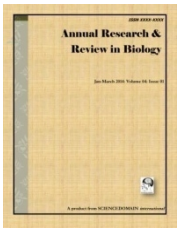

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Production Technology and Applications of Kojic Acid

Jignesh Chaudhary[1*], A. N. Pathak[1] and Sudarshan Lakhawat[1]

[1]Amity Institute of Biotechnology, Amity University Rajasthan, NH-8, Kant Kalwar, Jaipur, Rajasthan, India.

Authors' contributions

This work was carried out in collaboration between all authors. Author JC designed the study, managed the literature searches and wrote the first draft of the manuscript. Authors ANP and SL managed the analyses of the study and the applications part. All authors read and approved the final manuscript.

Review Article

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ABSTRACT

Kojic acid is produced industrially by *Aspergillus* species in aerobic fermentation. The production of kojic acid is increasing because of its commercial value in industry. Kojic acid has various applications in several fields. It is widely used in cosmetic industry, medicine, food industry, agriculture and chemical industry. Nowadays, kojic acid plays a crucial role in cosmetic, especially skin care products because it can enhance the ability to prevent exposure to UV-radiation. Kojic acid continues to attract attention because of its economic potential in medical field as an anti-inflammatory drug and painkiller. In food industry, kojic acid is used in post harvest process as an anti-speck and an anti-browning agent for agricultural product. Due to various usage of this organic molecule, the demand of kojic acid has been increasing rapidly. Thus the studies to improve the kojic acid production are still extensively conducted.

Keywords: Kojic acid; Aspergillus; de-pigmentation; food industry; anti-inflammatory.

**Corresponding author: Email: jigneshchaudhary23@rediffmail.com;*

ABBREVIATIONS

Symbol	Name
w/v	*Weight per volume*
g	*Gram*
ml	*Milliliter*
g/l	*Gram per liter*
V/V	*Volume per volume*
$Y_{p/s}$	*Product yield coefficient*
C:N	*Carbon to nitrogen ratio*
KA	*Kojic acid*
DOT	*Double oxygen tension*
NF-kB	*Nuclear factor kappa-light chain enhancer of B cells*
SSF	*Solid state fermentation*
SMF	*Sub-merged fermentation*
Y	*Yield*
Q	*Productivity*
S	*Substrate concentration*
P	*Product concentration*
rpm	*Revolution per minute*
Spp.	*Species*
ROS	*Reactive oxygen species*
BMOV	*Bis-(maltolato) oxovanadium*
MIC	*Minimum inhibitory concentration*
TLR 4	*Toll-like receptor 4*

1. INTRODUCTION

Kojic acid is an organic acid and it is a secondary metabolite secreted by several microorganisms of *Aspergillus* genus such as *Aspergillus oryzae, Aspergillus tamarri, Aspergillus parasiticus* and *Aspergillus flavus* [1]. There have been 58 different strains used for production of kojic acid are *Penicilium, Mucor, Aspergillus* etc [2]. Kojic acid can also be produced by using several plants such as *Kigella* African [3]. Kojic acid name was derived from “Koji”, a fungus or starter inoculums used in oriental food fermenters, many years ago in Japan. This crystalline substance was firstly isolated by Saito in 1907 [4], from the mycelia of *Aspergillus oryzae* grown on steamed rice. The chemical structure was determined as 5-hydroxy-2-hydroxymethyl-δ-pyrone by Yabuta in 1924 [5]. It is multifunctional and having weak acidic property. The natural origin of kojic acid confirms its non-hazardous bio-degradation makes it attractive and profitable skeleton for development of biologically active compounds by its derivation. Kojic acid crystallizes in form of colorless and prismatic needles [6]. Although kojic acid can be synthesized artificially from chemical conversion of various substrates, commercially it is produced by aerobic fermentation of *Aspergillus* species and it is one of the best practices is being conducted in industries.

Kojic acid has several economic uses in various fields. In medical field, kojic acid is used as an anti-bacterial and anti-fungal agent. In chemical industries it has been successfully used

to make azo-dyes and some other important and bio-degradable compounds. In the food industries, kojic acid is used as an anti-speck and anti-melanosis (blackening of product) agent for agricultural products. Since India has many agricultural product varieties, the use of kojic acid could be economically important in the post harvest process. In addition, kojic acid is also used as a chelating agent and activator in insecticide production. Recently, a new application of kojic acid is found in the cosmetic industry [1]. It is used as a skin whitening agent and ultraviolet filter in skin care products widely available in cosmetic market. Because of its effectiveness and good results in skin whitening, it has become more popular in Asia continent (especially China and Japan).

Although, kojic acid has been produced and applied industrially, attempts to improve kojic acid production are still been intensively studied. Kojic acid is in market since last 40-50 years and it was firstly manufactured by Pfizer company in U.S.A. Pfizer company has patented methods of production of kojic acid and its recovery by extraction as well [1]. Also derivatives of kojic acid were produced to use as bio-pesticides. However, there was no more market area due to its limited applications. Two main areas which are normally considered for the improvement of kojic acid production are the improvement of strain and development of the fermentation process. Screening of high kojic acid secretor from various strains and also improvement of strain through various mutation processes had been conducted in the last few decades [7]. Mutations in genes secret kojic acid by Ultraviolet or Gamma radiation cause overproduction of kojic acid which is more advantageous for kojic acid manufacturers [2]. Though, works on optimization of medium composition and environmental condition for kojic acid production by microorganisms have been studied extensively. Kojic acid can be produced in ample amount by using different carbon and nitrogen sources, also using agriculture based waste under aerobic fermentation strategies. To date, glucose has been described as a high kojic acid yielding raw material. However, use of some mutated strains of *Aspergillus flavus* has shown good yield of kojic acid by using potato starch, sago starch, corn starch etc.

Kwak MY & Rhee JS [8,9] found the use of immobilized viable cells as an approach to enhance kojic acid production. Information on the influence of aeration condition on kojic acid production is also scarce. Also an approach to enhance kojic acid production by different pH, temperature and dissolved oxygen strategies have not been studied extensively.

Researchers have stated that kojic acid production in very high amount by applying double oxygen tension (DOT) strategies, double phase pH and Temperature strategies. It has been investigated that very small changes in cultural pH and temperature cause very high effect on kojic acid production.

Kojic acid has many industrial applications and its demand is increasing as it is being applied to various areas regarding to healthcare, agriculture, food processing, cosmetic industries and many other chemical industries etc.

2. PROPERTIES OF KOJIC ACID

Kojic acid structure plays an important role in determination of some chemical and physical properties it possesses.

2.1 Structure of Kojic Acid

2.1.1 Physical properties

1) Structure of kojic acid is determined as 5-hydroxy-2-hydroxymethyl-δ-pyrone (Fig. 1) [5].
2) Molecular formula of Kojic acid is $C_6H_6O_4$ and molecular weight is 142.11g/mol [10].
3) Kojic acid crystallizes in form of colorless, prismatic needles that sublime under vacuum conditions without any changes.
4) Melting point of kojic acid ranges between 150-160ºC [11,12,13].

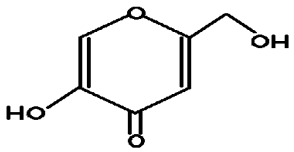

Fig. 1. Kojic acid Structure

2.1.2 Chemical properties

1) Kojic acid is soluble in polar substances like water, ethanol, ethyl acetate etc. On the contrary, kojic acid is very less soluble in chloroform, ether etc.
2) Kojic acid is classified as multifunctional reactive γ-Pyrone with weekly acidic properties.
3) Kojic acid molecule is reactive at every position on a ring.
4) At carbon 5 position hydroxyl (OH^-) acts as weak acid, which is capable to form salts with few metals such as Sodium, Zinc, Copper etc which make it more reactive [14].
5) Kojic acid and its derivatives with saccharin molecule are soluble in water.
6) Structure of kojic acid can be modified by glycosylation [15].
7) The side chains of carbon 5 behave as a primary alcohol whose reactivity can be enhanced by the adjacent oxygen atom in the nucleus [16].

2.2 Quantitative Determination

Kojic acid forms a complex with ferric ions to produce reddish purple color which has maximum absorption at 500 nm. This colorization is very useful as a principle method for the quantitative determination of kojic acid [17,14].

2.3 Kojic Acid Production

Kojic acid is produced industrially by *Aspergillus* species in aerobic fermentation. Industrial kojic acid production includes following main three stages (Fig. 2):

1) Inoculum development
2) Bio-production of kojic acid
3) Extraction & Purification of kojic acid

There are some factors that affect fermentation of kojic acid include use of high kojic acid yielding microorganisms, production media, type of fermentation operation, physiological conditions, aeration & agitation and minerals.

