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NANOFLOWERS- Recent Advances and Future Aspects for Multi Applications

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Abstract- Given their outstanding stability and increased efficiency, nanoflowers are garnering the interest of scientists and industry. Nanoflowers can be used in optoelectronics or sensors, catalysis, and solar cells. Nanoflowers have been identified to have a high potential for prospective applications in nanotechnology, such as sensors for hydrogen peroxide and glucose, as well as field emission features.

Nanoflowers have a variety of range of uses, including purification of enzyme, dye and heavy metal clearance from water, and gas presence detection utilizing nickel oxide. Recent research demonstrates the 3D structure of nanoflowers for improving surface sensitivity using a methodology called Raman spectroscopy. Based on its great face area with volume proportion & exceptional adsorptivity effectiveness on its husk, the nanoflowers are future of optimum advanced drug delivery system.

This article covers its extent on recent advances of nanotechnology related to nanoflower mainly in pharmaceutical field and future prospects of multi-application of nanoflower technology in field of pharma.

Keywords- Nanotechnology, Nano flower, Biosensor, Synthesis, Drug Delivery.

I. INTRODUCTION

Nanoflowers have a lot of potential when it comes to improving drug delivery systems and enzyme immobilization techniques. People have tried different methodology like microspheres, micro encapsulation, cross-transfer, and various binding and catalytic techniques to make enzymes work better, but now, hybrid/cross nanoflowers based inorganic enzymes given exceptional catalytic mechanism than independent enzymes or traditional immobilized enzymes. Similarly, now researchers are interested in the cation based [CDs] i.e. cyclodextrin or alginate oligosaccharide poliglusam nanoflowers as a system of drug delivery for 5-fluorouracil drug.
The study of nanoflowers is gaining popularity because they are easy to form using organic and inorganic materials or a combination of both. This helps to enhance the balance and efficient reactions occurring on facet. Nanoflowers act as organized or random form husk or leaflet, which provides a increased facial area in a compact organization. Because of this, they can be used in various applications like catalytic activity, biosensors, and drug delivery. [1-4]

In case to provide insight to the ongoing discussion following are the major topics that are tended to be discussed as per the flow of the subject. Majorly this review paper talks about Nanotechnology application and Nanoflower as a promising source of drug delivery system in micro drug delivery technology.

Various topics to be discussed are following-:
- About Nanoflowers
- Types of Nanoflowers
- Various Methodology for synthesis of Nanoflowers

- Organic and Inorganic Composition for synthesis of nanoflower
- Various recent advances in nanoflowers technology
- Future Aspects of Nanoflower technology system.

Thus, a comprehensive and detailed information with efficiently structured and thoroughly reviewed information is presented in this paper for better understanding and quick retrieval of knowledge required for nanoflowers and their synthesis and application.

Below table gives close details regarding different types of element used and their organic part and thus the target profile they tend to act on. [5-6]

	Metal ion	Organic material	Target
1	Cu	LALBA	-
		Urushiol oxidase	Catecholamines, phenols
		Carbonic dehydrates	
		Lipase	
2	Cu	Urushiol oxidase	Phenol
3	Cu	GOx and horseradish peroxidase	Glucose
4	Cu	Tripsin	HRPa, BSAb
5	Cu	HRPa	Hydrogen peroxide, phenol
6	Ca	a-Amylase	Cnp-G3c
7	Ca	Chitosan ascorbate	Hydrogen peroxide
8	Mn	Igg G	Ractopamine
		Anti-ractopamine ab	
		BSA	
9	Cu	DNA	Specific cell
10	Cu	Bovine liver catalase	Hydrogen peroxide

1. Types of Nanoflowers in Origin & Synthesis Method

In case to understand the diverse world of nanoflowers, it's important to note that their composition varies depending on the type of element used during their synthesis. This component plays a crucial role in determining their target profile, essentially deciding how these nanoflowers will interact with their surroundings.

