



“COMPREHENSIVE STUDY OF BIOCATALYST”

GUIDED BY:MS. JYOTI NAKUM

PRESENTED BY:AASHOON J. SHAH

DEPARTMENT OF INDUSTRIAL CHEMISTRY PARUL INSTITUTE OF APPLIED SCIENCES, VADODARA

PARUL UNIVERSITY

ABSTRACT

Green chemistry is the branch of chemistry that involves tools techniques and technologies. It is helpful to chemists and chemical engineers in research, development and production, for development of more eco- friendly and efficient products which may also have significant financial benefits. It is going to now become an essential tool in the synthetic chemistry. It is a new way of looking at organic synthesis and the design of drug molecules, offering important environmental and economically advantages over traditional synthetic processes. The recent interest in green chemistry has posed a new challenge for organicsynthesis in that new reaction conditions need to be found which reduce the emission of volatile organic solvents and the use of hazardous toxic chemicals. They improve selectivity, reduces reaction time, and simplifies separation and purification of products than the conventional methods.

OBJECTIVE

Green chemistry is a crucial component of organic synthesis chemistry and is vigorous in protecting the environment from hazardous and damaging catalysts. Lemon juice has the potential to be involved in a number of organic transformations in fruit juice catalysed chemistry, which is an essential component of green chemistry. The use and significance of lemon juice in synthetic organic transformation as well as the creation of various types of nanoparticles and catalysts are summarised in this review article (from 2011 to 2020). This review article can aid scientists in planning the most cost-effective and environmentally friendly way to synthesise different scaffolds, small

molecules, nanoparticles, and catalysts. Crucial component of organic synthesis chemistry and is vital in protecting the environment from hazardous and damaging catalysts.

INTRODUCTION

Easy and environmentally friendly synthetic methods are significant challenges in organic synthesis. For organic chemists, developing gentle and safe routes for the synthesis of biologically active molecules has become a source of inspiration and motivation [1]. Fruit juice that is readily available serves as the creation of a biocatalyst satisfies nearly all the requirements of green chemistry, which attracted the pursuit of knowledge [2]. The majority of fruits are often inexpensive, readily available, and can be simple to extract. This review's objective is to examine current elements of organic fruit juice transformations [3].

Chemists have taken an interest in the use of naturally occurring fruit juice in organic synthesis, particularly from the perspective of green chemistry. This paper outlines the various synthetic processes in which fruit juice has been used as a biocatalyst. Organic synthesis frequently makes use of the fruit juices of lemon, pineapple, tamarind, *Acacia concinna*, *Sapindum trifoliatum*, and coconut [4]. A number of reactions, including the three-component synthesis of dihydropyridines, triazoles, Schiff bases, and bis-, tris-, and tetra indoles, were found to be catalysed by lemon juice. Dihydropyridines and bis-, tris-, and tetra indoles have each been synthesised using pineapple juice and tamarind juice, respectively [5].

The public is now much more aware of the potentially dangerous compounds utilised and created during the chemical reactions, which led to the development of the "green and Bearable chemistry is now a thing. The primary goal of this idea is to produce the finding slick, clean routes is important techniques to lessen the use of harmful substances Reagents, solvents, challenging reaction circumstances, and pricey catalysts [6]. The public is considerably more aware of the dangerous compounds used and generated during the chemical industry with the dawn of the twenty-first century [7].

Reactions, leading to the idea of "green and Sustainability in chemistry has developed. The primary goal of this approach is to increase the smooth, unpolluted routes, and to discover innovative methods to decrease the use of harmful severe reaction conditions, solvents, and costly catalysts [8].

The testing of a new catalyst in an environmentally friendly manner has been far more crucial in modern times. The common method of synthesising huge amounts of harmful waste are produced by chemicals similar by-products [9]. These dangers show that it must be crucial to create procedures which abide with environmental standards [10].

The development of greener, eco-friendly techniques that use alternative reaction media instead of toxic, expensive catalysts or volatile, dangerous solvents like benzene, toluene, and methanol, which are frequently used in organic synthesis, has been a major focus of modern organic research. Many organic changes have been done in water recently [11].

It is a special solvent since it is widely accessible, affordable, harmless, secure, and ecologically friendly. Applications for aqueous extracts of various fruit juices have advanced quickly [12]. Fruit juice's increasing popularity is mostly due to its biocatalysts, environmentally friendly nature, non-hazardous nature, and cost-effectiveness [13]. Numerous organic reactions have been documented in the literature using natural catalysts such as clay, phosphates, gold, and animal bone. Recent years have seen a lot of interest in chemical processes employing plant cell cultures and plant parts as biocatalysts [14].

This growing interest is caused by the enzymatic reactions' extensive biotechnological potential. The employing edible plants for biocatalytic reactions, plant extracts from plant roots, plant tubers, and plant leaves can be used in a variety of chemical processes [15]. Fruit juice, which also occurs naturally and has been utilised as a biocatalyst in organic synthesis. Fruit juice is already consistently utilised in organic for diverse selective transformations of simple and complex compounds, homogeneous catalysts are synthesised. The goal of this review is to provide an overview of the benefits of various fruit juices with a focus on current synthetic uses literature coverage is up to 2013 [16].

The growing interest in green chemistry has created a new challenge for organic synthesis since novel reaction conditions must be discovered that limit the use of dangerous toxic compounds and the emission of volatile organic solvents [17]. Compared to the traditional procedures, they increase selectivity, shorten reaction times, and make product separation and purification simpler [18].

Green chemistry's inception is generally seen as a response to the desire to lessen the harm that man-made materials and the manufacturing processes that create them cause to the environment [19]. A cursory review of green chemistry concerns over the previous ten years reveals numerous techniques that safeguard public health and the environment while still being profitable in that it welcomes the same creativity and innovation that have always been at the heart of classical chemistry, green chemistry is similar to traditional chemistry [20]. By using safe raw materials during the production process, a threat can be removed more quickly and easily in accordance with the principles of green chemistry [21].

