	Root	-	10 min	Pink	10–35	Spherical	Antifungal activity	**Swain *et al*., 2016**
	Cannabis sativa	Leaves							
77	*Delonix regia*	Leaves	Aqueous	10 min	-	4–24	Spherical	Catalytic activity of nitro-organic pollutant	**Dauthal and Mausumi, 2016**
78	*Artemisia capillaris*	Aerial	Aqueous	-	Violet	17-30	Spherical	Catalytic activity of 4-nitrophenol reduction	**Soo *et al*., 2016**
79	Turnip	Leaves	Aqueous	-	Pinkish-red	45	-	Methylene blue reduction	**Kannan and Hyun, 2015**
80	*Sargassum muticum*	Marine algae	Aqueous	30 min	Purple	5.42	Spherical	Pharmaceutical and biomedical	**Namvar *et al*., 2015**
81	*Abroma augusta*	Bark	Ethanol	-	Red	23-33	Spherical, triangular, pentagonal, hexagonal	4-nitrophenol reduction	**Subhajit *et al*., 2015**
82	*Zoogloea ramigera*	Biomass	-	24h	Red	4–16	Spherical	Antibacterial activity (4 pathogens)	**Srivastava and Mausumi, 2014**
83	*Magnolia kobus*	Leaves	Aqueous	24h	Purple	64	Triangles, pentagons, hexagons	-	**So Hyun *et al*., 2014**
84	*Saraca indica*	Bark	Aqueous	-	Pinkish red	15-23	Poly shaped	Catalytic reduction of 4-nitrophenol	**Shib *et al*., 2014**
85	*Prunus armeniaca*	Fruit	Aqueous	8 min	Purple	<20	Spherical	Scavenging activity	**Dauthal and Mausumi, 2013**
86	*Padina gymnospora*	Leaves	Aqueous	5 min	Ruby red	53–67	Spherical	Cosmetics, foods and medical applications	**Singh *et al*., 2013**

S. No	Plant name	Part of the plant used	Extract	Time of formation	Colour	Size (nm)	Shape	Proposed application	References
87	*Aspergillus fumigatus*	-	-	72h	Pinkish red	85–210	Spherical and irregular	-	**Vasanthi *et al*., 2013**
88	*Lycopersicon esculentum* (red tomato)	Fruit	Aqueous	-	Reddish violet	5-10	Spherical	Detect and estimate the pesticide, methyl parathion	**Barman *et al*., 2013**
89	*Eucalyptus macrocarpa*	Leaves	Aqueous	1h	Brown	20-100	Spherical, triangular, pentagon, hexagonal	Antibacterial activity	**Poinernr *et al*., 2013**
90	*Semecarpus anacardium*	Leaves	Aqueous	15-20 min	Pink	13-55	Poly-dispersed	-	**Raju *et al*., 2011**
91	*Psidium guajava*	Leaves	Aqueous	5 min	Purple	27	Spherical	-	**Raghunandan *et al*., 2009**
92	*Albizia amara*	Leaves	Aqueous	10 min	Purple	34-64	Hexagonal	Antioxidant, antibacterial and anticancer activity	**Balasubramani *et al*., 2016**
93	*Mappia foetida*	Leaves	Aqueous	4 min	Wine red	10–20	Spherical	Cancer activity against MDA-MB-231, HeLa, SiHa and Hep-G2	**Yallappa *et al*., 2015**
94	*Gymnema sylvestre*	Leaves	Aqueous	30 min	Red	26	Spherical	Radical scavenging efficacy and antiproliferative activity against Hep2 cells	**Nakkala *et al*., 2015**
95	*Fagopyrum esculentum*	Leaves	Ethanol	15s	Ruby red	8.3	Spherical, hexagonal, triangular	Cytotoxicity against human cervical, breast cancer and IMR-32 cancer cell lines	**Babu *et al*., 2011**

2.4 Synthesis of reduced graphene oxide using green sources

Reduced graphene oxide is one of the carbon-based nanosheets with hexagonally arranged monolayers (atomic thickness). It consists of unique properties (chemical, physical, and mechanical), and it exhibits a large area with uniform surfaces. It has important functional groups like epoxide (-O-), carboxyl and hydroxyl (-OH). It also has plane edges and basal surfaces based on their oxidation and reduction properties. Optical absorption of rGO is high during near NIR light and elevated temperature **(Kim *et al*., 2014)**. Chemical methods are widely used to reduce GO. The plant materials are an alternative resource to reduce GO into rGO. The plant-based reduced graphene oxide is given in **Table 5**.