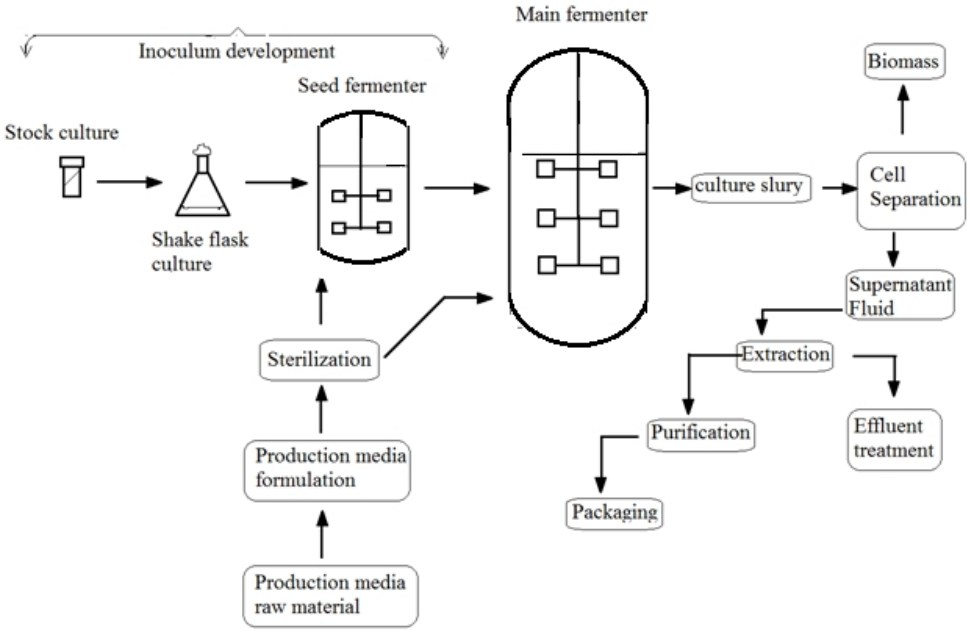

Fig. 2. Schematic representation of fermentative kojic acid production

2.4 Microorganisms

Moulds from the genus of *Aspergillus* are capable to produce large amount of kojic acid, especially, strains such as *A. oryzae, A. flavus, A. parasiticus* and *A. tamarii* [18]. Mycelium can make kojic acid when re-suspended in glucose buffer solution [19].

During fermentation process, the characteristics of moulds should be considered especially, mycelium formation rate. The excretion of enzymes that occur during mycelium formation before kojic acid production starts are not considered but the mycelium formation during and after production could influence the total yield of kojic acid [12].

Nevertheless, producing kojic acid especially, moulds from genera of *Aspergillus* and *Penicillium* species produce various types of aflatoxins, which are causative and chemically carcinogenic to human and animal (Table 1) [16,20,21,22].

Table 1. Toxins produced by *Aspergillus* spp

Fungus	Toxins
A. flavus	Aflatoxins, Aflatrem, Aspergilic acid, Cydoplazonic acid, β-nitropionic acid and Serigmatocyctin
A. oryzae	Aspergilic acid, Cycopiazonic acid and Maltoryzine β-nitropropionic acid, Ochtratoxins
A. parasiticus	Aflatoxins, Aspergillic acid and Sterigmatocyctin
A. toxicarius	Aflatoxins

2.5 *Aspergillus flavus*

Aspergillus flavus can be describe as a cosmopolitan, filamentous fungus that can be found in soils, plant products, particularly oil-rich seeds, and in living plant (Table 2) [23]. Generally, the colonies are yellow, light green-yellow and brown-yellow; some strains turn brown on aging. There are some characteristics of *A. flavus* have been described in Table 2 [24,25].

Table 2. Characteristics of *A. flavus*

Criteria	Characteristics
Pathogenicity	Usually a contaminant but also known to cause disease; commonly associated with aflatoxins.
Macroscopic morphology	Velvety, yellow to green or brown, Reverse goldish to red brown
Microscopic morphology of conidiophores	Variable length, rough, pitted, spiny
Microscopic morphology of phialides	Uni-seriate and bi-seriate, cover entire vesicle, point out in all directions

A. flavus has been reported as a high kojic acid yielding strain among all kojic acid producing microorganisms. Some UV and Gamma mutated strains have shown high kojic acid production in batch fermentation and glucose as a best carbon source. It was investigated that UV and Gamma mutated parent strain AFNS 9 of *A. flavus* gives high amount of kojic acid production 61.78g/l and 60.31g/l respectively by using glucose as a carbon source [2].

During fermentation process, glucose that accumulates in the culture converts into kojic acid through the action of cell-bound enzymes [11]. The cell-bound enzymes system consist glucose-6-phosphate dehydrogenase, hexokinase and gluconate dehydrogense, which are involved in the direct synthesis of kojic acid from glucose [26]. It is well known that glucose acts as a precursor in kojic acid synthesis. Kojic acid production ceases when all the glucose in the culture depletes [27].

According to Bajpai P et al. [28] glucose oxidase activity was too low to be account for the formation of kojic acid but the other enzymes had sufficient activities. Correlation of the pattern of enzymes activities under different experimental condition with kojic acid concentration provided evidence for the involvement of glucose dehydrogenase and gluconate dehydrogenase in kojic acid biosynthesis. Based on these data a possible pathway for biosynthesis of kojic acid is presented. Gluconic acid-δ-lactone and at least one of the three compounds 3-ketogluconic acid lactone, 3-ketoglucose and oxy-kojic acid, are believed to be intermediates in this pathway (Fig. 3) [28,25].

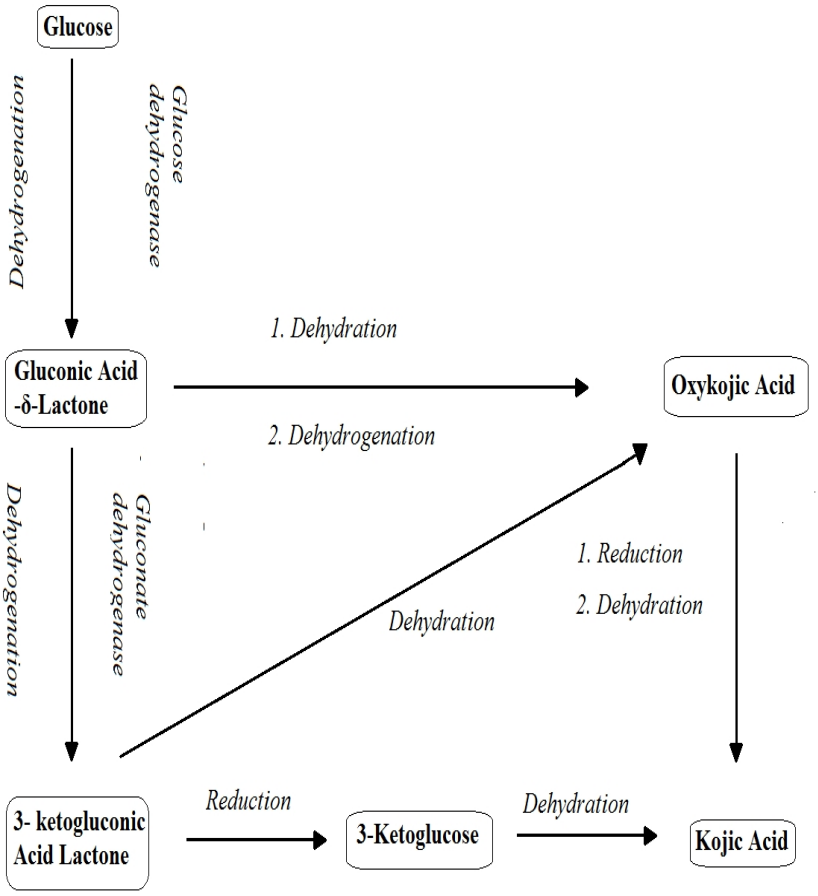

Fig. 3. Kojic acid biosynthesis pathway of *A. flavus*

2.6 *Aspergillus oryzae*

Kojic acid was produced at very first time by using *Aspergillus oryzae* strain. Wild strain of *Aspergillus oryzae* does not produce high amount of kojic acid by using glucose as a supreme carbon source. However, recent investigations on genetically modified or mutant strains of *Aspergillus oryzae* have demonstrated very high yield of kojic acid (Table 3). It is stated that *Aspergillus oryzae* wild strain B008 modified by using ion beam implantation and ethyl methane sulphonate treatment to obtain mutant strain M866 producing kojic acid with a high yield about 40.2g/l. In comparison of wild B008 strain of *Aspergillus oryzae* produce kojic acid with yield about 23.8g/l mutant strain produce 1.7 times more kojic acid [29].

It was investigated that *Lae A* gene regulates kojic acid production in *A. oryzae* strain. *Lae A.* deleted *A. oryzae* strains were not able to synthesize kojic acid [30].

A. Oryzae var effusus NRC14 has been reported a high kojic acid secretor strain among all *A. Oryzae* strains with yield of 42g/l [31].

2.7 *Aspergillus parasiticus*

Aspergillus parasiticus is one of the high kojic acid yielding fungi. In comparison of *Aspergillus flavus* and *Aspergillus oryzae* it is less productive. It was found that strains with deleted *msnA* produced more conidia and elevated kojic acid production as mechanism of oxidative stress relief [32].

2.8 *Aspergillus tamarii*

Generally, *Aspergillus tamarii* isn't suitable for industrial production of kojic acid because of its less productivity. It was found that *Aspergillus tamarii* NRC 18 showed low productivity of kojic acid (15.2g/l) in comparison of other *Aspergillus* species such as *A. flavus, A. oryzae, A. parasiticus* etc [31].

