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Review Article

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Kojic acid-derived tyrosinase inhibitors: synthesis and bioactivity

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Abstract

Tyrosinase is a key enzyme for melanin biosynthesis, catalyzing the oxidation of L-tyrosine to L-dopaquinone. The tyrosinase inhibition is an effective approach to control hyperpigmentation in human skin and enzymatic browning in fruits and vegetables. Kojic acid is a naturally-occurring tyrosinase inhibitor which has been clinically used to treat the hyperpigmentation of skin. However, kojic acid as a hydrophilic small-molecule has insufficient inhibitory activity and stability, with considerable toxicity. To overcome these drawbacks, synthetic derivatives of kojic acid were developed, which exhibited enhanced tyrosinase inhibitory activity and more favorable stability relative to kojic acid. In this context, the synthesis and biological activity of kojic acid derivatives as tyrosinase inhibitors have been highlighted.

Keywords: Kojic acid, 4*H*-pyran-4-one derivatives, enzyme inhibitors, structural modifications

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Introduction

Melanin is a dark pigment produced by about 10% of skin cells in the innermost layer of the epidermis (1). This compound is a heteropolymer of indole derivatives and is produced inside melanosomes through a series of oxidative reactions involving the amino acid tyrosine in the presence of the enzyme tyrosinase (Fig. 1). The type and amount of produced melanin in the melanosomes generates the actual color of the skin (2). Melanogenesis is a physiological process, which plays an important role in the prevention of sun-induced skin injury. Although the melanin production in human skin is a major defense mechanism against ultraviolet (UV) light, the accumulation of an excess of epidermal pigments can causes various hyperpigmentation disorders, such as melasma, age spots, and sites of actinic damage (3).

Tyrosinase (EC 1.14.18.1), also known as polyphenoloxidase (PPO), is a copper-containing bifunctional enzyme with a molecular weight of approximately 60–70 kDa in mammals and is found exclusively in melanocytes (1,4). It catalyzes two distinct reactions of melanin synthesis (Fig. 1); the hydroxylation of

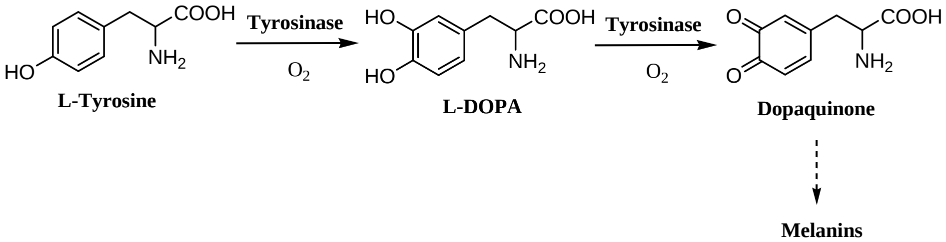

Figure 1 The role of tyrosinase enzyme in the melanogenesis process

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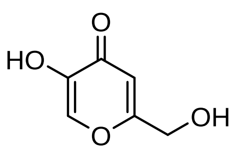
L-tyrosine to form 3,4-dihydroxyphenylalanine (L-DOPA) by monophenolase action and the oxidation of L-DOPA to the corresponding *o*-dopaquinone by diphenolase activity (5). Dopaquinone is highly reactive and can polymerize spontaneously to form melanin in a series of reaction pathways (6). The tyrosinase can be considered as a rate-limiting enzyme in the melanin biosynthesis (7). Accordingly, tyrosinase inhibitors significantly reduce pigmentation in melanosomes and avoid excessive melanogenesis. Some tyrosinase inhibitors have useful application in cosmetics and pharmaceutical products for the prevention of the overproduction of melanin in the epidermis (8). On the other hand, melanogenesis affects the color quality and flavor of foods. The enzymatic action of tyrosinase causes the browning in fruits and vegetables. Thus, tyrosinase enzyme plays an important role in loss of nutritional and market values of foods. In the food industry, tyrosinase inhibitors, especially from natural sources have great potential in controlling the quality and economics of fruits and vegetables (9). Many efforts have been spent in the search for effective and safe tyrosinase inhibitors, and a large number of naturally occurring and synthetic tyrosinase inhibitors were extensively reported (10-12). However, only few of them are practically applicable due to their weak intrinsic activities or safety concerns. Therefore, it is still necessary to search and develop novel tyrosinase inhibitors with potent activity and lower side effect (4).