Table 5. List of green sources for synthesizing reduced graphene oxide

S.No	Plant name	Part of the plant	Proposed application	References
1	*Polyalthia longifolia*	Leaves	Conductive transparent film	**Chamoli *et al*., 2016**
2	*Ficus religiosa*			
3	*Mangifera indica*			
4	*Terminalia chebula*	Seeds	-	**Maddinedi *et al*., 2015**
5	*Nigella sativa*	Seeds	Antibacterial and anticancer activity	**Awad *et al*., 2017**
6	*Paederia foetide*	Leaves	Electrical conductivity	**Tapas *et al*., 2016**
7	*Artemisia herba alba*	Leaves	Optoelectronic	**Khenfouch *et al*., 2016**
8	*Terminalia bellirica*	Fruit	-	**Maddinedi and Mandal, 2016**
9	Caffeine	-	Biomedical applications	**Thu Ha Thi Vu, 2015**
10	*Salvadora persica* L.	Roots	Technological and biological fields	**Khan *et al*., 2015**
11	*Ginkgo biloba*	Leaves	Biocompatibility for human breast cancer (MDA-MB-231)	**Gurunathan *et al*., 2014**
12	*Peltophorum pterocarpum*	Pollen grains	Electrochemical applications	**Rahman *et al*., 2014**
13	*Amaranthus dubius*	Whole	-	**Firdhouse and Lalitha, 2013**
14	*Prunus serrulate*	Leaves	Biomedical applications	**Lee and Kim, 2014**
15	*Citrullus colocynthis*	Leaves	Cytotoxicity against DU145 cells	**Zhu *et al*., 2016**
16	*Ocimum sanctum* L.	Leaves	-	**Mahata *et al*., 2018**

S.No	Plant name	Part of the plant	Proposed application	References
17	*Eichhornia crassipes*	Whole	-	**Firdhouse and Lalitha, 2014**
18	*Shewanella* cells	Bacteria	Electrochemical	**Wang *et al*., 2011**
19	Grapes	Seed	Antimicrobial activity against *E. coli* and *S. aureus.* Anti-proliferative activity (88%) against HCT-116 cell line	**Yaragalla *et al*., 2016**
20	*Camellia sinensis*	Green tea	Supercapacitor and biosensor fields	**Selvakumari *et al*., 2016**
21	*Vitis vinifera*	Fruit	Removal of organic dyes from waste water	**Upadhyay *et al*., 2015**
22	*Platanus orientalis*	Leaves	Cytotoxicity against cardiac cell lines of *Catla catla*	**Xing *et al*., 2016**
23	*Eichhornia crassipes*	Whole	Anticancer activity against MCF-7 cell lines	**Santhiya and Lalitha, 2020**
24	*Melissa officinalis*	Leaves	Proliferative activity	**Elif *et al*., 2017**

2.4.1 Pharmacological application of reduced graphene oxide

The antibacterial activity was tested using combined reduced graphene oxide with luminescent Eu (3+) complex and vancomycin under NIR illumination. The combined complex provided more heat and the ability to kill drug-resistant bacteria **(Xinjian *et al*., 2014)**. The cell viability of *P. aeruginosa* bacteria decreases depending on the dose and the treatment time of rGO. The antibacterial activity was confirmed by cell death, followed by induced oxidative stress, and DNA fragmentation **(Sangiliyandi *et al*., 2012)**. Ag–rGO nanocomposite prepared using *Potamogeton pectinatus* extract, showed enhanced antibacterial activity due to the combination of Ag–rGO nanocomposite **(Sedki *et al*., 2015)**. AgNPs-AuNPs-rGO nanocomposites showed antibacterial activity and bacterial DNA damage at a lower level of metal ions **(Alazzam and Alsharaeh, 2015)**.