2.9 Trichoderma spp

Trichoderma reesei and *Trichoderma viride* secret very low kojic acid during fermentation, yield of kojic acid from *Trichoderma reesei* and *Trichoderma viride* were 3g/l and 5g/l respectively [33].

Table 3. Improvement of Kojic acid production by using Mutant strain

Microorganism	Carbon source(S)	P (g/l)	References
A.Oryzae M866	Glucose (75%) Xylose (25%)	40	Yan S et al. [29]
AFG7 of parent strain AFNS9	Glucose	60.31	El-Aziz ABA [2]
AFUV 8 of parent strain AFNS 9	Glucose	61.78	El-Aziz ABA [2]

**AFG: Aspergillus flavus Gamma mutated*
**AFUV: Aspergillus flavus Ultra-violate mutated*

2.10 Production Media

Every fermentation process requires specific medium but certain basic requirements components must be present within fermentation media. All microorganisms used in fermentation require water, various carbon sources, nitrogen, certain minerals, vitamins and oxygen if process is aerobic.

For kojic acid production wide range of carbon and nitrogen sources have been used are described in following table (Table 4):

Table 4. Composition of production media

Element	Source
Carbon	Glucose Xylose Starch Sucrose Lactose Arabinose Ribose

	Cane molasses Beet molasses Cereal grains Corn Stover/stalk
Nitrogen	Corn steep liquor Ammonium salts Urea Nitrates
Minerals	$ZnSO_4$ KH_2PO_4 $M_gSO_4.7H_2O$ $CuSO_4.5H_2O$
Oxygen	Natural sterile air (Through aeration)

Glucose is the best carbon source for Kojic acid production. Pentose and methyl pentose are rarely used carbon sources with very less kojic acid productivity [11].

2.11 Effect of C: N Ratio on Kojic Acid Production

One of the important factors in medium optimization for kojic acid production is C: N ratio. The optimal kojic acid production was reported at a C: N ratio between 75 and 100 [12]. This suggests that kojic acid production enhances in nitrogen-limited fermentation. In order to improve kojic acid production, the amount of nitrogen source added to the medium must be increased with the amount of carbon source at an optimal C: N ratio [34].

2.12 Nitrogen Sources

Addition of nitrogen source has shown drastic improvement in kojic acid production. Different nitrogen sources such as yeast extract, ammonium nitrate and peptone have been used in medium optimization for improvement of kojic acid production in laboratories and industries as well. Yeast extract of concentration 1 w/v% is very effective for higher kojic acid production than peptone (Fig. 4) [35]:

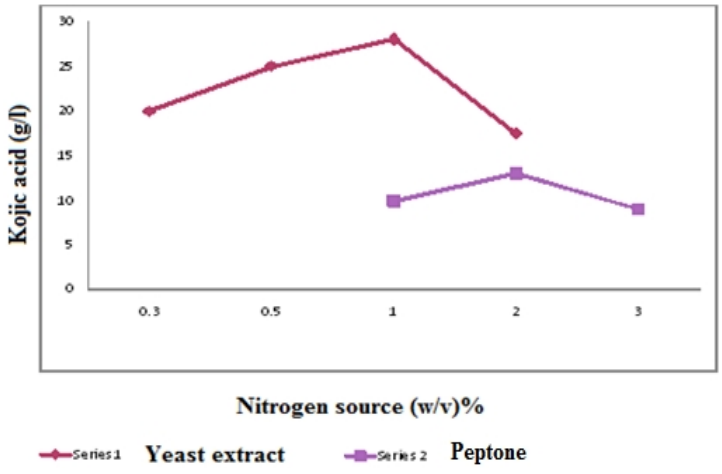

Fig. 4. Effect of Nitrogen source on kojic acid production

2.13 Effects of Substrate Concentration on Kojic Acid Production

Substrate concentrations also deeply affect kojic acid production. El-Aasar SA [35] found the effect of substrate concentration on kojic acid production by taking different sugar substrates such as glucose, sucrose and beet molasses in different amount in which glucose found supreme carbon source of concentration 6w/v is depicted in figure given below (Fig. 5).

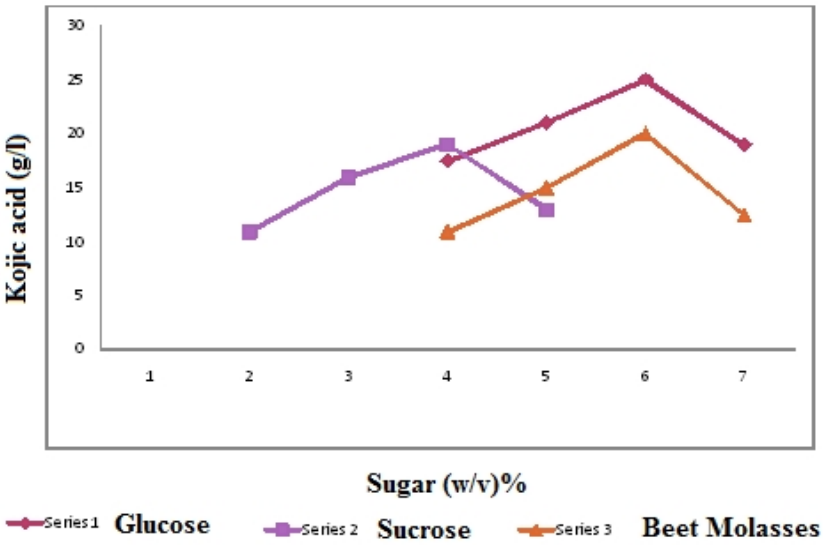

Fig. 5. Effect of substrate concentration on kojic acid production Fermentation techniques

There are mainly two types of fermentation techniques include solid state fermentation (SSF) and submerged fermentation (SMF) and different modes of operations such as batch fermentation, semi-batch fermentation and continuous fermentation have been employed industrially for the development of kojic acid fermentation.

Kojic acid is an extracellular product which is not growth associated. Batch fermentation is most suitable process for secondary metabolites production. Sometimes semi-batch fermentation used to enhance secondary metabolite production when there is a problem of substrate limitation.

2.13 Solid State Fermentation (SSF)

Solid state fermentation (SSF) is known as the technique where molds or yeasts are grown on a solid or liquid medium without any requirement of agitation or shaking of the fermentation vessel [36].

Solid state fermentation has not been extensively studied for the production of kojic acid. To date various solid wastes have been used to improve yield and productivity of kojic acid as well. Solid-state fermentation (SSF) by using *Aspergillus flavus* produces very less amount of kojic acid comparison of submerged fermentation (SMF).

It was found that *Aspergillus flavus* link-44 by using pineapple as carbon source produces 0.415g/l kojic acid which is lesser than fermentation in submerged culture [37]. Also they have found that in solid state fermentation at 70% moisture content can be resulted into high production of kojic acid.

On the contrary, it has been stated that solid state or surface fermentation produces higher kojic acid than submerged fermentation (Table 5) [31].

2.14 Submerged Fermentation

Mostly kojic acid is produced by using submerged fermentation (SMF) method. It is highly reliable method for kojic acid biosynthesis [12,36,38]. The growth of aerobic micro-organisms in a submerged fermentation (SMF) is controlled by the availability of substrates (sugars), energy and enzymes produced by microorganisms. Production cultures are always of a heterogeneous nature hence, the rates of biochemical reactions can't be limited by the rate of substrate or product transfer at a particular interface [1].

Various modes of fermentation such as batch, fed-batch and continuous for the improvement of kojic acid production are also possible to apply in submerged type of fermentation in order to achieve economical and efficient fermentation process [12,39].

2.15 Batch Fermentation

Kojic acid is an extracellular metabolic product in which the production is not associated with mould growth since the production phase occurs after growth reached a stationary phase.

El-Aziz A.B.A. (2013) [2] noted high production of kojic acid 50.27g/l and 48.95g/l in batch culture by *A. Flavus* using glucose and sucrose as carbon sources respectively (Table 6 and 7).