Because they are poisonous and flammable, organic solvents like benzene and chlorinated hydrocarbons, which are used in organic synthesis operations, have wreaked havoc on the environment [22]. In comparison to conventional solvents, solvent-free processes typically require shorter reaction durations, smaller reactors, simpler and more efficient build up procedures, more improved selectivity, and simpler separations and purifications. Chemists have taken an interest in the use of naturally occurring fruit juice in chemical synthesis, notably from the perspective of green chemistry [23]. This demonstrates how fruit juice can be used synthetically in a variety of processes as a biocatalyst [24].

One of the most recent issues facing organic chemists is the development of non-hazardous synthetic techniques for organic synthesis [25]. Growing environmental awareness necessitates the creation of efficient, cost-effective methods where even less dangerous outputs are undesirable. Since the majority of solvents are either hazardous or flammable and significantly raise the cost of a synthesis, solvent-free organic reactions have grown in favour in recent years [26]. Compared to conventional solvent-based processes, these solvent-free ones often require shorter reaction durations, smaller reactors, simpler and more effective work-up procedures, higher selectivity, and simpler separations and purifications [27].

Green chemistry as a concept and its applications in synthetic organic chemistry have become important solutions for the creation of safer and cleaner chemical processes. For this objective, numerous approaches and procedures have been created [28]. Environmentally friendly synthetic techniques have attracted a lot of attention recently, and certain protocols without solvents have been created the mixing of solid anilines and solid benzaldehydes to produce a variety of benzylideneaniline derivatives [29]. described the use of microwave

irradiation for the solvent-free, clay-catalysed synthesis of imines and enamines. The creation of ecologically friendly solvents and reaction conditions has received significant attention from the chemical industry as well as from academic study [30].

APPLICATION OF SCHIFF BASE

Imines are compounds with an azomethine group ($\text{HC}=\text{N}$), often known as Schiff bases, and their typical formula is $\text{R}_3\text{R}_2\text{C}=\text{NR}_1$. They are the abbreviated by goods of aldehydes or ketones, and Hugo Schiff originally identified them in 1864. Schiff was the first to explain how to make Schiff bases synthetically, which involves condensing primary amines with carbonyl compounds during azeotropic distillation while also eliminating water. The structural similarities between these compounds and biological molecules found in nature, the ease with which they can be produced, and the flexibility with which they may be produced have all contributed to the interest in them.

They serve as a well-known intermediary in the production of several other derivatives, including azetidinone, thiazolidinone, formazone, arylacetamide, and metal complexes. One of the most potent classes of chemicals, Schiff bases have a wide range of biological uses, including antitubercular, anticancer, antibacterial, anti-inflammatory, antifungal, antitumor, diuretic, insecticidal, herbicidal, anthelmintic, anti-HIV, antiproliferative, anticonvulsant, and antihypertensive properties in addition to antiparasitic activities. For more than a century, the Schiff's base derivatives have been thoroughly studied and used in a variety of fields, such as magnetochemistry, non-linear optics, photophysical research, catalysis, materials chemistry, chemical analysis, and the absorption and transport of oxygen.

New environmentally friendly catalysts and techniques have been researched that are both economically and technologically possible because of their advantageous qualities, environmental demands, and strong interest in the development of green chemistry. For the synthesis of Schiff bases, the current work also uses certain natural, cheap, and eco-friendly catalysts, such as lime juice, orange juice, and butter milk.

GLASSWARES, MATERIALS, AND OTHER ASSEMBLIES:

All the glassware used in the dissertation project were clean, oven dried and washed thoroughly after and before use with detergents and solvents. The

Glassware used were of Borosil, Glassco, Fisherbrand company.

Glass Apparatus: Beakers, Round Bottom flask, magnetic beads, measuring cylinders, glass rod, conical flask, spatula, etc

Other Apparatus: Weighing balance, hot plate magnetic stirrer, Ph meter, Thermometer

Materials: Aniline, Benzaldehyde, Glacial acetic acid, Methanol, Orange juice, Limetta juice, Butter milk