Poly-l-lysine (conjugated with anti-HER-2 antibody) functionalized rGO successfully delivered the drugs (doxorubicin) into the nucleus of HER-2 cancer cells. The higher drug loading and colloidal stability of the complex improve MCF-7 and HER-2 cell lines **(Zheng *et al*., 2016)**. rGO synthesized using pyrogallol shows toxicity against HeLa cell lines in a dose-dependent manner (80% higher concentration) **(Luo *et al*., 2017)**. Ag–rGO nanocomposite prepared using *Pulicaria glutinosa*, shows excellent anticancer activity against A549 cell lines. The activity increases while increasing the concentration of silver on rGO **(Khan *et al*., 2016)**. The interaction between rGO and lung cancer cell lines (A549 and SKMES-1) was studied by oxidative stress potential and induction of apoptosis. Late apoptosis and necrosis occur based on the dose-dependent manner **(Tanveer *et al*., 2017)**.

In blood-brain barrier (BBB) paracellular tightness, transitory decrease by injecting rGO in the thalamus and hippocampus of rats. The activity was confirmed at anatomical, subcellular and molecular levels. Due to this, BBB impermeability of rGO is useful for brain disorders **(Mendonca *et al*., 2015)**. ZnO/rGO composite was prepared using Aloe vera gel as a surfactant by the facile hydrothermal route. These nanocomposites are used in the applications of radiation dosimetry and antioxidant activities **(Kavyashree *et al*., 2016)**. Due to the excellent stability (2 years) of Paclitaxel loaded rGO is effectively involved in the drug delivery for MRC-5 and nasopharyngeal cancer cells **(Geetha *et al*., 2017)**.

The composite of rGO-tetraethylenepentamine effectively detected cervical cancer by electrochemical method **(Wu Dan *et al*., 2016)**. Prostate cancer biomarker was detected using palladium nanoparticle-rGO at the concentration of 10 pg mL^{-1} **(Vinod *et al*., 2014)**. rGO has more advantages in photoacoustic imaging and therapeutic agent for tumour **(Sheng *et al*., 2013; Shi *et al*., 2013)**. Glucose-mediated rGO showed fragmentation in cancer cells, and the agglomeration was prevented by gluconate ions **(Akhavan *et al*., 2012)**. rGO-AgNP composites reduced by vitamin C, show anticancer activity against A549 cancer cell lines **(Kavinkumar *et al*., 2017)**. These composites detect flutamide (an anticancer drug) in the detection range 0.1 to 0.3 mM with a detection limit of 1.16 µM **(Banerjee *et al*., 2017)**.

2.4.2 Photo and electro catalytic applications of reduced graphene oxide

ZnO-rGO **(Gong *et al*., 2014)**, TiO_2-rGO nanocomposite **(Zhou *et al*., 2013; Loryuenyong *et al*., 2016)**, CdS-TiO_2-rGO **(Pant *et al*., 2016)**, TiO_2-rGO-Ag nanocomposite **(Fu Li *et al*., 2015)** and rGO-WO_3 nanoplates **(Chai Bo *et al*., 2014)** showed photocatalytic degradation of methylene blue. The degradation of methylene blue occurs with rGO-CdO under UV irradiation **(Kumar *et al*., 2016)**. A luminescent nanocomposite aided graphene and a luminescent moiety are used in the luminescent film in electronic waste components **(Thalappil *et al*., 2014)**. CdS-rGO composites showed photocatalytic activities due to minimized photoinduced electrons, holes and the high surface areas in rGO sheets **(Liu *et al*., 2014)**.

The $PrPO_4$-rGO nanocomposites show the excellent photocatalytic activity of methylene blue under sunlight irradiation, contributing to the separation of electron-hole pairs and an increase of OH atmosphere in the reaction system **(Lv Hongwei *et al*., 2014)**. Furthermore, Cu_2O-rGO composites showed an excellent photocatalytic reduction of CO_2 without any co-catalyst **(An Xiaoqiang *et al*., 2014)**. In addition, TiO_2-rGO-Ag nanocomposites **(Arif *et al*., 2013; Zhang *et al*., 2017)**, TiO_2-rGO nanocomposite **(He Rong and He Wei, 2016)**, Cu_2O_2-rGO nanoparticles

(Zheng *et al*., 2016a) and TiO_2-rGO-COOH **(Pei *et al*., 2013)** showed Rhodamine B degradation activity.

The Cu-rGO-TiO_2 composite is used for the photo electrocatalysis of CO_2. The production of formic acid and methanol are the main products of this reaction (32.47%) **(Hasan *et al*., 2015)**. TiO_2–rGO composites are used in degrading methyl orange **(Lu Zeyu *et al*., 2015)**. *Psidium guajava* aided rGO and Ag nanoparticles are used for detecting methylene blue (at a concentration of 10^{-8}M and enhancement factor is 4.6×10^{5}) **(Chettri *et al*., 2017)**. rGO functionalized phenylboronic acid nanocomposite is used in glucose detection (2 to 75 mg/mL detection limit) **(Basiruddin and Swain, 2016)**.