Table 5. Kojic acid production in solid state and submerged fermentations

Method	Microorganism	Carbon source(S)	Nitrogen source	Y *p/s* (g/g)	Q (g/L. h)	Reference
SSF	*A. flavus*	Pineapple peel (glucose, Fructose)	--	0.263	0.415	Nurashikin S et al. [37]
SSF	*A. oryzae*	Sucrose (20%)	Yeast Extract (2%)	0.31	--	Chaves FC et al. [22]
SSF	*A. flavus* NRC13 *A. oryzae var effesus* NRC14 *A. parasiticus* *A. tamarri* NRC18	Glucose (100g)	Ammonium Nitrate (1.125g)	0.41 0.42 0.17 0.22	0.602 0.626 0.307 0.379	Hazzaa MM et al. [31]
SMF	*A. flavus* NRC13 *A. oryzae var effesus* NRC14 *A. parasiticus* *A. tamarri* NRC18	Glucose (100g)	Ammonium Nitrate (1.125g)	0.17 0.25 0.05 0.15	0.288 0.447 0.08 0.262	Hazzaa MM et al. [31]

Table 6. Kojic acid production in batch culture by using different carbon sources

Microorganism	Carbon source(S)	P (g/l)	References
A. oryzae	Glucose	24	Kitada M et al. [13]
A. oryzae B008	Glucose (75%) Xylose (25%)	23.8	Yan S et al. [29]
A. flavus	glucose	28.9	Ariff AB et al. [39]
A. flavus	Corn starch	19.2	Rosfarizan et al. [7]
A. flavus	Sago starch	0.3	Rosfarizan et al. [27]
A. flavus	Glucose	39.9	Rosfarizan et al. [7]
A. flavus	Xylose	35.1	Ariff AB et al. [38]
A. flavus	Sucrose	14.8	Ariff AB et al. [38]
A. oryzae	Corn starch	40	Futamura T et al. [40]
A. flavus	Glucose	50.27	El-Aziz ABA [2]
A. flavus	Sucrose	48.95	El-Aziz ABA [2]

Table 7. Kojic acid production per gram of reducing sugar consumed

Microorganism	Carbon source (S)	Y *p/s* (g/g)	Q (g/L. h)	Reference
A. oryzae M866	Glucose (75%) Xylose (25%)	0.36	0.17	Yan S et al. [29]
Trichoderma viride	Sucrose	0.062	0.016	Saleh RM et al. [33]
Trichoderma reseei	Sucrose	0.0375	0.0096	Saleh RM et al. [33]
A. flavus	Glucose	1.49	0.234	El-Aziz ABA [2]

2.16 Semi-Batch Fermentation

Semi-batch or fed-batch culture can be determined as a batch culture, which is fed continuously or sequentially with medium without removal of culture fluid [41]. The purpose is to promote product formation instead of biomass. A proper feed rate, with the right component constitution is required during the process [42]. The majority of large-scale industrial fungal fermentations involved fed-batch culture in which biomass is grown initially in batch culture until a chosen component of the substrate is fully utilized [43].

The optimization of kojic acid production by *Aspergillus flavus* Link 44-1 using gelatinized sago starch as carbon source, using different fermentation modes (batch and fed-batch with different feeding mode) was conducted in 8 liter stirred tank fermenter [27]. During kojic acid fermentation, the dissolved oxygen tension (DOT) was controlled at high level (40%-50% saturation) during the active growth phase, which was required for the enhanced secretion of α-amylase used for saccharification of starch and also for the formation of mycelia with higher ability in synthesizing kojic acid (Table 8). The yield (0.164g kojic acid/g starch) and overall productivity (0.97g/L/d) of kojic acid in fed-batch culture was 2 and 3 times higher than for batch fermentation (0.045g kojic acid/g starch, 0.45g/L/d) [27].

Table 8. Kojic Acid Production in fed-batch Culture

Microorganism	Strategy	S	P (g/l)	References
A. flavus	DOT control strategy	Glucose	28.90	Ariff et al. [39]
A. flavus	pH control strategy	Sago starch	31.23	Rosfarizan et al. [27]
A. flavus	pH control strategy	Glucose	48.69	Rosfarizan et al. [27]

2.17 Fix Volume Fed-Batch Fermentation

Fix volume mode is conducted by feeding the limiting substrate without diluting the culture. The dilution decreases the biomass concentration and result in an increase in the specific growth rate. As the feeding continues, the growth will decline gradually, as biomass increase and approaches the maximum sustainable in the vessel once more, at that point the culture may be diluted again [44]. A constant volume can be achieved by feeding substrate as neat gas, liquid or solid [45].

Rosfarizan M et al. [27] employed different feed strategies in which fixed volume fed batch fermentation by adding very small volume of highly concentrated gelatinized sago starch (140g/l) in initial media with starch concentration (60g/l) intermittently in active fungal culture at 2 days interval. Using this strategy DOT could be controlled at high levels (40-50%) during active growth that was required for secretion of required enzymes for improved kojic acid production (31.23g/l).

2.18 Variable Volume Fed-Batch Fermentation

A variable volume fed-batch culture refers to culture where the volume changes with the fermentation time due to the substrate fed. This volume changes dependent on the requirements, limitations and objectives of the operator. Rosfarizan M et al. [27] employed different feed strategies in which variable volume fed batch fermentation by adding high volume of gelatinized sago starch (140g/l) in initial medium with starch concentration (60g/l) showed almost 4 times high kojic acid (16.43g/l) than batch operation of kojic acid production (4.51g/l).But in comparison of fixed volume fed-batch fermentation variable volume fed batch fermentation gave poor performance.

2.19 Continuous Fermentation

Generally, kojic acid is produced in stress-full conditions. It is extra-cellular product secreted by various fungi of *Aspergillus*. Rarely, continuous fermentation has been employed for kojic acid production as batch fermentation is much productive in comparison of continuous operation.

Kitada M et al. [13] reported kojic acid production by *A. oryzae* strain and using peptone (Nitrogen source) as the growth limiting nutrient with the use of two-stage continuous fermentation process. Slightly lower concentration of kojic acid at steady-state (6-7g/L) in the first fermentation vessel and second fermentation vessel was obtained as compared to the kojic acid production using similar factors in batch fermentation process (9g/L). The continuous fermentation is an ideal method for the production of microbial biomass and other growth associated processes (to get primary metabolites) such as ethanol production rather

than use for the production of secondary metabolites or extra cellular products such as kojic acid.

3. Effects of Physiological Conditions on Kojic Acid Production

Following physiological factors affect kojic acid production:

1) pH
2) Temperature
3) Moisture

3.1 Effects of Ph on Kojic Acid Production

High kojic acid production requires proper pH control strategies throughout production phase. Rosfarizan M et al. [27] investigated that highest kojic acid production can be achieved at pH between 4.5 to 6 during growth phase for the production of enzymes useful to convert glucose into kojic acid and pH between 2 to 3 during stationary phase for kojic acid production (Fig. 6).

Rosfarizan M et al. [34] found that more than 20% increment in kojic acid about 62g/l was reported by using double phase pH strategies rather than fermentation without any pH control strategy for kojic acid production about 49g/l was reported.

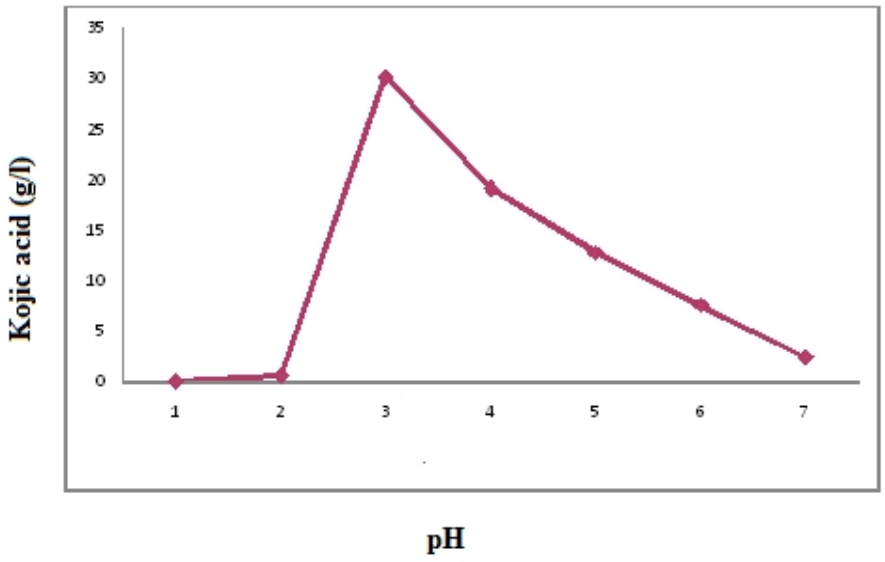

Fig. 6. Effects of pH on kojic acid production

3.2 Effects of Temperature on Kojic Acid Production

Temperature is one of the major kojic acid production parameters. High production of kojic acid can be achieved through maintaining optimum temperature during production period. El-Aasar SA [35] found that an optimum temperature for kojic acid production ranges from

24ºC to 28ºC. He has shown an effect of temperature on kojic acid production in figure given below (Fig. 7):

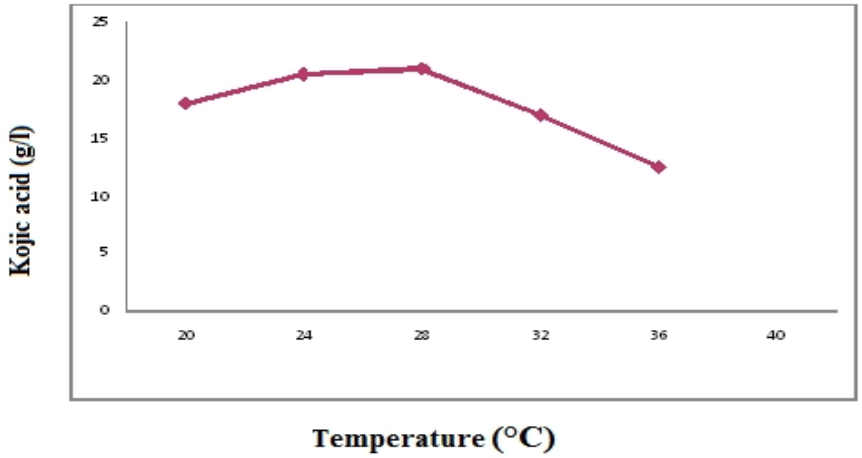

Fig. 7. Effect of temperature on Kojic acid production

3.3 Effects of Moisture on Kojic Acid Production

Generally, moisture is vital parameter for solid-state fermentation (SSF) in which it is required for proper growth of kojic acid secretory fungi. Nurashikin S et al. [37] found high kojic acid production using pineapple peel waste as substrate was reported at moisture level 70 %. Following figure shows effects of moisture level on kojic acid yield (Fig. 8):