SYNTHESIS METHOD

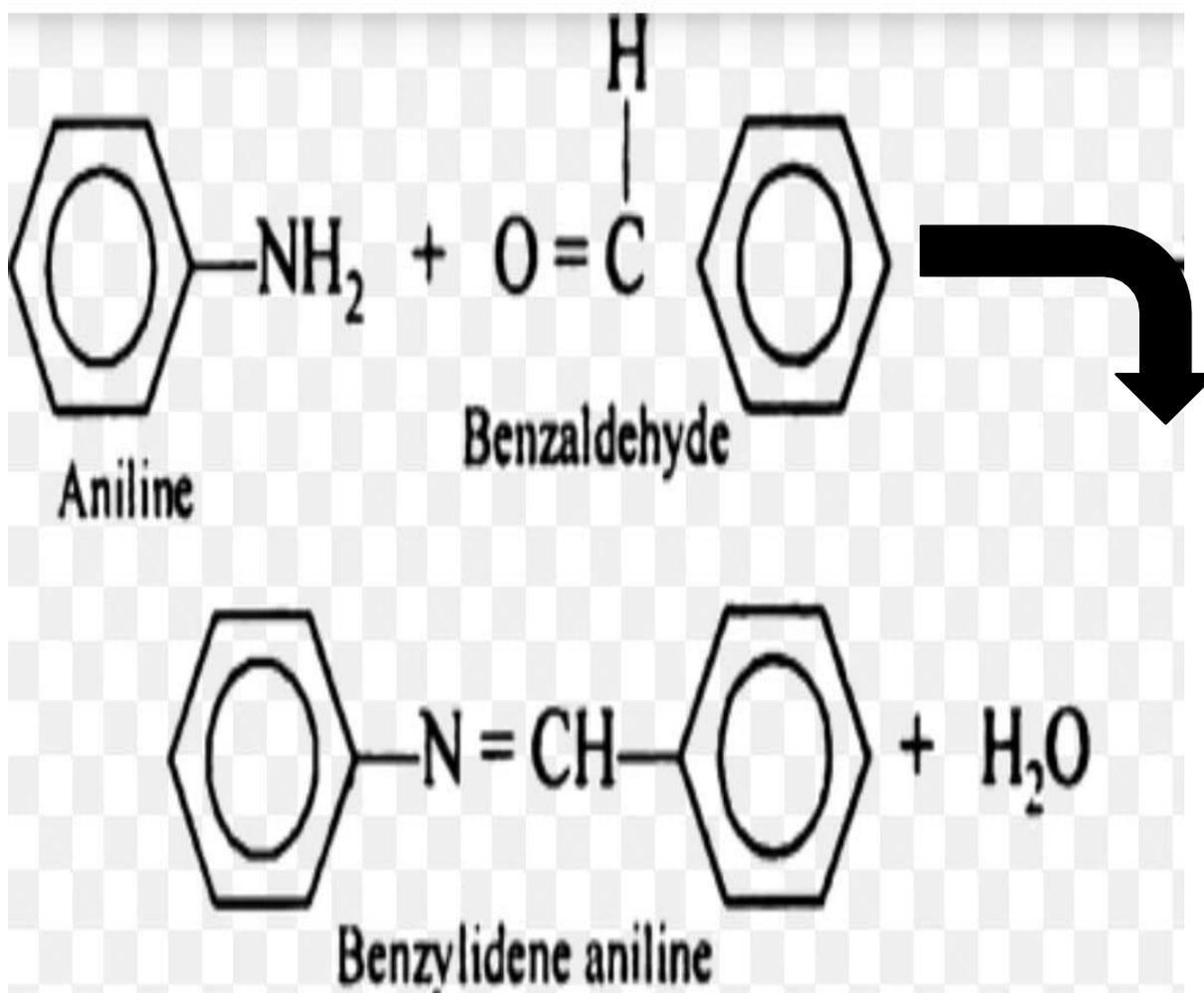
1) Formation of catalyst: Orange and limetta fruits were purchased locally for the preparation of the catalyst. limetta was then squeezed into a fruit juice and filtered through cotton to get liquid juice. While peeling the luscious orange fruits with a knife and Fruit pieces were put through a juicer to produce semisolid juice. bulk, which was later cotton-filtered to obtain liquid juice to act as a catalyst.

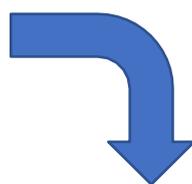
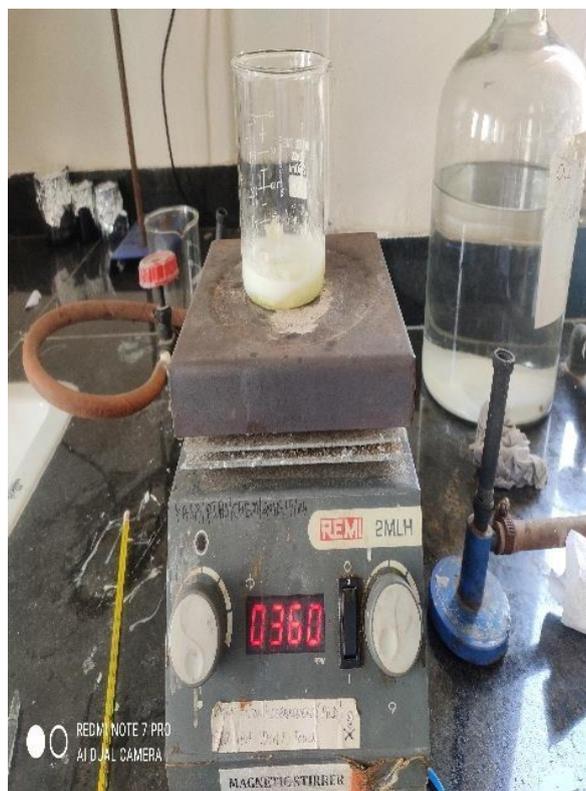


Butter-milk

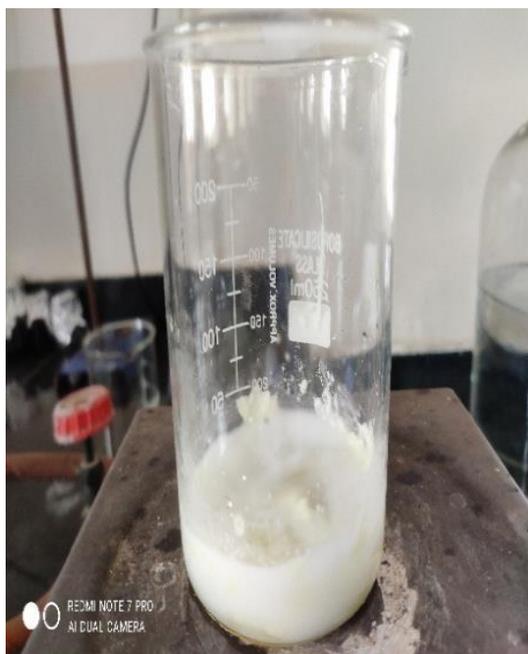
2) Orange juice is used in the Schiff base synthesis. The equaliser quantity of aniline and benzaldehyde (1 mol) (1 mol) was collected in various beakers. These reaction combinations contain orange juice, a bio-catalyst, was added in varying amounts. quantities (1 ml, 2 ml, 3 ml, 4 ml, 5 ml, 6 ml, and 7 ml) then stored 15 to 20 minutes. Each reaction mixture was further stirred. at room temperature, for 10 to 15 minutes. Solid pale- yellow colour obtained after the reaction was finished, a crude product appeared. rinsed in distilled water and was made pure by recrystallization using the least amount of methanol possible. The Repeat the process with limetta juice and a buttermilk.