The prepared rGO thin films have the potential in detecting gas molecules. The gas sensing properties of the films showed 27% and 54% for variable concentrations of Cl_2, and NO_2 gases. The films also exhibit good electrical conductivity (1.04 S cm^{-1}) at room temperature, and they also exhibit antibacterial activity on *E. coli* and *B. cereus* **(Nagesh *et al*., 2015)**. rGO-TiO_2 nanocomposite showed an energy conversion efficiency of 4.74%, and on the surface of TiO_2 the efficiency is 5.83% in the solar cell **(Lim *et al*., 2015)**.

A DSSC was fabricated using rGO-Cu_2S quantum dots. It has high transparent performance, excellent conductivity and catalytic activity. The conversation efficiency of the device is 7.12% compared to Pt-based device **(Bi Enbing *et al*., 2015)**. The electron transportation and collecting capability of rGO with TiO_2 improves solar cell efficiency (conversion efficiency values up to 19.54%) **(Cho *et al*., 2016)**. The degradation of phenol, 2-chlorophenol and 2-nitrophenol was attained at 3h, 3h and 5h, respectively, using Au-Pd-rGO **(Darabdhara *et al*., 2016)**.

The photocatalytic activity increases with increasing rGO content in ZnO-rGO, and it significantly showed surface hydrophilicity and photoinduced super hydrophilicity. Increases in the ratio of rGO-ZnO enhanced the activity of charge separation (from ZnO conduction band to graphene). It increases super hydrophilicity and decreases particle size suggesting use in self-cleaning applications **(He H-Y, 2015).** rGO-ZnO was tested for its supercapacitor and energy storage performance **(Bu and Huang, 2015)**. One-dimensional Ag nanowire-doped rGO integrated CdS nanowires is an efficient co-catalyst in artificial photosynthesis **(Liu *et al*., 2015)**.

2.5 Applications of gelatin/chitosan/alginate film-based materials

2.5.1 Drug delivery applications of gelatin/chitosan/alginate film-based materials

Chitosan is a naturally available mucopolysaccharide from marine chitin and it is linked with 2-amino-2-deoxy-D glucan. This possesses various properties such as nontoxic nature, biodegradability and biocompatibility and hence finds application in the biomedical field. Chitosan has the potential for electrostatic interactions with other anionic polysaccharides. It possesses high mechanical strength and increases the tensile property. The chitosan molecular weight and solvent system control the physical and mechanical properties of the chitosan film **(Bhuvaneshwari *et al*., 2011)**.

Lactobacillus rhamnosus containing alginate and gelatin capsules were prepared using a calcium chloride crosslinker. Alginate (1% w/v) and gelatin (0.1% w/v) showed the highest cell viability (4.2 x 109 CFU/g). The interaction between alginate and gelatin was confirmed by FTIR and TGA studies. The encapsulation of *Lactobacillus rhamnosus* formed 109 CFU g^{-1} viable cells in an alginate-gelatin bead. After four months the survival cells (105 CFU g^{-1}) were maintained in prepared beads. These results suggest the use of formulation in producing probiotic microcapsules **(Lopes *et al*., 2017)**.

Temephos (insecticide) loaded chitosan, alginate and gelatin were crosslinked with glutaraldehyde capsules and tested against *Culex pipiens* larvae. The highest *in vitro* release of the insecticide was found on the first day. The prepared capsules (0.05g) showed the highest mortality (100%) up to 120 and 92 days by stagnant and running water experiments, respectively. The prepared capsules exhibited high residual activities against the larvae for a long time (more than six months), and these biodegradable capsules are suggested as temephos carriers **(Mohamed *et al*., 2015)**. Norethisterone acetate loaded chitosan-sodium alginate has high drug loading efficiency (48.92-75.56%) for 10-23 days **(Sohani *et al*., 2016)**.

The release of the anticancer drug of 5-fluorouracil (5-FU) increased by increasing the amount of gelatin and drug and decreased with an increase in crosslinker. The prolonged-release rate was observed up to 12h **(Mallikarjuna *et al*., 2015)**.