Kojic acid

Kojicacid (5-hydroxy-2-(hydroxymethyl)-4*H*-pyran-4-one, **1**) (Fig. 2) is one of the metabolites



(1)

Figure 2 The structure of kojic acid

produced by various fungal or bacterial strains such as aspergillus and penicillium and has been used in many countries as a skin-whitening agent because of its tyrosinase inhibitory activity on melanin synthesis. The biological activities of kojic acid are attributed to its γ-pyranone structure that contains an enolic hydroxyl group. If the enolic hydroxyl group is protected, its tyrosinase inhibitory activity is completely lost. It acts by chelating copper at the active site of the tyrosinase enzyme (13). Melanocytes that are treated with kojic acid become nondendritic and have decreased melanin content (14). Kojic acid was reported to have a high-sensitizing potential and to potentially cause irritant contact dermatitis. However, it is useful in patients who cannot tolerate hydroquinone and it may be combined with a topical corticosteroid to reduce irritation (15). Additionally, it also acts as an antioxidant and scavenges reactive oxygen species that are released excessively from cells or generated in tissue or blood (16). The reaction of kojic acid with metal salts of aluminium, chromium, cobalt, copper, gold, indium, iron, nickel, manganese, palladium, vanadium, and zinc results in the formation of stable metal kojate complexes (17-20). Due to its iron chelating activity, kojic acid and its derivatives play an important role in the management of iron-overload diseases such as *β*-thalassemia or anemia (21-25). Moreover, various biological effects including antibacterial (26, 27), antifungal (28, 29), antiviral (30), anti-inflammatory (31), antineoplastic (32-34), pesticide (35), radioprotective (20), antidiabetic (36), and anticovulsant (37, 38) activities have been reported for kojic acid and its derivatives.

Kojic acid derivatives as tyrosinase inhibitors

Kojic acid is a naturally-occurring tyrosinase inhibitor, which has been clinically used to treat the hyperpigmentation of skin. However, kojic acid has insufficient inhibitory activity and stability, with considerable toxicity. To overcome these drawbacks, synthetic derivatives of kojic acid were developed. We discuss here the major modifications which were made on the kojic acid structure.

Conversion of γ-pyranone to 4-pyridinone: O-1 modification

The replacement of oxygen in the γ-pyranone ring with nitrogen resulted in 4-pyridinone analogs of kojic acid (Fig. 3).

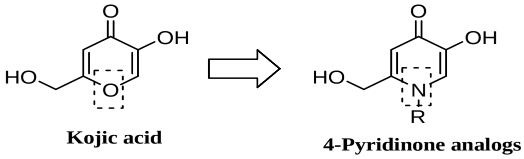


Figure 3 O-1 modification in kojic acid

Accordingly, a series of hydroxypyridinone–L-phenylalanine conjugates **5** (Fig. 4), starting from kojic acid was synthesized by Li et. al. and evaluated against mushroom tyrosinase (39). It was found that compound containing 1-octyl moiety (R= n-C_8H_{17}) had potent inhibitory effect against mushroom tyrosinase. MTT assay indicated that this compound was non-toxic to tested cell lines. For the synthesis of compounds, firstly kojic acid (**1**) was *O*-benzylated by benzyl chloride, and then reacted with an appropriate alkylamine to give compound **3**. The alcoholic compound **3** was conjugated with *N*-protected L-phenylalanine by using EDC and DMAP. Finally, *N*-deprotection was carried out by hydrogenation at 30 psi H_2 for 5−6 h at room temperature to give target compounds **5** (39).

Saghaie et al. synthesized a series of 3-hydroxy-4-pyridinones **9** starting from kojic acid in high yield (Fig. 5), and evaluated them for their inhibitory activity toward tyrosinase enzyme using dopachrome method (40). As illustrated in Fig. 5, the amine insertion in the *O*-benzyl kojic acid (**2**) resulted in compound **6**, which subsequently oxidized to aldehyde **7**. Condensation of aldehyde **7** with aniline derivatives gave Schiff base **8**. Reduction of C=N bond and *O*-debenzylation in compound **8** by using Pd/C hydrogenation afforded final compounds **9**. Their biological results show that all synthesized compounds have inhibitory effect on tyrosinase activity. Among compounds studied those containing two free hydroxyl group were more potent than their analogues with one hydroxyl group. Also, substitution of a methyl group on position N_1 of the hydroxypyridinone ring seems to confer more inhibitory potency (40).

Esterification of 2-(hydroxymethyl) group of kojic acid

The primary alcoholic group of kojic acid can be esterified with different acids. However, the convenient method for preparation of kojic esters is via chloro-kojic acid (**10**) and subsequent nucleophilic substitution with a suitable carboxylate salt (Fig. 6).

Nitric oxide (NO) is an important inflammatory mediator, synthesized by inducible nitric oxide synthase (iNOS).