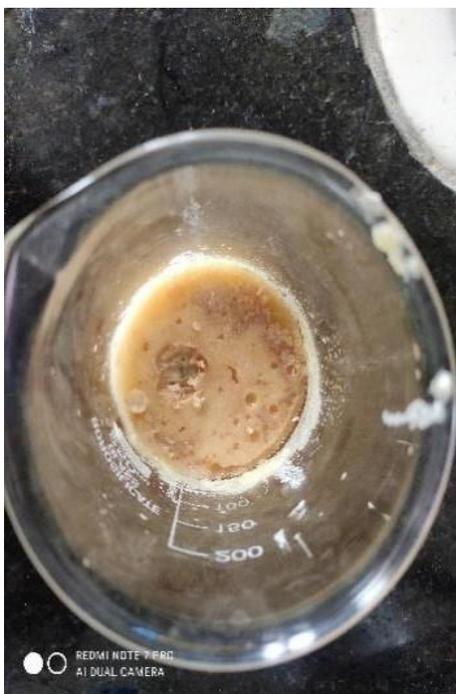
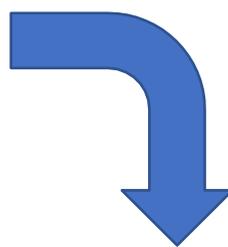
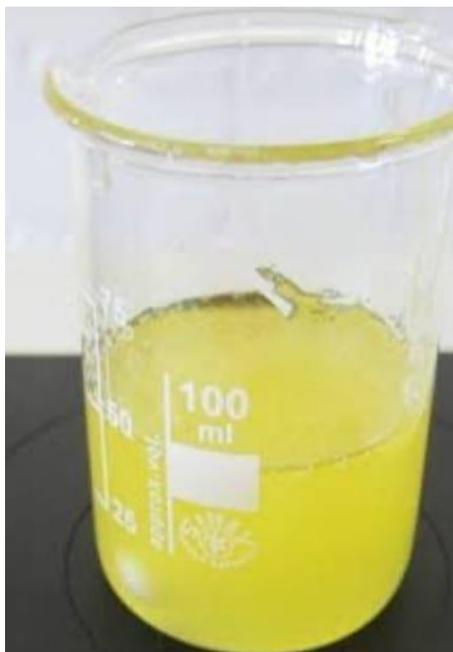
Schiff base reaction:



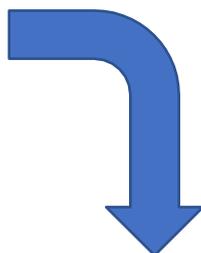


Schiff base reaction (butter milk catalyst)





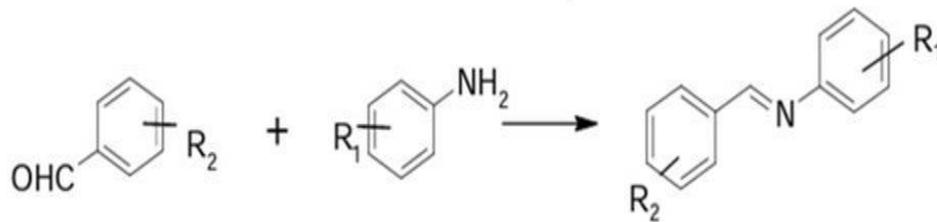
Schiff base reaction (orange juice catalyst)