Nanoflower	Method of synthesis	Function
Dual functional nanoflower	Green synthesis method: technique utilizes plant extract for synthesis.	Biosensor for detection of α-fetoprotein in human serum
Zinc oxide nanoflower	Precipitation method: reaction of liquid phase to form solid phase called precipitate and separation.	Biosensor for detection of amyloid in diabetics
Concanavalin A nanoflower	Mild one-pot process (Biomeneralization): Minerals like silicates, carbonates, phosphates, etc. are used for fabrication.	Detection of food pathogen using pH meter
Cationic cyclodextrin, chitosan and alginate-based nanoflower	Ionotropic gelation technique: cross-linking of oppositely charged ions i.e. anionic and cationic polymer.	Carrier for oral drug delivery of 5-FU for cancer treatment
Multifunctional and programmable DNA nanoflower	Liquid crystallization technique: transfer of mass from liquid to crystalline solid phase.	Drug delivery, intracellular imaging, cell targeting
	Precipitation method	Purification of soya bean peroxidase enzyme
	Hydrothermal method: in this method minerals are solubilized in hot water under high pressure.	Removal of heavy metals from water
	Facile one-pot method	Removal of dye effectively
Platinum cobalt nanoflower	One pot synthesis using precipitation technique	Enhance redox reaction
	Hydrothermal method and calcination	Gas sensing
Zinc oxide and silver nanoflower	Two-step hydrothermal technique	Substrate for surface enhance Raman scattering
Titanium dioxide nanoflower	One-step hydrothermal technique	Photoanode for dye sensitized solar cells
Gold nanoflowers	Microbial synthesis	Drug delivery, photo imaging and diagnosis

2. Different Method of Synthesis

Nanoflowers Synthesis Using [Cu] (II) Ions and Proteins

Since the advent of cross nanoflowers made up of [Cu] (II) ions and proteins, researchers are diligently studying their effectivity and stability. The focus has primarily been on copper-protein cross nanoflowers, leading to a relatively comprehensive understanding of their synthesis mechanism and applications.

To delve deeper, four different types of cross nanoflowers were manufactured using various biogenic molecules like LALBA, urushiol oxidase, carbonic dehydrates, and tri glycerol lipase. Nanoflowers which were made exhibited remarkable capabilities in detecting phenols and oxidizing catecholamines. In fact, their efficiency surpassed that of traditional free enzyme solutions, ranging from 95% to 650%. Several factors contribute to this increased efficiency: out of which few are

- The nanoflower's enhanced facet area, which rules out any limitation based on mass transferring.
- The enhanced interaction among enzyme subunits fixed inside nano particle molecule in nanoflower

Furthermore, researchers have explored the idea of multi-enzymatic hybrid nanoflowers by incorporating notatin (an oxidoreductase enzyme) and horseradish peroxidase enzyme. This breakthrough demonstrates the possibility of including two or more enzymes within a single hybrid nanoflower.[9]

Thus being greatly available in terms of extensive surface area captivity nanoparticles in nanoflowers system act as great source of drug delivery system. It is also possible that nanoflowers could also utilize more methods of micro manufacturing of particle to be used in nanoflower technology like 'nano sphere' & 'nano capsules'.

The matrix arrangement and capability of carrying both solid and liquid material through a bilayer system proves to be effective in case of nanoflower technology through above mentioned two models.

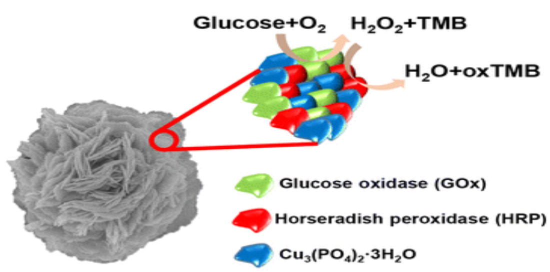

Fig.1- The enzyme based domino reaction of various enzyme embedded in cross nanoflower for detection of glucose [10]

Nano flowers Synthesis Using Calcium Ions and its Proteins

The researchers also put forward a hypothesis regarding the increased activity observed during the enzyme immobilization procedure. They suggested that the structure of the functional site was influenced by [Ca] ions, which occupy the regulatory sites of the ptyalin enzyme. Normally, ptyalin enzymes are inactive when calcium ions are absent, as the due to restraint of operative site. However, when calcium ions are present, they bind strongly to the enzyme and activate it.