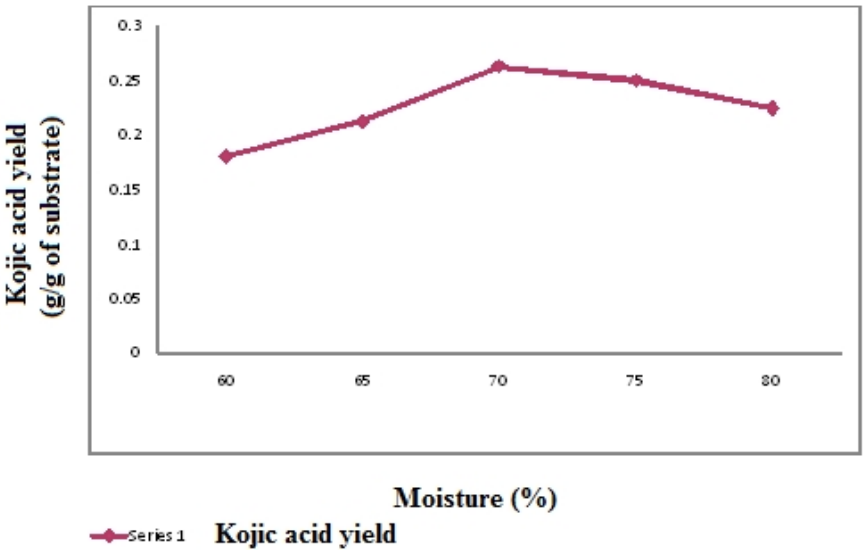

Fig. 8. Effect of moisture on Kojic acid production

3.4 Effects of Aeration and Agitation on Kojic Acid Production

Fermentations of kojic acid at pilot as well as at industrial scales are generally carried out in stirred tank reactors to ensure efficient oxygen transfer into the production culture. Some studies on the effects of aeration and agitation on kojic acid production in stirred tank fermenters at industrial scale had been conducted by Kitada M et al. [13]. The highest kojic acid production (32g/l) in a 300 l stirred tank fermenter was obtained at 1vvm (Air volume flow per unit of liquid volume per minute (volume per volume per minute)) and 240rpm (impeller tip speed = 8.04m/s) which gave the value of oxygen transfer rate (OTR) coefficient of 11.2×10^{-6} g/mol O_2/min. atm. ml [1].

Double oxygen tension (DOT) is also an important strategy for improvement of kojic acid production has been employed in several research projects. Ariff AB et al. [39] investigated that DOT level at about 80% saturation during growth phase for high growth of mycelia and DOT level at about 30% during kojic acid production phase resulted in high yield of kojic acid than same DOT level throughout fermentation.

3.5 Effects of Minerals on Kojic Acid Production

Czapek-dox medium containing Sucrose, Sodium nitrate, Di-potassium phosphate, Magnesium sulfate, Potassium chloride, ferrous sulfate are preferred for the growth of kojic acid producing and high kojic acid production [26,36]. However, the medium with carbon and nitrogen sources containing only di-potassium phosphate and Zinc chloride as minerals can also be used for a good growth of *A. parasiticus* and high kojic acid production [46]. Phosphate is an essential nutrient for the growth of most kojic acid producing fungi. It incorporates in bio-molecules such as nucleic acids, phospholipids and sugar phosphate; and plays an important role in energy metabolism. The proper concentration of phosphate in the culture broth gives a significant improvement on kojic acid production by *A. oryzae* [11]. Previous studies have shown that high phosphate concentration in the culture broth (0.6-13 mM) resulted in rapid kojic acid production. On the Contrary, at lower concentrations of phosphate (0.006-0.06mM), the rate of kojic acid was very much Lower and concentration of kojic acid obtained was about 2 times lesser than those which were obtained in fermentation using with high concentrations of phosphate.

4. EXTRACTION

Extraction of kojic acid from fermentation broth is carried out by using different polar compounds. Kojic acid is highly soluble in ethyl acetate than water makes an immiscible layer with first solvent which can be separate out easily by opening a stopcock of separatory funnel. Various extraction agents have been used by researchers to get high separation are listed below (Table 9):

Table 9. Extraction solvents used to separate kojic acid from fermentation broth

Extraction solvent	References
Ethyl acetate	Hazzaa MM et al. [31] Chaves FC et al. [22]
Chloroform	Parrish FW et al. [47] Hazzaa MM et al. [31]

4.1 Crystallization

Purification of kojic acid involves re-crystallization from water. If the sample is discolored a preliminary treatment with charcoal should be included. For small amounts, it is better to dissolve the crude kojic acid in the minimum volume of warm methanol and, after charcoal treatment if appropriate, add ether slowly to the point of incipient crystallization. Kojic acid crystallizes on standing [17].

Kojic acid crystallizes in yellow long needles a most purified form of kojic acid. Hazzaa MM et al. [31] found recovery of kojic acid from culture broth by keeping it under refrigeration about 5ºC, after one night of storage they observed long yellow needles with an average 39 gram of crystal/L were observed.

Saleh RM et al. [33] observed crystal growth at 0ºC after overnight storage of culture broth containing kojic acid. Same yellow long needles with highest production 3 to 5g/l were observed.

4.2 Kojic Acid Derivatives

Kojic acid represents an attractive multifunctional skeleton for development of biological active compounds. Inventors have prepared great varieties of kojic acid derivatives with selective properties. Thus, kojic acid derivatives are promising and advantageous to make human or veterinary medicines and also to prepare more biological active compounds with preferable properties. Since it is freely soluble in polar compounds such as water, ethanol, acetone, kojic acid was profitably employed for the preparation more than one hundred and fifty various kojic acid derivatives, many of them even represented new chemical compounds which were never synthesized before.

There are some potent developed derivatives of kojic acid listed below:

1) Chlorokojic acid (2-chloromethyl-5-hydroxy-4*H*-pyran-4-one)
2) Allomaltol (5-hydroxy-2-methyl-4*H*-pyran-4-one)
3) Iodokojic acid (2-iodomethyl-5-hydroxy-4*H*-pyran-4-one)
4) Fluorokojic acid (2-floromethyl-5-hydroxy-4*H*-pyran-4-one)
5) Comenic acid (5-Hydroxy-4-oxo-4H-pyran-2-carboxylic acid)
6) Pyromeconic acid (3-hydroxy-4*H*-pyran-4-one)
7) 2-substituted aryl (Indolyl) kojic acid
8) Vanillin-Kojic acid ligand
9) Bis (maltolato) oxovanadium(IV)
10) Bis ((5-hydroxy-4-oxo-4H-pyran-2-yl)methyl-2-hydroxy-benzoatato) oxovanadium

4.3 Chlorokojic Acid

Chlorokojic acid (Fig. 9) can be synthesized by simply a chlorination of the 2-hydroxymethyl moiety of kojic acid molecule using thionyl chloride ($SOCl_2$) at room temperature forms chlorokojic acid, with the ring hydroxyl being unaffected. Reaction of kojic acid molecule with thionyl chloride is given below (Fig. 10) [48,49].

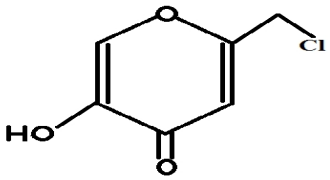

Fig. 9. Chlorokojic acid

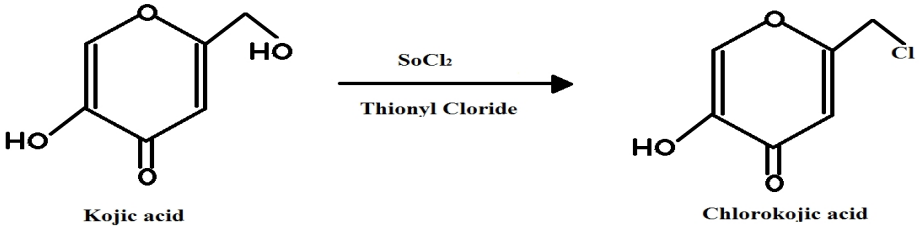

Fig. 10. Chlorokojic acid synthesis

4.3.1 Allomaltol

Reduction of chlorokojic acid with zinc dust in concentrated hydrochloric acid results in the production of allomaltol. It is two steps reaction after synthesis of kojic acid (Fig. 11) [48,49].