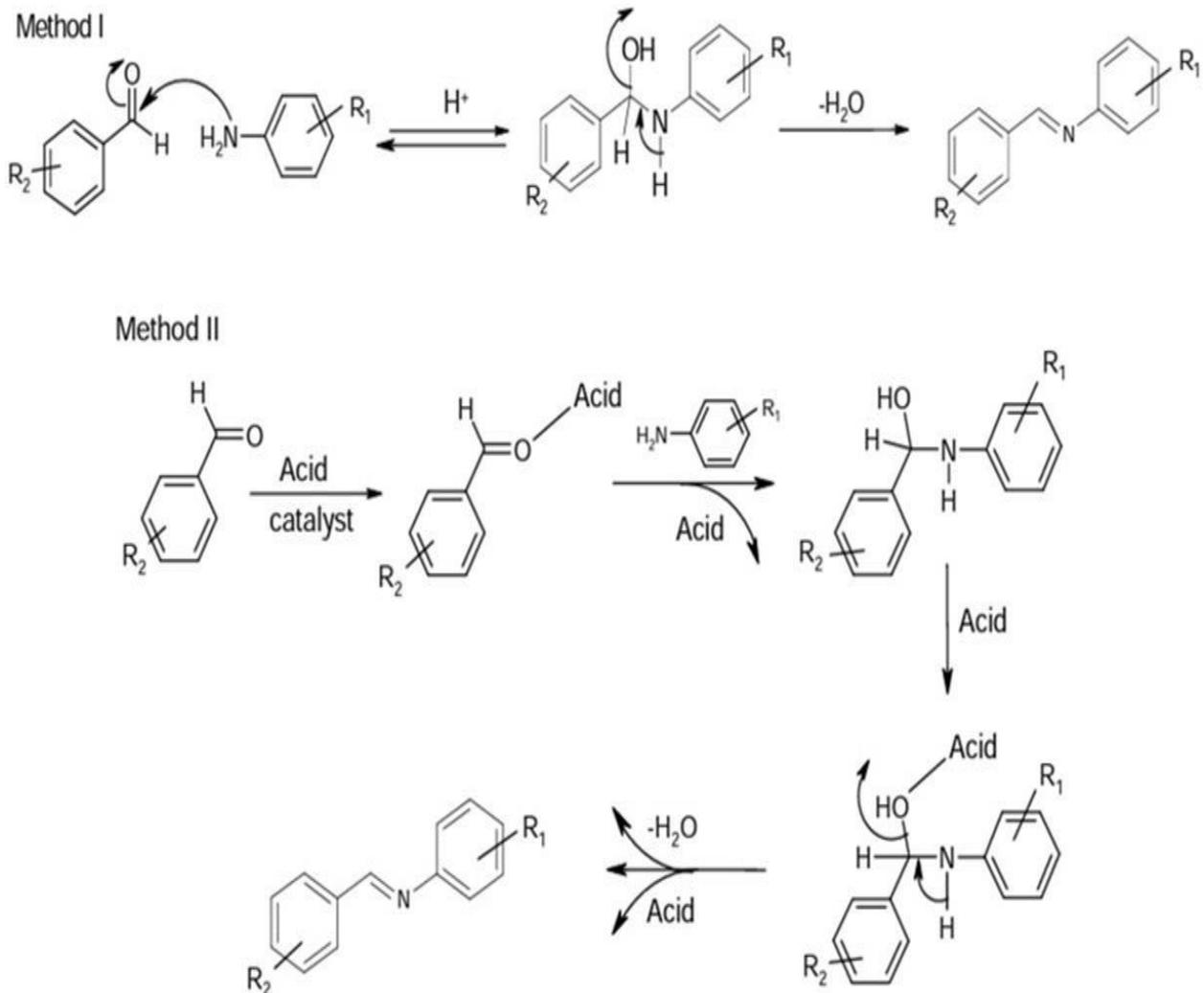
Schiff base reaction (limetta juice catalyst)

RESULT AND DISCUSSION

Scheme I : Schiff bases synthesis



Scheme II: Mechanism for acid catalyzed Schiff base synthesis



If we concentrate on the process that turns amines and aldehydes into Schiff bases, a primary amine's nucleophilic attack on carbonyl carbon provides Dehydration of a hydroxyl molecule results in Schiff bases are formed in the subsequent phase. depends significantly on the rate of water removal from the combination for reactions.

The amine combines with the aldehyde or ketone in the first phase of the process to create the unstable addition chemical known as carbinolamine. Carbinolamine then loses water in the second stage via acid- or base-catalysed pathways. Being an alcohol, carbinolamine prefers to undergo acid-catalysed dehydration. The good electrophilic and nucleophilic characteristics of the carbonyl and amine groups, respectively, are observed to make the condensation between a carbonyl molecule and an amine leading to the creation of Schiff bases an easy reaction.

If the reaction takes place in an acidic solvent medium, carbinolamine production and protonation happen in the same step, whereas in an acid-catalysed process, protonation comes first.

Obviously, fruit juices were used in this work to protonate heteroatoms throughout the organic transition. Citric acid, among other organic acids, is the main component in many fruit extracts. Other organic acids found in fruit juices include tartaric acid, malic acid, oxalic acid, succinic acid, and amino acids. Because of the acidic nature of fruit juices, these organic acids are to blame. They also give a favourable pH to catalyse the condensation reaction, which is utilized to create Schiff bases.

Effect of catalyst loading on the product yield

Sr. no.	Amount of catalyst (ml)	Product yield obtained with orange juice	
		Product yield (gm)	Percentage yield %
1	1 ml	8.023 gm	80.23%
2	2 ml	8.421 gm	84.21%
3	3 ml	8.795 gm	87.95%
4	4 ml	8.884 gm	88.84%
5	5 ml	8.521 gm	85.21%
6	6 ml	8.123 gm	81.23%
7	7 ml	8.256 gm	82.56%

Table:1 Product yield obtained with orange juice

Sr. no.	Amount of catalyst with limetta (mousambi juice) (ml)	Product yield with limetta (mousambi juice)	
		Product yield (gm)	Percentage yield %
1	1 ml	8.234 gm	82.34 %
2	2 ml	8.532 gm	85.32 %
3	3 ml	8.674 gm	86.74 %
4	4 ml	8.941 gm	89.41 %
5	5 ml	8.527 gm	85.27 %
6	6 ml	8.425 gm	84.25 %
7	7 ml	8.342 gm	83.42 %

Table:2 Product yield with limetta (mousambi juice)

Sr. no.	Amount of Catalyst (ml)	Product yield with butter milk	
		Product yield (gm)	Percentage yield %
1	1 ml	8.325 gm	83.25 %
2	2 ml	8.549 gm	85.49 %
3	3 ml	8.726 gm	87.26 %
4	4 ml	8.954 gm	89.54 %
5	5 ml	8.741 gm	87.41 %
6	6 ml	8.513 gm	85.13 %
7	7 ml	8.412 gm	84.12 %