Nimodipine loaded alginate beads show the drug release at pH 6.8 **(Rajendran and Sanat, 2009)**. Chitosan and sodium alginate nanocomposite were blended with different ratios of Cloisite® 30B, which is used as a co-emulsifier. Curcumin-loaded nanocomposites showed more pronounced drug release in the basic medium than the acidic medium **(Malesu *et al*., 2011)**.

The sago starch intermediated LDPE biodegradable polymer was synthesized using TMPTA cross-linker and EB irradiation. An increase in starch content increases modulus, mass loss and elongation. The polymer strength and ductility increased with an increase in the concentration of starch, TMPTA, and EB irradiation **(Hoque *et al*., 2013)**. Sodium alginate treated with octylamine and kaempferol overcome the hydrophilic nature.

Hydrogel films cross-linked by calcium chloride exhibit an inter-porous structure, show good swelling and mechanical properties. In addition, the films had 70.4% encapsulation efficiency and drug release up to 80h, which did not show any cytotoxicity on human umbilical vein endothelial cells **(Guarino *et al*., 2015)**.

The gelatin–sodium alginate microparticles are porous and sensitive to pH. It is effectively used as a drug carrier for ascorbic acid **(Devi and Dilip, 2013)**. Microencapsulation of prebiotics with alginate-gelatin significantly protects *Lactobacillus acidophilus* and *Lactobacillus casei* against an adverse condition of human gastro-intestinal condition. Alginate–gelatin microcapsules might be potentially used as a safe and protective vehicle delivery for controlling viable probiotic bacteria **(Mathews, 2017)**.

Alpha-lipoic acid-chitosan and Alpha-lipoic acid-alginate/gelatin-zinc are encapsulated by dipping method with controlled conditions. The α-lipoic acid encapsulation is 53.9% and it showed antioxidant activity at a satisfactory level. The alginate/gelatin hydrogel microparticles did not release Alpha-lipoic acid **(Bojana *et al*., 2016)**. Sodium alginate/gelatin (SA/Gel) hydrogel beads were prepared using Ca^{2+} and glutaraldehyde (GTA) crosslinker. It provided higher encapsulation efficiency and a prolonged drug-releasing profile **(Liu *et al*., 2012)**.

Catechin (isolated from *Cassia fistula*) is loaded into the nanoparticles of chitosan-alginate and chitosan-carrageenan composite. The prepared polymers have mucoadhesive properties, encapsulation efficiency (65%-97%) and catechin stability (99.56%-94.5%). Due to this activity, it effectively releases and distributes catechin into plasma and tissues **(Manikkam and Pitchai, 2014)**.

2.5.2 Tissue engineering application of gelatin/chitosan/alginate film-based materials

EDC treated chitosan-gelatin scaffold showed high porous, viscoelasticity and mechanical strength. The mixture of the scaffold was further treated with sodium alginate hydrogel, which showed cyto-biocompatibility. It is a potential agent for cartilage tissue engineering. The prepared scaffold is used as a hydrogel in Chondrocyte culture **(Zhao *et al*., 2014)**. The Chitosan-gelatin-glycerol phosphate disodium salt hydrogels show the efficiency of the 3D carrier for Nucleus Pulposus cell culture. The addition of gelatin increases the gelation, and these hydrogels are suitable for potential application in the tissue engineering of IVD **(Cheng *et al*., 2010)**.

Purified silica from geothermal power plants was incorporated into chitosan-based scaffolds using lyophilization. The presence of silica in scaffolds decreases the swelling ability and increases the mechanical strength of chitosan, alginate and geothermal silica (1:1:1). In addition, pores in scaffolds help to increases the cell infiltration and nutrient absorption for cell growth. The prepared bio-composites are biocompatible and promising scaffolds for tissue engineering applications **(Kusumastuti *et al*., 2018)**.

An artificial collagen skin prepared using a composite of nano-TiO_2–chitosan showed moderate water absorptivity (110 wt%), biodegradability (<3.0%), low density and favourable time-dependent biodegradability. The composite contains TiO_2 showed bactericidal activity with high pseudo-first-order kinetic coefficients (0.1736–0.0360 min). *In vivo* studies revealed faster recovery of wounds, and these results suggested the prepared composite for use in preparing artificial skin substitutes **(Peng *et al*., 2008)**. Alginate-gelatin crosslinked hydrogel showed a high degradation rate, fibroblasts attachment and proliferation **(Sarker *et al*., 2014)**.