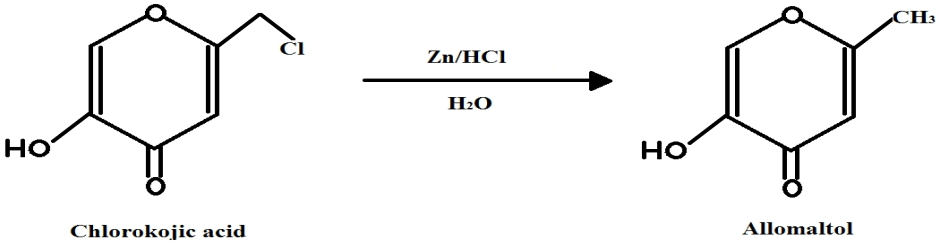

Fig. 11. Allomaltol synthesis

4.3.2 Iodokojic acid

Iodokojic acid is prepared from chlorokojic acid by treatment with potassium iodide in acetone (Fig. 12) [48].

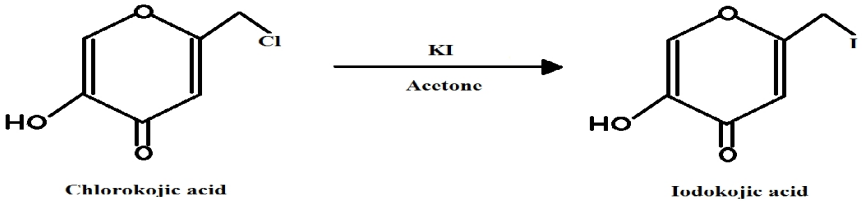

Fig. 12. Iodokojic acid synthesis

4.3.3 Fluorokojic acid

Iodokojic acid can be synthesized by treatment of any halokojic acid with various metal fluorides such as mercuric fluoride, silver fluoride etc (Fig. 13) [48].

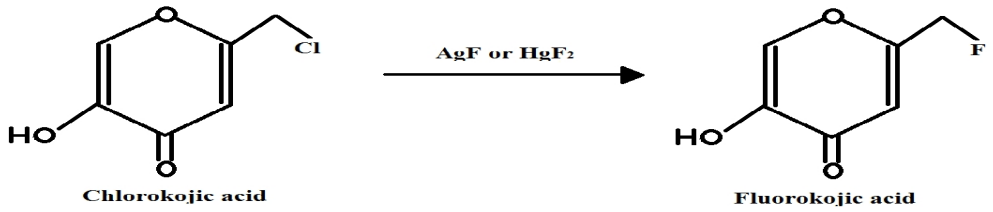

Fig. 13. Fluorokojic acid synthesis

4.3.4 Comenic acid

Comenic acid is prepared by simply an oxidation of kojic acid molecule. Oxidation reaction is given below (Fig. 14) [49].

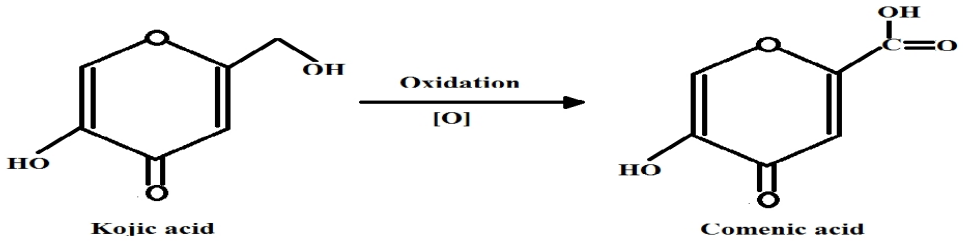

Fig. 14. Comenic acid synthesis

4.3.5 Pyromeconic acid

Synthesis of pyromeconic acid is one step further of comenic acid making from kojic acid molecule. It can be synthesized by decarboxylation of carboxyl group located at 5 position of comenic acid (Fig. 15) [49].

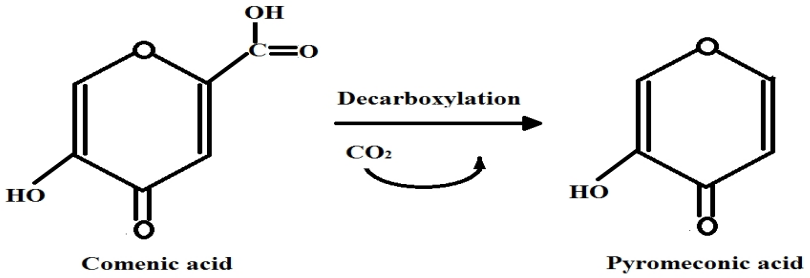

Fig. 15. Pyromeconic acid synthesis

4.4 2-Substituted Aryl (Indolyl) Kojic Acid

2-substituted aryl (Indolyl) kojic acid can be synthesized by coupling of iodole, aldehyde and kojic acid using catalytic amount of Indium chloride. Another approach is to use kaolin and Ag nanoparticles as reusable catalyst (Fig. 16) [50,51].

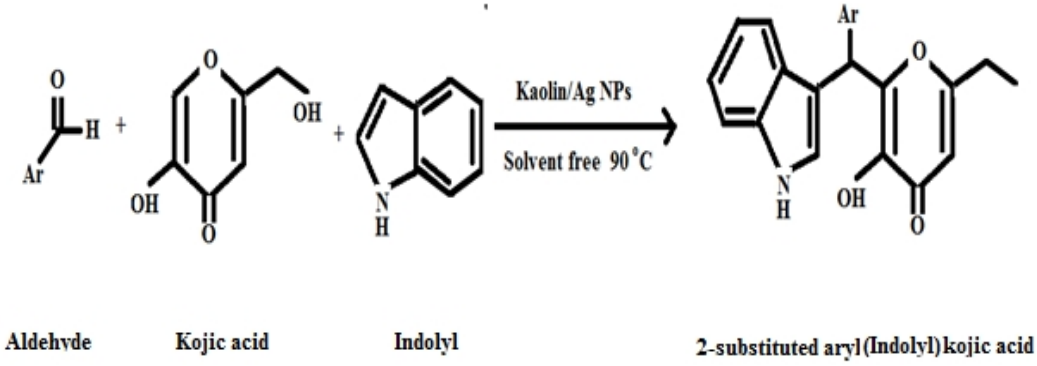

Fig. 16. 2-substituted aryl (Indolyl) kojic acid synthesis

4.5 Vanillin-Kojic Acid Ligand

Vanillin-Kojic acid ligand is designed by adding vanillin molecule in linker which makes strong ligand with two kojic acid molecules which is powerful chalator of iron (III) and aluminium (III) (Fig. 17) [52].

4.6 Bis (Maltolato) Oxovanadium (IV)

BMOV can be synthesized by complexation of maltol with vanadyl sulfate in refluxing aqueous solution, adjusting pH 8.5 using KOH (Fig. 18) [53].

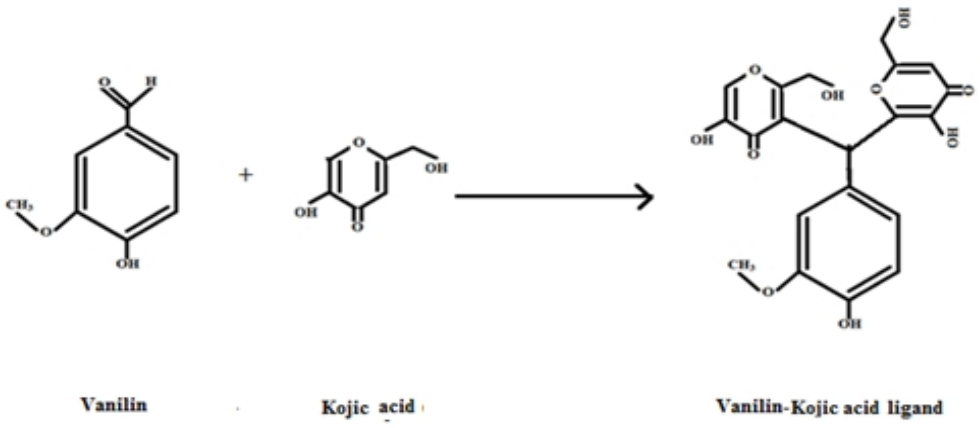

Fig. 17. Vanillin-Kojic acid ligand synthesis

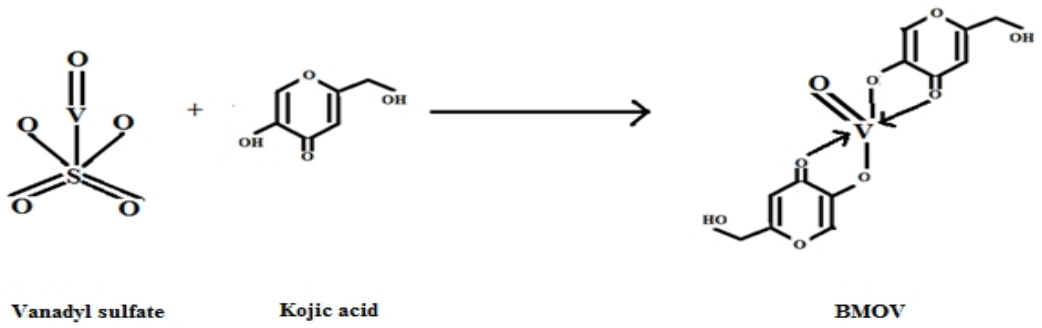

Fig. 18. Bis (maltolato) oxovanadium (IV)

4.7 Bis ((5-Hydroxy-4-Oxo-4H-Pyran-2-Yl) Methyl-2-Hydroxy-Benzoatato) Oxovanadium

Bis ((5-hydroxy-4-oxo-4H-pyran-2-yl) methyl-2-hydroxy-benzoatato) oxovanadium or BBOV can be prepared by adding vanadyl sulfate drop-wise in aqueous solution containing (5-hydroxy-4-oxo-4H-pyran-2-yl) methyl benzoate which is made by dissolving chlorokojic acid and sodium benzoic acid in DMF (Fig. 19) [54].