Table: 3 Product yield with butter milk

Entry	Product		Time min	Yield %	m.p. °C		Ref.
	R ₁	R ₂			Found	Reported	
1	H	H	60	89	65-67	66-68	31
2	H	4-OH	60	89	191-194	--	--
3	H	2-OH	45	90	47-49	--	--
4	H	4-OCH ₃	90	93	84-85	80-82	31
5	H	4-N(CH ₃) ₂	90	97	78-82	--	--
6	H	4- NO ₂	60	85	65-70	--	--
7	4-CH ₃	H	25	90	112-115	--	--
8	4-CH ₃	4-OH	50	94	210-212	--	--
9	4-CH ₃	2-OH	30	94	102-105	--	--
10	4-CH ₃	4-OCH ₃	45	92	93-95	--	--
11	4-CH ₃	4-N(CH ₃) ₂	30	84	92-95	--	--
12	4-CH ₃	4- NO ₂	50	83	121-125	128-130	31
13	4-OCH ₃	H	20	96	154-157	--	--
14	4-OCH ₃	4-OH	42	93	215-220	--	--
15	4-OCH ₃	2-OH	15	91	145-149	--	--
16	4-OCH ₃	4-OCH ₃	30	84	142-145	--	--
17	4-OCH ₃	4-N(CH ₃) ₂	55	93	132-135	--	--
18	4-OCH ₃	4- NO ₂	30	100	68-72	--	--
19	4-Br	H	120	82	61-62	63-65	31
20	4-Br	4-OH	90	87	169-170	--	--
21	4-Br	2-OH	65	88	175-177	--	--
22	4-Br	4-OCH ₃	120	no reaction	--	--	--
23	4-Br	4-N(CH ₃) ₂	145	78	189-192	--	--
24	4-Br	4- NO ₂	110	80	172-175	176-178	31
25	4-NO ₂	H	180	75	138-140	141-143	31
26	4-NO ₂	4-OH	85	42	158-161	--	--
27	4-NO ₂	2-OH	90	85	158-162	--	--
28	4-NO ₂	4-OCH ₃	300	no reaction	--	--	--
29	4-NO ₂	4-N(CH ₃) ₂	180	72	178-182	--	--
30	4-NO ₂	4- NO ₂	150	81	110-117	--	--

Table:4 physical characterisation of Schiff base

Characterization of the products:

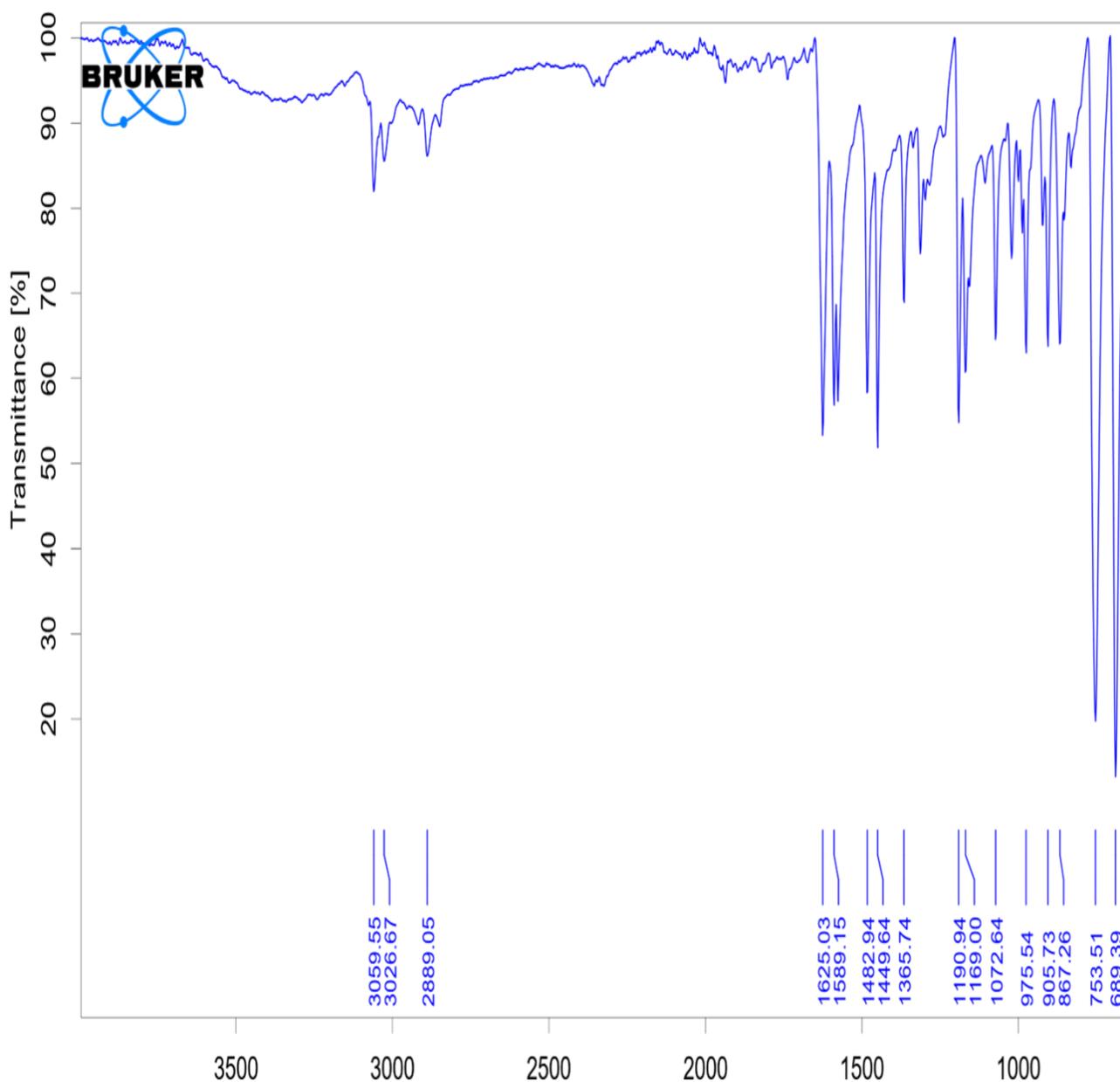
Both goods are stable at ambient temperature and do not require hygroscopic.

They are soluble but disintegrate at high temperatures.