Chitosan-alginate hydrogel shows a porous structure and highly hydrophilic nature. The cell proliferation study showed that OECs and NSCs proliferated and this result indicates a potential application in neural tissue engineering **(Wang *et al*., 2017a)**. The hemostatic sponge was prepared using carboxymethyl chitosan and sodium alginate. $CaCl_2$ used as a crosslinking agent in this preparation. The water absorption and porosity ratio of the sponge is 30.50% and 67.23%, respectively. Thus, this sponge had significant *in vitro* procoagulant activity, and it could be a potential agent in novel medical biomaterial **(Z Hu *et al*., 2017)**.

Compared to gelatin/sodium alginate 3D bioprinting materials, gelatin/sodium alginate/carboxymethyl chitosan shows potential applications in tissue engineering scaffolds and 3D bioprinting due to the excellent mechanical and degradation property. It also exhibits good antibacterial activity **(Huang *et al*., 2016)**. Nanofibers were prepared using gelatin, sodium alginate with chitosan oligosaccharide by spiral-electrospinning. The large-scaled produced nanofibers have a high potential for application in biomedical fields **(Lu Weipeng *et al*., 2016)**. *Lactobacillus plantarum* TN9 were encapsulated into alginate, chitosan and gelatin composite. Encapsulated *lactobacillus plantarum* TN9 enhanced the antibacterial activity in the presence of chitosan and not in a gelatin medium. The effect is due to the production of organic acids, and it is suggested as the agent for food and animal feed industries **(Trabelsi *et al*., 2014)**.

2.5.3 Wound dressing applications of gelatin/chitosan/alginate film-based materials

Skin substitute was prepared with the composition of mPEG–PCL–grafted gelatin/hyaluronan/chondroitin sulphate/sericin. The *in vitro* biocompatibility with fibroblasts and keratinocytes by cellular adhesion and proliferation was studied. This composite improves the 2^{nd}-degree burn wound healing activity **(Bhowmick *et al*., 2018)**. Artificial skin was prepared using chitosan and Bacterial cellulose (BC is obtained from *Acetobacter xylinum* bacteria). It exhibits better-wound healing activity by providing a moist environment to a wound, resulting in better wound healing properties, but it did not show any antimicrobial activity. The addition of chitosan improves the antimicrobial activity of the skin **(Lina *et al*., 2009)**.

Chitosan nanoparticles loaded calcium alginate hydrogel accelerate the wound healing activity by regulating the inflammation and neovascularization. It shows antibacterial activity against *E. coli* and *S. aureus*. It shows excellent wound healing and re-epithelialization activity on mice. The nanoparticle aided hydrogel can generate ROS production and used as a wound healer **(Wang *et al*., 2017)**.

Chitosan shows faster blood clotting by an increase in concentration. The decrease in the concentration of sodium and calcium alginate reduces the clotting time. The blood clotting time of nano-sized biopolymer is less than raw biopolymers. These nano biopolymers used as a wound dressing material for blood clotting applications **(Sivakumar *et al*., 2017)**. Fibrin–chitosan–sodium alginate composite sheet possesses high mechanical properties because of strong ionic interaction between the amine (chitosan) and carboxyl groups (alginate). The prepared films showed excellent wound dressing efficiency on wounded dogs **(Devi *et al*., 2012)**.

2.5.4 Other applications of gelatin/chitosan/alginate film-based materials

Nanofibers were prepared using poly-ion complex (chitosan-alginate) with polyvinyl alcohol (PVA). The formation of a larger diameter of nanofibers were formed in the presence of higher concentration of acetic acid. The higher concentration of sodium alginate increases the conductivity and viscosity of the complex **(Kusumastuti *et al*., 2016)**.

Chitosan-based productive coatings were prepared and coated on veal surfaces, rabbit meat, boiled sausages, smoked sausages, and smoked-boiled pork. The composition of chitosan and organic acids with gelatin (2% solution) showed the most potent bacteriostatic effect, increasing self-life on meat and meat products. This showed the most substantial suppressing effect on microflora, and it has the strongest stabilizing effect on samples with the least amount of basic hurdle factors **(Denis *et al*., 2013)**.

An increase in chitosan concentration in the composite of gelatin/CMC/chitosan, increases the crystalline nature and reduces the amorphous character. These formulations exhibit significant weight loss after the burial in soil (5 days). The film properties were not be affected by changing the temperature, and hence can be used as food packaging materials **(Jahit *et al*., 2016)**.