This experiment utilized ptyalin, which is known to exhibit allosteric phenomena. By investigating the formation of a cross nanoflower using [Ca] ions and ptyalin, the researchers were able to observe the activation and strong binding of calcium ions to the enzyme.

In summary, while most nanoflowers are synthesized using calcium ions, the utilization of calcium ions in the creation of nanoflowers has also been explored. The unique flower-like morphology of the calcium-based nanoflower was confirmed, and the researchers proposed a possible explanation for the increased activity observed during the enzyme immobilization. The presence of calcium ions was found to affect the structure of the functional site of ptyalin, leading to its activation. These findings contribute to our understanding of nanoflower synthesis and the role of calcium ions. [11]

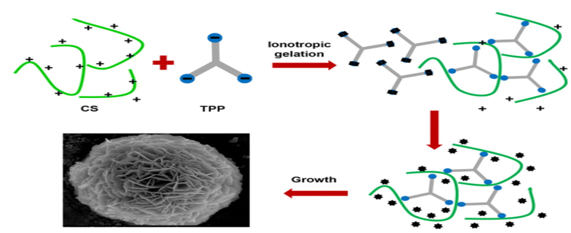

Fig.2- Simplified method of formation of chitosan ascorbate–(Ca) ion cross nanoflower. [12]

Nanoflowers Synthesis Using Manganese (II) Ions and Proteins

The specific enzyme used in this experiment is called ptyalin, which exhibits allosteric phenomena. Essentially, this means that without the presence of manganese ions, α-amylases remain inactive as the operational site is restrained. However, when calcium ions are introduced, they bind to the allosteric sites of ptyalin, thereby influencing the structure of the operational site. The researchers utilized this concept during the formation of the cross nanoflower, incorporating calcium ions alongside ptyalin. Interestingly, the manganese ions exhibited activation properties and formed strong bonds with ptyalin. This interaction played a crucial role in enhancing the overall activity of the nanoflower. By understanding the influence of calcium ions on ptyalin, researchers have uncovered a remarkable connection between nanomaterials and enzymatic activity.

Exploring different elements, such as copper and calcium, in the synthesis of nanoflowers allows scientists to push the boundaries of this field even further. By expanding their understanding of the factors that impact the properties of nanoflowers, researchers can unlock new potentials for various applications. From medicine to energy, nanoflowers hold immense promise in shaping the future of science and technology.[13]

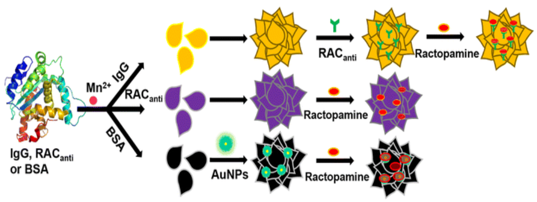

Fig.3- Basic representation of the formation of (Mn)-based cross nanoflowers as a biosensor for the spotting of ractopamine. [14]

Nanoflower Formation Using [Cu] (II) Ion and DNA

As Deoxyribonucleic acid is very much soluble in hydrated medium and has a high proportion of nitrogen in its proteinic structure, acting as a great source for forming of cross nanoflowers by combining ions from metal sources. The DNA based cross nanoflower framing was bonded with the Förster resonance energy transfer also known as electron energy transfer (EET) method to produce images of great resolution of cells or to use for tracing of drug delivery systems. Basically, researchers formed a place in DNA arrangement for combining of drug and fluorescence producing dye like (Fluorescein amidite, Cyanine3) thus they formed a cross nanoflower attached to the distorted DNA by a technique known as unidirectional nucleic acid replication with the ions of metal entity. Thus, able to produce a better resolution image based on electron energy transfer (EET) between the fluorescence producing dyes using high-wavelength light source, which does not affect the cells. Thus through the tracing of light path from the source we can judge the route and efficiency of drug delivery pathway accordingly. [15]

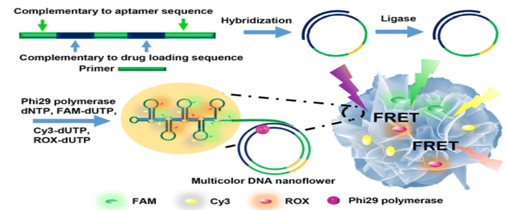

Fig.4- Sequence-individualistic auto-assembly of multi colour EET (electron energy transfer) DNA cross nanoflower [16]