Among halokojic acids, chlorokojic acid has been reported to have fungicidal activity against certain organisms. Even though, it has certain shortcoming to use as fungicidal. It is irritant and hazardous to handle. Like chlorokojic acid, other halokojic acids also have some shortcoming to use as fungicides. Iodokojic acid releases iodine atom on warming or with-standing prolonged period. Another disadvantage of using halokojic acid in agriculture is to face difficulty in making formulation with compounds as halokojic acids are very light in weight and very fine powder form float on water when use to make suspensions.

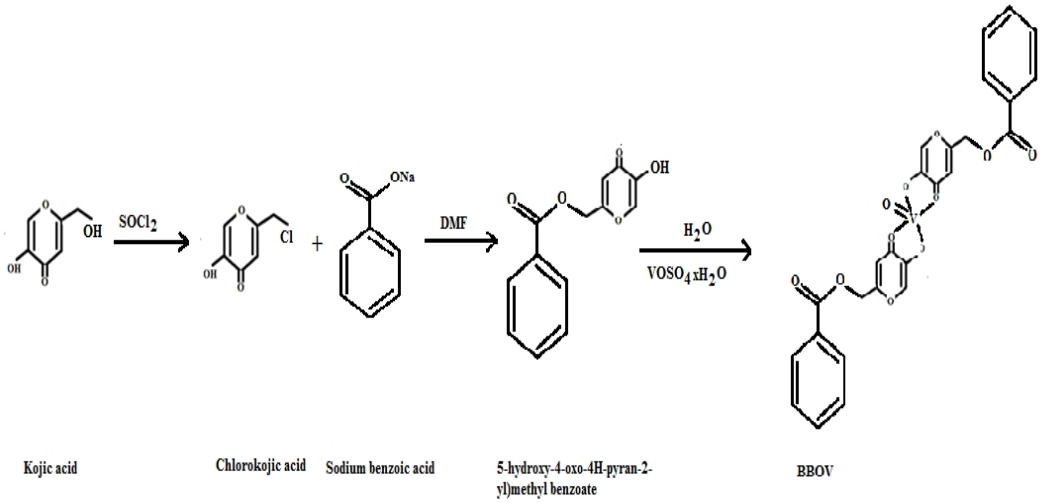

Fig. 19. BBOV synthesis

To overcome all this disadvantages inventors have used appropriate and valuable metal chelates. Metal chelates are very dense and not soluble within formulation compounds weather solid or liquid. They are also non hazardous for human being except come into contact by breathing. Additionally, they have been reported as toxic for larvae of numbers of insects.

Proper structures of metal chelates of these inventions are not known. It is thought that a divalent atom having co-ordination number 4 like copper in which salt formation tales place in between metal ion and two halokojic acid molecules at 5-Hydroxyl group. Further covalent bonding occurs at the carbonyl oxygen atom located at 4th position on halokojic acid and presents bi-cyclic structure in which metal containing both coordinate and covalent bonds. Tri and quadric-valent metals with coordination numbers 6 and 8 respectively make bonding with three and four halokojic acid molecules respectively. Divalent metal bonded with two halokojic acid is shown in figure given below (Fig. 20) [48].

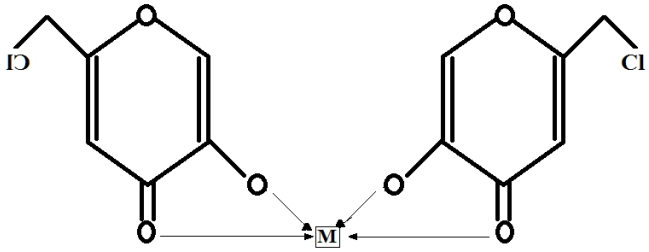

Fig. 20. Divalent metal chelation by two chlorokojic acid

It has been stated that all above metal chelates and those which are prepared from chlorokojic, iodo kojic acid, bromokojic acid possess metals which have atomic numbers in between 12 to 82. Metals like zinc, calcium, magnesium, cobalt, copper, aluminium, iron, mercury, barium which are appropriate, valuable fungicidal properties. All these metal shave coordination numbers in between 4 to 8 which are preferable to use them to make effective fungicides. Because of this property, these metals are applicable both in agriculture and industrially as well. Many other rare earth elements which atomic numbers are more than 82 aren't used to make fungicides because of rare availability.

The term fungicide is meant to include not only the property of destroying fungi but also the property of inhibiting the germination of the spores or the sporulation of the fungi, a property sometimes referred to as fungi-static or fungi-toxic.

5. Applications

Kojic acid has many applications (Table 10) and economic uses in various fields. Nowadays, the primary application of kojic acid in market is in the cosmetic industry in which it plays key role in skin care treatments [55]. Kojic acid has the ability to prevent ultra-violet radiation and inhibit tyrosinase activities which cause pigmentation [56].Intercalation of kojic acid in hydro-talcite-like compounds in order to stabilize kojic acid and to reduce its photolability which is very effective in melanin synthesis inhibition for skin treatment [57]. Kojic acid loaded nanotechnology based drug delivery systems can modulate drug permeation through the skin and improve the drug activity for the treatment of skin aging [58].

In the medical field, Kojic acid is widely used in medicinal and cosmetic formulations as a skin-lightening agent based on its de-pigmenting activity. Kojic acid is used as a pain killer and anti-inflammation drug [59]. Kojic acid and its peptide derivatives has also been reported as potential antibacterial agents [60]. Among them, 7-iodo kojic acid has the most potent activity against *staphylococcus aureus*. The factor that enhances the anti-microbial activity is attributable to the high hydrophobicity of the substituent at the end 7 position. Emami S et al. in [61] found novel mannich bases of 7-piperazinylquinolones with kojic acid and chlorokojic acid showed significant effect as antibacterial agents. Particularly chlorokojic acid derivative is most potent compound against *staphylococcus aureus* and *pseudomonas aeruginosa.* It showed activity about 4-8 times higher than standard drug norfloxacin. Immobilized kojic acid on magnetic nanoparticles grafted with chitosan and PEG has significant antimicrobial activity on both gram positive and negative bacteria [62]. In addition, kojic acid and its derivatives at concentration of about 100ppm are very effective to inhibit growth of many fungi [60]. Kojic acid can modulate macrophage activation through its cytoskeleton rearrangement, increase cell surface exposure and enhance the phagocytic process and ROS (Reactive Oxygen Species) production [63]. The study demonstrates a new role for Kojic acid as a macrophage activator. Kojic acid can highly protect Chinese hamster ovary (CHO) cells against ionizing radiation with low toxicity. In short, Kojic acid provides marked radio-protective effects both in vivo and in vitro [64].

Kojic acid derivative, O3-Acyl kojic acid as a potent and selective human neutrophil elastase inhibitor for the treatment of chronic and acute inflammatory lung diseases [65]. New bis-kojic derivatives induced faster clearance from main organs as compared with the mono-meric analog. So kojic acid could be applied as aluminium chelating agent in the treatment of aluminium related diseases [66]. Many kojic acid derivatives show significant effect on inhibition of D-Amino acid oxidase (DAAO) which is responsible for schizophrenia [67].Several kojic acid derivatives (RHS-0110, RHS-0111& RHS-0108) which are capable to suppress proliferation and induction of C6 glioma cells. Moreover, these kojic acid derivatives (organic acid chain addition) are found to modulate TLR4- mediated functional activation of macrophages, as assessed by No production under lowered or non-cytotoxic concentrations of kojic acid compounds. So, these results suggest that kojic acid derivatives, including RHS-0110, RHS -0111 and RHS-0108 could be useful as novel anti-cancer drugs with anti-proliferative and anti-TLR4-mediated micro-environmental formation features [68].

Kojic acid can be used as antioxidant iron chelator for topical treatment of wound healing [69]. Conjugation of kojic acid with vanadium improves its efficacy and safety to use as anti-diabetic agent. Effects of kojic acid derivative BSOV on streptozotocin induced diabetic rats with bis-maltolato oxovanadium (BMOV) effectively lowered blood glucose level that shows it can be use as an effective and safe anti-diabetic agent [70].

Dung TTM et al. in [71] found that MHNC treatment suppress a series of upstream signaling cascades consisting of IκBα, AKT, PDK1, Src and Syk for NF-κB activation. MHNC prevented inflammatory symptoms of the stomach in mice treated with HCl/EtOH by reducing phospho-IκBα levels.