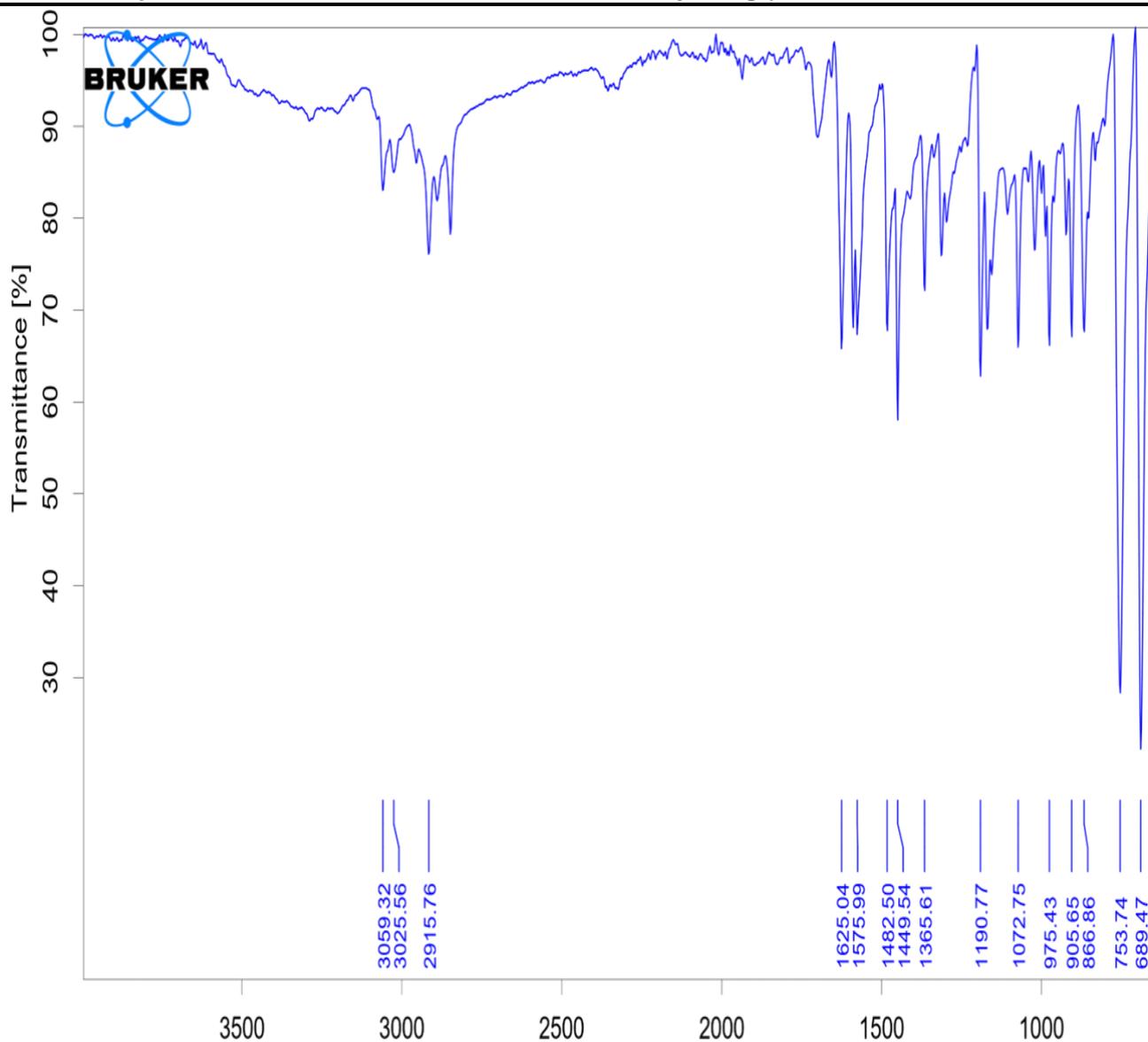
Solubility in ethanol and water. Consequently, characterising Products' physical characteristics were used, melting mass spectra, TLC, and datapoints. TLC was carried out using methanol and chloroform (1:9), one spot on TLC plate that displays the product's purity.

Sr. no	Product name and chemical formula	Product colour	Product smell	Physical state	solubility	Melting point	Rf value
1	N-benzylidene Aniline (C ₁₃ H ₁₁ N)	Pale yellow	Disagree- Able	Crystal- lain solid	Methanol ethanol	48	0.610