II. RECENT ADVANCES & APPLICATION OF NANOFLOWERS IN PHARMACEUTICAL SECTOR

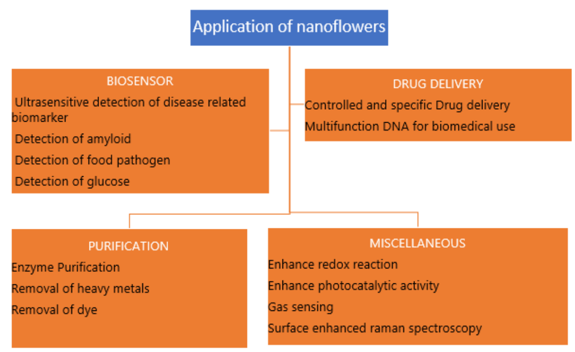

Fig.5 Flowchart showing application of application nanoflower **[17]**

1. Recent Advances in Nanoflower Technology

Biosensors as Analytical Devices

Biosensors are devices for analysis of origin of biological molecules that are largely used to identify different variety of disease and health disorder that are comprised of transducing signal based methodology attached with a detection unit which process out various signal based on analysed result. [18]

Binary Functionalism Approach Applied in Nanoflower for Extremely Sensitive Extremely Sensitive Spotting of Disorder Bio Marker

Sensors based on principle of calorimetric are thoroughly used as a biomarkers for various disorder and health problems like diabetes, Parkinson's disease, etc. Protein based bio markers are readily used in cases were samples are in low concentration and thus the scenario of evaluation of result become difficult to analyse. Variation in amplification signal was a extensively utilized technique in order to increase the magnitude of sensitivity of calorimetric bio markers. ELISA (enzyme-linked immunosorbent assay) involving southern blotting and western blotting technique was carried out experimentally by such calorimetric bio marker method for optimum analysis.

The binary functioned nanoflower were prepared by a green synthesis ny utilizing plant extract method in which nano scale metal particles utilizing tetrameric biotin binding protein known as streptavidin, as a source of biologically originated recognition system unit and [HRP] enzyme also known as horseradish per oxidase enzyme obtained from horseradish roots, compositely utilized as a signal modular and amplifier unit in photometric analysis through combing of [Cu] ions in Phosphates which are buffered at room temperature and streptavidin - Horseradish peroxidase-$Cu_3(PO_4)_2$ nanoflower was obtained. These nanoflower mainly established association between the biotin binding protein and the [Cu] ion.[19] Streptavidin has an inclination towards both for biotin binding protein and Horseradish peroxidase acting as a catalyst for catalysis of the oxidation of [3,30,5,50]-tetra methyl benzidine (TMB) in a blue substance in the presence of hydrogen peroxidase. Therefore, the introduction of SA and HRP demonstrated that dual functioning nanoflowers helped in the development of a highly efficient ultrasensitive colorimetric analysis technique for the determination of alpha-fetoprotein.[20]

(Con A) Lectin Based Nanoflower for Spotting of Escherichia Coli in Food

Escherichia coli infection is majorly caused by spoiled food. Escherichia coli is a dangerous microorganism affecting human health by infecting RBCs and causes gastrointestinal irregularity and vomits. Therefore, bio sensors are being prepared to detect this harmful microorganism. (Con A) is also known lectin and is a carb-bounding protein. GOx is an enzyme that is immobilized on the (Con A) atom by help of metal ions i.e. (Ca ion).This entity of (Con A, GOx and Ca+ ions) were extensively utilized for formation of bio sensor help in spotting of Escherichia coli.[21]

Zinc Oxide Nanoflower for Amyloid Detection

Amyloids are aggregates of proteins that fold and stick together to form fibrils. Amyloids are detected in diseases such as amyloidosis and neurodegenerative diseases such as Parkinsonism, Alzheimer's disease, etc. Fluorescent dyes have been widely used to detect amyloid, but the main disadvantage was photo stability and low intensity. Detecting amyloid at very low concentrations is difficult and therefore a sensitive technique for detecting amyloid is required. Thioflavin T is the thiazole salt Benzo (dye).