In food industry, kojic acid is used as an agent to prevent undesirable melanosis (blackening) of agricultural products such as vegetables, fruits and crustaceans during storage. Kojic acid has the ability to inhibit the action of polyphenol oxidase (PPO) enzyme when these products are exposed to oxygen [72]. Apart from that, it is also used as an 'anti-speck' agent in raw noodles during production processes. This is to avoid the color changes and black spot formation on noodles by inhibiting the tyrosinase enzyme [73,10]. Anti-

bacterial activity of kojic acid grafted chitosan oligosaccharide derivative that supports for developing new antimicrobial agents and explore the scope of application of kojic acid in food industries [74]. Metal complex of Kojic acid–phenylalanine inhibits mushroom tyrosinase activity as much as Acid–phenyl alanine and reduce melanin contents in melanocyte efficiently [75].

In the chemical industry, kojic acid can be used as an analytical tool for ion determination since the reaction of kojic acid with the trace of ferric ion can form deep red complex [76]. Kojic acid also has been used as a substrate for chemicals synthesis of comenic acid and 2-methyl-4-pyrone [77]. Comenic acid is an important intermediate for the synthesis of maltol and its derivative, while 2-methyl-4-pyrone is a compound which is normally associated with natural pigments. Szklarzewicz J et al. [78] found novel chemical complexes of Mo (IV) in reaction with compounds maltol, ethyl maltol and kojic acid.

Kojic acid is widely used in agriculture as a chelating agent and insecticide activator for insecticide production. Newly designed two ligands composed of vaniline and O-vaniline molecules, each molecule with two kojic acid molecules joined with methylene group which have been proved as powerful chelators of iron and aluminium [52].

The addition of 5% kojic acid increases the toxicity of nicotine insecticide from 5 to 35% [49,76]. Natural compounds that pose no significant side effects in medicinal or environment field are potential sources of antifungal agents in agriculture either in normal form or as a structural backbone for more effective and efficient organic derivatives. Kojic acid greatly lowers minimum inhibitory (MIC) or maximum fungicidal (MFC) concentrations of commercial antifungal agents used in medicinal and agricultural field Amphotericin B (AMB) and strobilurin respectively against pathogenic yeasts and fungi. *A. fumigatus* cause human invasive aspergillosis, with H_2O_2 or AMB indicate a chemo-sensitizing activity of kojic acid is most effective in rupture of fungal antioxidant system. So kojic acid can be applied as chemo-sensitizer to improve the conventional fungal drugs or fungicides effectiveness and efficiency as well [98]. Kojic acid is potent chemo-sensitizing agent of complex III inhibitors disrupting the mitochondrial respiratory chain in fungi. Addition of kojic acid greatly lowers the minimum inhibitory concentrations of complex III inhibitors tested against certain filamentous fungi [99].

5.1 Future Work

To improve kojic acid production, future work should be carried out for improvement of productivity. Some suggestions are listed below:

1) Temperature & pH control strategy during complex carbohydrates hydrolysis and enhancement of kojic acid for higher productivity.
2) Aeration & agitation requirements of kojic acid production by using poly-carbohydrates.
3) Kojic acid production in fed-batch culture using variable mode volume.
4) Kojic acid production using different organic wastes such as whey (by product of dairy industry), Agro-processing industries etc.
5) Optimizing scale up criteria by KLa studies.

Table 10. Applications of kojic acid in different fields

Field	Functions	References
Cosmetic	Whitening agent (SPF 15), Melasma treatment, Hyper pigmentation cure, Pigmentation inhibitor, De-pigmentation, Skin aging treatment	Kobayashi Y et al. [79], Lim JT [80], Jimbow K, Minamitsuji Y [56]; Kim DS et al. [81], Ambrogi V et al. [57], Cho JC et al. [82], Goncalez ML et al. [58]
Medicine	Anti-fungal & anti-bacterial agent, Painkiller & anti-inflammation drug, Free radical preventer, Cytotoxic agent, Anti-inflammation pills, Type-3 copper protein, Tyrosinase inhibitor, Ultra-violet B inhibitor, Ultra-violet A inhibitor, Radio-protective agents, Macrophage activation, De-pigmentation medicines, Wound healing, Modulatory effect on cancer cell proliferation, Anti-diabetic agent, Schizophrenia, Aluminium chelator, Lung disease treatment,	Beelik A [16]; Kasser JH et al. [83]; Emami S et al. [61]; Hussein-Al-Ali SH et al. [62], Kayahara H et al. [60]; Dung TTM et al. [71], Niwa Y, Akamatsu H [84], Novotny L et al. [85], Ozturk G et al. [59], Tepper A et al. [86], Ahn KS et al. [87]; Reelfs O et al. [88]; Emami S et al. [89]; Hosseinimehr SJ et al. [90]; Wang k et al. [64], Rodrigues APD et al. [63], Lajis AFB et al. [91], Mohammadpour M et al. [69], Yoo DS et al. 68],Wei Y et al. [70], Raje M et al. [67], Toso L et al. [92], Lucas SD et al. [65],
Food industry	Antibacterial agent, Anti-speck agent, Anti-melanosis agent, Tyrosinase inhibitor, Anti-browning agent for fruits Xanthan gum	Liu X et al. [74] ; Morton HE et al. [93], Uchino K et al. [10], Kobayashi Y [55]; Kwak SY et al. [75], Kaatz H et al. [73], Son S et al. [72], Weiss RM, Ollis DF [94]
Chemical industry	Reagent for ion determination Substrate for comenic acid synthesis, Substrate for 4 (1H)-pyridone derivatives synthesis, Substrate for 2-methyl-4-pyrone synthesis, Novel chemical complexes of Mo(Iv)	Butcha K (1983) [76]; Tatsumi C et al. [77], Ozturk G et al. [59], Hasizume K et al. [95], Szklarzewicz J et al. [78]
Agriculture	Insecticidal mycotoxin, Chelating agent, Insecticides activator, Pesticides, Fungicides	Beard RL, Walton GS [96], Beelik A, Purves CB [49], Nurchi VM et al. [52], Butcha K [76], Dowd PF [97], Reddy BVS et al. [49]; Saleh RM [33]; Kim JH et al. [98]; Kim JH et al. [99]; Aytemir MD et al. [100]

6. CONCLUSION

Kojic acid is mostly secreted by more than 58 fungal strains of *Aspergillus* genus. *A. flavus* produces high amount of kojic acid by using glucose and yeast extract as carbon and nitrogen sources respectively. It can be produced by using variety of sugars such as sucrose, lactose, galactose, arabinose, ribose, starch and also using different organic wastes via microbial fermentation. Kojic acid is safe for human kind and because of its antioxidant and tyrosine inhibition property it is widely applicable in food sector as well as in medical research practices. Apart from medical uses it has shown effective results as antibacterial, antifungal and pesticide agent. More than 150 derivatives of kojic acid have been identified as bio-pesticide, bio-fungicide in agriculture. Numbers of chemicals can be made by reactions with other chemical or biochemical molecules. Because of its biodegradable property it has been more popular in agriculture sector. Very low dose of kojic acid about 1% to 3% is more effective in de-pigmentation of skin. Furthermore, studies in these fields will help to improve human ability to combat with microbial infections and disease such as cancer.

7. SUMMARY

Kojic acid is an organic acid and it is a secondary metabolite secreted by several microorganisms of *Aspergillus* genus such as *A. oryzae, A. tamarri, A. parasiticus* and *A. flavus*. There have been more than 58 different strains used for production of kojic acid are *Penicilium, Aspergillus* etc. Kojic acid can also be produced by using several plants such as *Kigella* African. It is produced by both solid state and submerged fermentation by using batch, semi-batch and continuous operations. Glucose and yeast extract are high yielding carbon and nitrogen sources respectively by various fungal strains. Kojic acid molecules are reactive at every position on a ring and have numbers of application in chemical industry to make metal chelates, pyridines, pyridines, azodyes etc. Kojic acid has several economic uses in various fields. In medical field, kojic acid is used as anti-bacterial and anti-fungal agents. In chemical industries it has been successfully used to make azodyes and some other important and biodegradable compounds. In the food industries, kojic acid is used as an anti-speck and anti-melanosis (blackening of product) agents for agricultural products. Since India has many agricultural product varieties, the use of kojic acid will be economically important in the post harvest process.

Kojic acid represents an attractive multi-functional skeleton for development of biological active compounds. Inventors have prepared great varieties of kojic acid derivatives with selective properties. Thus kojic acid derivatives are promising and advantageous to make human or veterinary medicines and also to prepare more biological active compounds with preferable properties. Since it is freely soluble in polar compounds such as water, ethanol, acetone, kojic acid was profitably employed for the preparation more than one hundred and fifty various kojic acid derivatives, many of them even represented new chemical compounds which were never synthesized before.

Hence, Kojic acid has many industrial applications and its demand is increasing as it is being applied to many areas regarding to healthcare, agriculture, food processing, cosmetic industries etc. Though various fermentation approaches can be applied for kojic acid efficient production.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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