2.6 Reviews on unwanted hair removal

The growth of hair in the human body is a routine cyclic. The need for hair depends on the place in which it is grow. Photo-epilation is used to remove unwanted hair permanently. The mechanism of this technique involves the destruction of the hair follicle by using thermal energy. The permanent destruction is influence the hair growth cycle by the photo thermolysis-induced hair removal method **(Mandt *et al*., 2006)**.

The unwanted hair growth may be caused by androgen overproduction, circulation of androgens, metabolic and endocrine disorders. The hair removal efficiency is varied due to the efficacy of the treatment, comfort, and cost. The hair follicle is removed by surgery. Electrolysis is the only permanent treatment. Shaving, epilation, and depilation are the most common methods for facial hair removal with temporary results. Laser hair removal also shows a long-term effect but is yet to proven for the permanent treatment and Eflornithine is a simple method with minimal side effects **(Donald and Virginia, 2002)**.

Axillary hair is responsible for the odour in the underarm among men. Hair removal by clipping and soap washing did not show any immediate effect than soap washing alone. Shaving, waxing and soap washing showed good and immediate axillary odour reduction than soap washing alone **(Lanzalaco *et al*., 2015)**. Epilation showed comparable effects on surgery by visual acuity and corneal outcomes **(Rajak *et al*., 2011)**. Hair removal is important during surgery because it causes surgical site infections. Many awareness programs were created among patients through posters and education regarding self-hair removal. The percentage of self-hair removal significantly reduced from 41 to 27% from 2008 to 2011 and surgical site infection also significantly reduced **(Ng *et al*., 2013)**.

Laser hair removal is a suitable method for all skin types. It is a better treatment, and it does not show any long-term side effects. A longer wavelength is suitable for darker skin types. The short-term effects of laser hair removal show post-inflammatory, transient effects, erythema, hyperpigmentation, moderate pain, crusting and paradoxical hair growth in untreated areas **(Lim and Lanigan, 2006)**.

The effect of long-pulse alexandrite laser (spot size-18 mm, 3 ms pulse width and applied energy-755 nm) was evaluated on 322 patients between 1 to 12 months. According to the Fitzpatrick classification, the patients were classified by the basis of mechanism, efficacy, and possible side effects of treatment. The results of the study reveal that 80.8% of patients are independent of skin type, two patients' cases of hypopigmentation and 8 cases of hyperpigmentation post-treatment and no other complications noted. Hair removal through long-pulse alexandrite laser is effective, faster and provides patient comfort **(Tatlidede *et al*., 2018)**.

In microwave-heated hair removal, the waxes are unsafe to use and cause burn injury **(Angela *et al*., 2011)**. Therefore, a combination of capislow and long-pulsed Nd-YAG laser was compared with long-pulsed Nd-YAG laser alone. Both the methods are very well tolerated, and significant results were obtained for combined capislow with long-pulsed Nd-YAG laser **(Hisham *et al*., 2018)**. Imidiazolium-based ionic liquid were used in unhairing and fiber-opening processes. The ionic liquids are successfully employed in enhancing the activity of the unhairing enzyme **(Jaya Prakash *et al*., 2017)**.

Trimming, electrolysis, epilation (laser and light epilation are durable) show temporary results with repeated treatment. The improvements in advanced imaging, correct plan in radiation setting, and delivery strongly minimize the side effects **(Anderson *et al*., 2012)**. In addition, hair removal by clipper and depilation cream is more efficient than shaving, minimizing the surgical site infection **(Mohammed and Hamedan, 2016)**. The composition of sugar and lemon juice is used in removing unwanted hair. A gel was prepared using sugar (major), lemon juice (minor) and water. The hair is dissolved by applying the gel on unwanted hair and cover the absorbent layer **(Yasmin, 2012)**.

Hair removal methods on the surgical site were evaluated for 14 months. Patients who have used a razor (12.16%), depilatory cream (4.88%), and clipping (2.82%) showed severe infection. Skin injuries only occur while using a razor (10.81%) and also had erythema (12.16%). The razor and depilation method was responsible for 4.05% of rashes and 1.22% respectively. This study shows that the razor method had more complications than depilatory cream and clipping **(Chetty *et al*., 2017)**.