Thioflavin T is often used to visualize these folded proteins, i.e. H. Amyloids that bind to the beta sheet show increased fluorescence and red shift. Thioflavin T bound to the amyloid insulin beta sheet showed weak fluorescence. This problem could be solved by fabricating a biosensor with zinc oxide nanoflowers and thioflavin-T dye adsorbed on its surface and grown on a silver nanofilm coated on the glass surface. It showed increased fluorescence when detecting insulin amyloid.[22]

In Drug Delivery System

Cyclodextrins are extensively used as carriers of less water-soluble BCS drugs of classes 2 and 4. 5-Fluorouracil is a readily water-soluble anticancer drug that is widely used for treatment of cancer in GIT since past many years. The major drawback of 5-fluorouracil is its less half-life and various toxic side effects. Therefore, oral administration of this drug requires some properties such as improved biocompatibility, controlled drug release pattern, reduced drug toxicity, and increased therapeutic efficacy.[23] For oral administration of the drug, cationic b-cyclodextrin is used as a carrier due to its unique structure and lack of hydrolysis in the stomach.

When cationic b-cyclodextrin is conjugated with 5-fluorouracil, the drug is delivered specifically to the colon. The freeze-drying method was used to prepare the cation inclusion complex beta-cyclodextrin and 5-fluorouracil. Nano petals prepared from sodium alginate and chitosan were used for controlled and long-term drug delivery.[24]

Microfluidic Paper-Based Analytic Devices (Mpads) Biosensor for Detection of Glucose

The biosensor was fabricated by Zhu (2017) for sensitive and visual glucose detection. Novel paper-based microfluidic analyzers (mPADs) were fabricated from a hybrid nanocomplex composed of two enzymes, glucose oxidase (GOx) and horseradish peroxidase (HRP), with inorganic copper(II) phosphate nanocrystals embedded in the sensing areas using a wax printing technique.[25] The resulting complex resembled the structure of a flower, allowing co-immobilization of GOx and HRP in a biocompatible environment. The prepared nanoflowers facilitate the transport between the enzyme and its substrate and also preserve the activity and stability of the enzymes. The biosensor enabled rapid and sensitive detection of 0-glucose. Concentration range 1 to 10 mM with a limit of detection (LOD) of 25 µM.[26]

Multifunctional and Programmable DNA for Biomedical Application

DNA is an exogenous biocompatible material that is naturally soluble in water. DNA is used as a building block to develop various DNA nanoparticles with inherent functions for biomedical applications. The possibility of group programming and automated and controlled combination makes DNA nanostructures more useful biomedical nanotools. Assembling DNA strands to create a DNA nanostructure is laborious and expensive. DNA hybridization created scratches in the DNA backbone, which may reduce the biostability of conventional DNA nanostructures. Pinched DNA strands, hydrogen-bonded DNA nano assemblies, often have loose internal structures, which is problematic in therapeutic applications and biological imaging. Conventional DNA nanostructures lose their structural properties upon denaturation.[27] DNA nanoflowers make them ideal candidates for drug delivery. In this regard, color atoms ideally calm related entities (e.g. double-stranded templates 50-GC-30 or 50-CG-30 for anthracycline sedatives in this convention) and aptamers (sgc8c for CEM and HeLa cell lines) are grouped into format templates to functionalize nanoflowers as "smart nanotherapeutics" that are unique can carry anti-cancer medications. The DNA nanoflowers showed obvious special features such as high biocompatibility, adaptive programming, resistance to physiological impedance, as well as a high drug stacking limit of and traceability.[28]

III. FUTURE ASPECTS OF NANOFLOWERS

Nanoflowers are proven to be beneficial in various fields, let it be purification of water, anode unit in photometry, improved efficiency of reduction and oxidation reaction by improved action of enzymes through catalysis and efficient and enhanced activity of catalysis in photometry. Nanoflowers show promising results in advanced drug delivery system especially for drugs in available in low concentration, to deliver drug across various physiological barrier system, active and passive drug targeting in micro drug delivery system. We are already aware of the fact that nanoflowers has various useful application such as biosensor, bio catalyst and other analytical devices, nanosphere and nano capsule technology [29-31]. In near future nanoflower technology could be proven to be a great source of photodiode. Nanoflower could also be developed single dispersing system and enhancing optical analysis and suitable for various other applications. Different metallic component composition in metal alloy and core shell production can be developed with advance techniques.