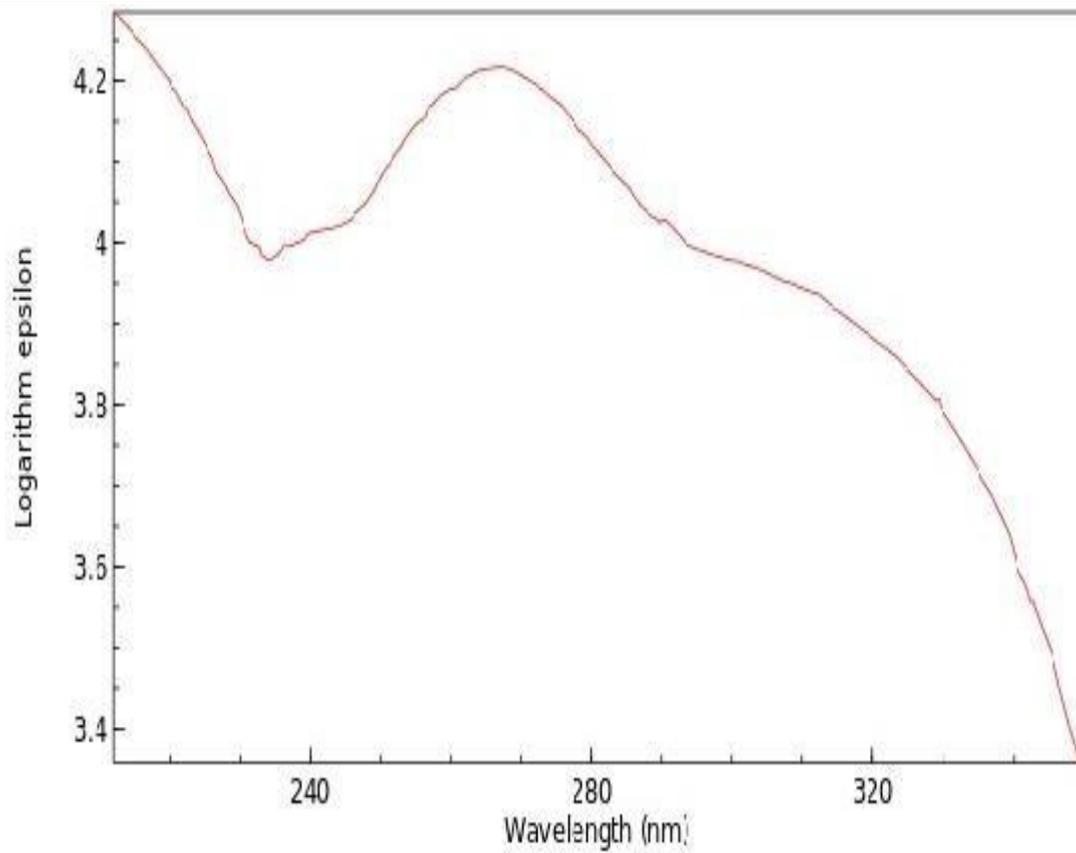
Table:5 TLC OF Product



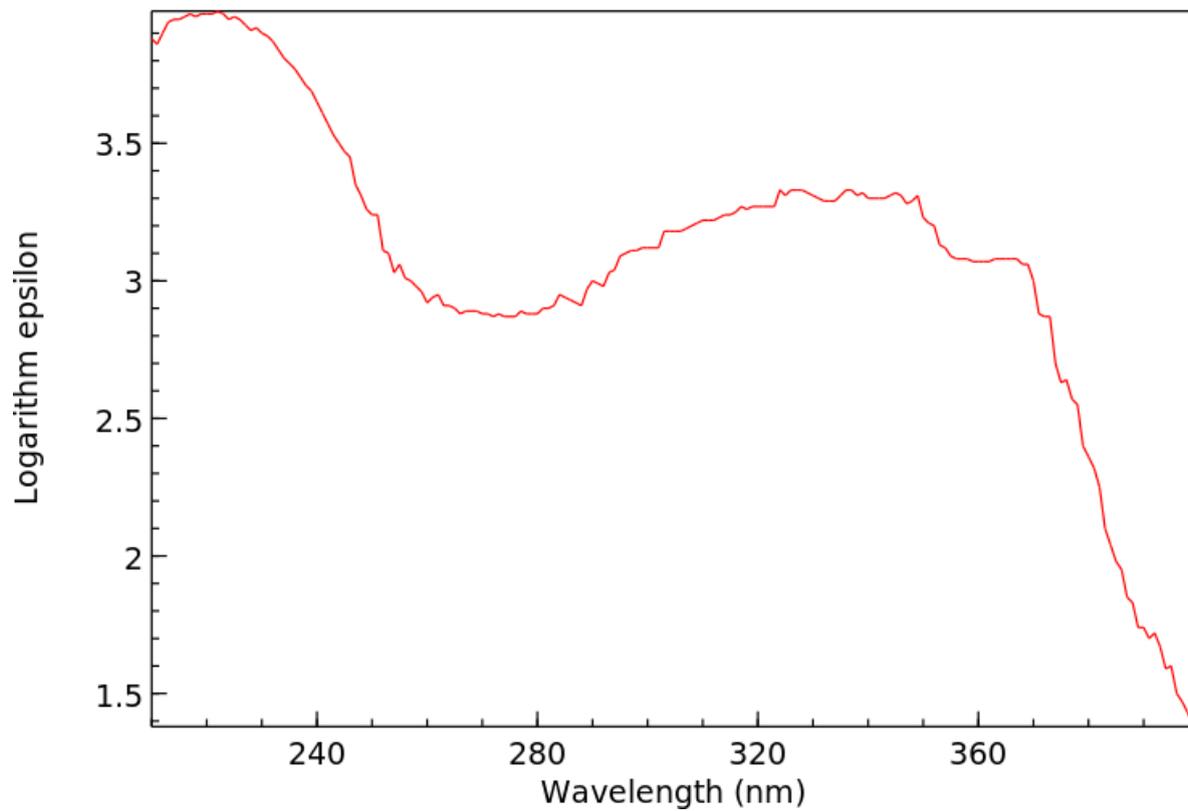
FT-IR Of benzylidene aniline (orange fruit juice as a catalyst)



FT-IR Of benzylidene aniline (Glacial acetic acid as a catalyst)



UV/VIS Spectroscopy of benzylidene aniline (orange juice catalyst)



UV/VIS Spectroscopy of benzylidene aniline (glacial acetic acid catalyst)

CONCLUSION

We are reporting a new eco-friendly route with good yield for the synthesis of Schiff bases by using orange Juice, Sweet lime juice and butter-milk the products can be purified by Recrystallization using appropriate solvents. This solvent-free approach is nonpolluting and does not employ any toxic materials, quantifying it as a green approach for the synthesis of Schiff bases. In addition to this, compared to traditional methods, this new method is cleaner, safer, cheaper and more eco-friendly, involving mild reaction conditions and simple workup. The reaction conditions such as reaction time, use of hazardous solvents can be reduced by maintaining good yield of product

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