Depilation cream is effective, non-toxic, cheaper than shaving, self-administer and very safe on granulating wounds. It does not promote any bacterial growth. After the shaving, discomfort and itching occur during hair regrowth, but this is not in depilatory cream users. The growing hair tip is soft when using depilatory cream **(Powis *et al*., 1976)**. Complete hair removal (88.6%), skin infections (2.5%) and skin injuries (3.8%) were noticed using a depilatory cream. Complete hair removal (61.6%), skin injuries (27.9%) and skin infection (12.8%) were noticed using a razor. The wound infection and degree of skin injuries are strongly associated with hair removal, and these commonly occur after shaving **(Adisa *et al*., 2011)**.

Shaving has some disadvantages during hair removal, which are difficult to remove hair in body folds, axillae, public, perineal and scrotal regions. If the infection is caused, shaving may be extremely painful. It does not a skilful technique, which causes skin excoriated and offering an infection. The shaving equipment may not be sterile; it involves transferring pathogenic microorganisms from one patient to another. Comparatively, depilation is an effective method for operative sites **(Prigot *et al*., 1962)**. The complete hair removal efficacy 91% observed in depilatory cream users compared to razor users (62%). The highest skin injuries (29%) were noted among the razor group, and 4% was observed among depilatory cream. Postoperative wound infection was (3%) occurred in depilatory cream and 13% among the razor group **(Suvera *et al*., 2013)**.

A simple procedure was developed to determine the efficacy of depilatories. Two parameters, such as initial cross-sectional hair diameter and the time of maximum hair swelling were involved in this study. The graph was plotted against both the length and the width of swelling hair vs time. Identical hairs in identical solutions and the breaking times from one hour to more than ten days were maintained throughout the study. Nearly 500 depilatory creams have shown reproducibility in this method with a high degree of precision. The simplicity of procedure and the paucity of necessary equipment make it ideally suited for the rapid evaluation of formulation modifications (**Harvey and Roberta, 1968)**. Quantitative methods are very important for a cosmetic chemist to evaluate hair products properly and significant results. The tensile strength of hair has proved to be accurate, reproducible, and sensitive in this method **(Beyak *et al*., 1969)**.

A new thermo-mechanical analyzer was used to determine the relative activity of depilatory preparations. In this method, the change in length of a bundle of human hair was immersed in depilatory solutions. Then, the graph was plotted against hair length after the immersion vs time and getting two straight lines denoted as the time was inversely connected to the efficacy of depilatory preparations **(Francis, 1987)**. Chemical depilatories are available in the form of creams and lotions. It involve in the function of the degradation of keratin protein. The hair removal creams contain calcium hydroxide and potassium thioglycolate, which play an important role for dissolving keratin proteins **(Elumalai *et al*., 2013)** and weaken body hair roots. Toxic metals in hair removal products cause the skin to red, aggravated and blacken the skin. It causes hormonal cancer, sexual problems and rashes on sensitive parts of the skin **(Qurat and Iftikhar, 2013).**

Thioglycolate is an effective agent for enhancing transdermal drug delivery. Fluorescent model drugs were allowed through human skin by depilatory cream for 10 min. The structural alternations of stratum corneum, disruption of cellular integrity and focal detachment of superficial corneocytes, the disruption of the intracellular keratin matrix were observed **(Lee *et al*., 2008)**. *Bacillus subtilis* is a key ingredient in the degrading of feather keratin. Large amounts of ammonia, sulphite, and L-cysteic acid were detected in the fermented liquid. Gamma-glutamyl transpeptidase, peptidase T, serine protease, and cystathionine gamma-synthase are also were identified in this degradation process. The mechanism of keratin degradation by *B. subtilis* provided similarity with Streptomyces B221 **(Zhou feng *et al*., 2018).**

Ethanol (75%), DMSO, and peppermint oil (ethanolic solution) are act as an exchanger, and it significantly reduces the tear resistance time (6 to 3.5 min) of hair. At the temperature range from 20 to 37°C, the exchangers reduce the tear resistance time by about four times. Hydration in boiling water also reduced the tear resistance time by about 1.5 times. It provides less skin irritation and reduces pH from 12 to 11.5 in a formulation **(Hamid *et al*., 2013)**. Depilatory cream were prepared using herbal formulations contains *Prosopis cineraria* (leaves) which was used as a depilating agent, and thioglycolic acid was used as an accelerator. The prepared cream was elegant in appearance, and it reduced the quantity and quality of hair samples **(Verma *et al*., 2011)**.