In addition, future nanomaterials research may include the combination of multiple ultra micro structured materials, such as graphite and various oxides wrapped in structure of nanoflower, Nickel containing nanoflower doped with nitrogen and Cu containing nanoflower, single layered of Platinum nanoflower on single-walled carbon nanotube membranes and non crystalline structured nanotubes. Nanoflowers of hybrid carbon modules in production of carbon fibre with more enhanced strength. [32-33] Thus nanoflower are going to be major player playing crucial role in various fields like water filtering and water treatment, aroma therapy, pollutant absorbent by their extensive use in catalytic converters in vehicles as adsorbate and will be largely used as Coating material in various industries like pharma, automobile etc.

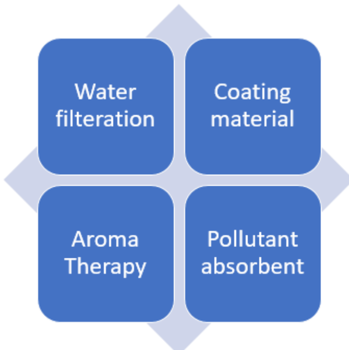

Fig.6 Future scope of nanoflowers [34]

IV. CONCLUSION

All the scientific progress made in context to nano materials which are formed in shape of flower is seen as source of immense achievement, combining various studies for methods regarding synthesis, drug distribution, drug delivery, intra cellular targeting of various drugs in low concentration & enhance catalytic properties, also extensively used in preparation of sustained release drug delivery system. Different nanoflowers based on elemental in synthesis are formed by various methods involving different approaches like: vapourised depositories, process including hydrothermal approach, chemical deposition through vaporised model, solvent-gel, natural extract (green synthesis), displacement through galvanic models, MAS and various other techniques like solvent emulsification evaporation and salting out.

In essence to research made from few decades the advancement in nanoflower technology has surely made an upgrade in all aspects, and also the number of published articles and paper are increased in significant number compared to previous stats. Concluding the matter we can say that, cross organic-inorganic nanoflowers have influenced many researchers working in field of nanotechnology to objectively work for development of nanoflower and utilise their full potential in terms of usage in drug delivery system and indeed various findings have been already published in various sources. Research in this field is actively motivated by purpose of personal virtue, simple method and quick result oriented study with less rigorous work. If such progress is continued is more accelerated pace then surely it will result in a advanced future of nanotechnology and will lead to solution for drug delivery system and lead to betterment of their efficacy. In addition to these nanoflower technology will also lead in advancement of used as carriers for radio nucleotides and for diagnostic purposes in nuclear medicine drug delivery.

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Rigorous efforts to achieve the target backed by self-confidence are the key to success. Well planned efforts in the right direction surely give us success. It is a moment of gratification and pride to look back with a sense of contentment at the long travelled path, to be able to recapture some of the fine moments, to be able to thank the infinite number of people, some who were with me from the beginning, some who joined me at some stage during the journey, whose rally round kindness, love and blessings has brought me to this day. We wish to thank each one of them with all our hearts. From the inner core of our heart, we express profound sincerity and gratefulness to our adored Dr. Rajesh Oswal, Principal, Genba Sopanrao Moze College of Pharmacy Wagholi. We are highly honoured to have worked under such a multitalented personality. This dissertation was possible mainly because of the opportunity presented to me by my guide Prof. Nikita Rane, Department of Pharmaceutics GSMCOP, Wagholi. We are very grateful to her for showing us path and vision that research is all about enthusiasm, keen observation, creativity, hard work and patience. Her heartily criticism and timely suggestion on research induced a continuous source of inspiration for us throughout the period of study, We are also thankful to our librarian as she has helped us to find such from our various science journal on internet. Working together with them allowed us to expand our mind and grow into a better pharmacist. We would like to convey our honest unforgettable regardful thanks to all teaching staff for their constant and valuable support, regular observations, kind co-operation, boundless enthusiasm, affectionate encouragement. Last but not least we would like to thank God lord Shri Ganesh and goddess Sharda for all blessings.

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