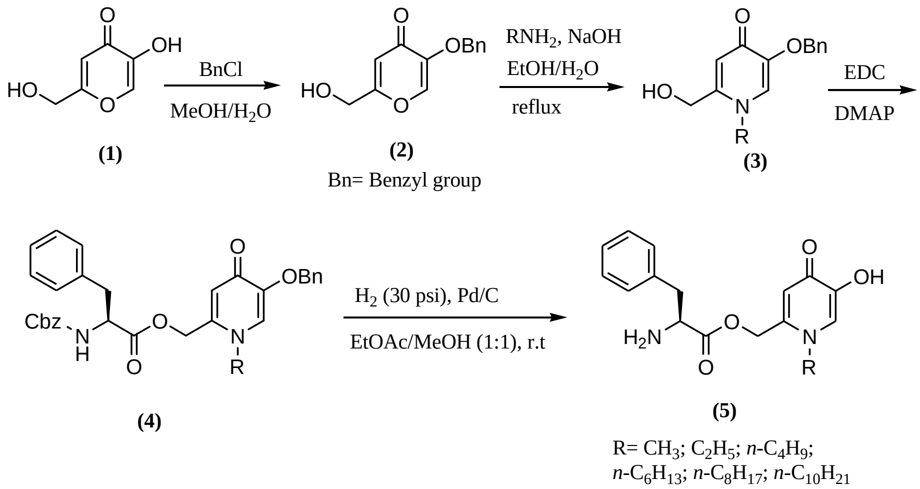


Figure 4 Synthesis of hydroxypyridinone–L-phenylalanine conjugates **5**

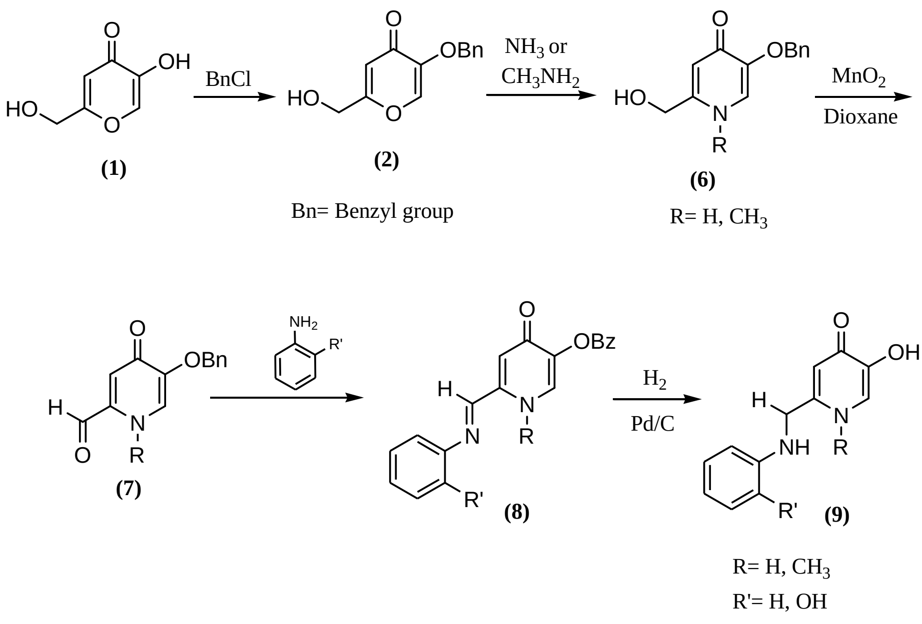


Figure 5 Synthesis of 3-hydroxy-4-pyridinones **9**

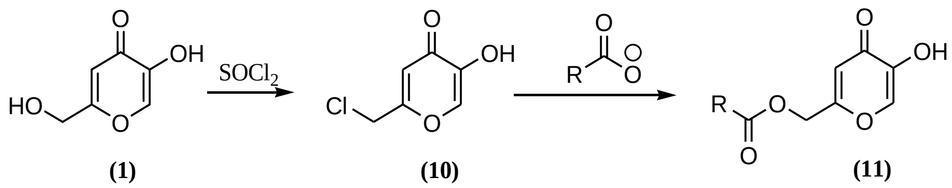


Figure 6 The convenient method for preparation of kojic esters **11**

Overproduction of NO can lead to inflammatory diseases (41). Thus, much effort has been focused on finding potent inhibitors of NO production. However, there have been few studies on the use of kojic acid and its derivatives as NO inhibitors. Recently, a series of kojic acid derivatives **12** containing ester linkage, and kojic-benzoates **16** containing adamantane moiety were synthesized and evaluated their inhibitory activities against tyrosinase and NO production. As depicted in Fig. 6, the reaction of kojic acid with thionyl chloride produces compound **10**, which is conveniently *O*-methylated to give compound **13** using dimethylsulfate and potassium carbonate in acetone under reflux conditions. Chlorides **10** and **13** react with potassium salts of benzoic acids or of cinnamic acids in DMF at 110–120 ºC to give the corresponding ester derivatives **12** (Fig. 7). Adamantylbenzoic acids **15** were synthesized by reacting benzoic acids with 1-adamantanol in trifluoroacetic acid (TFA) under reflux conditions. Adamantylbenzoic acids **15** reacted with potassium hydroxide in methanol to afford the potassium salts. After methanol removal, the potassium salts react with corresponding ester derivatives **16** (Fig. 8) (42). The obtained biological results revealed that 3,4-methylenedioxy cinnamic acid ester of kojic acid (**12c**) exhibited more potent inhibitory effect on tyrosinase than kojic acid. The structure of compound**12c** (Fig. 9) comprises two main parts: a chelating part of kojic acid and a hydrophobic part of cinnamic acid. However, the hydroxybenzoate derivatives of kojic acid containing the adamantyl moiety showed no inhibitory activity. The reason for no inhibition may be either steric hindrance of the adamantyl moiety or insufficient copper chelating tendency

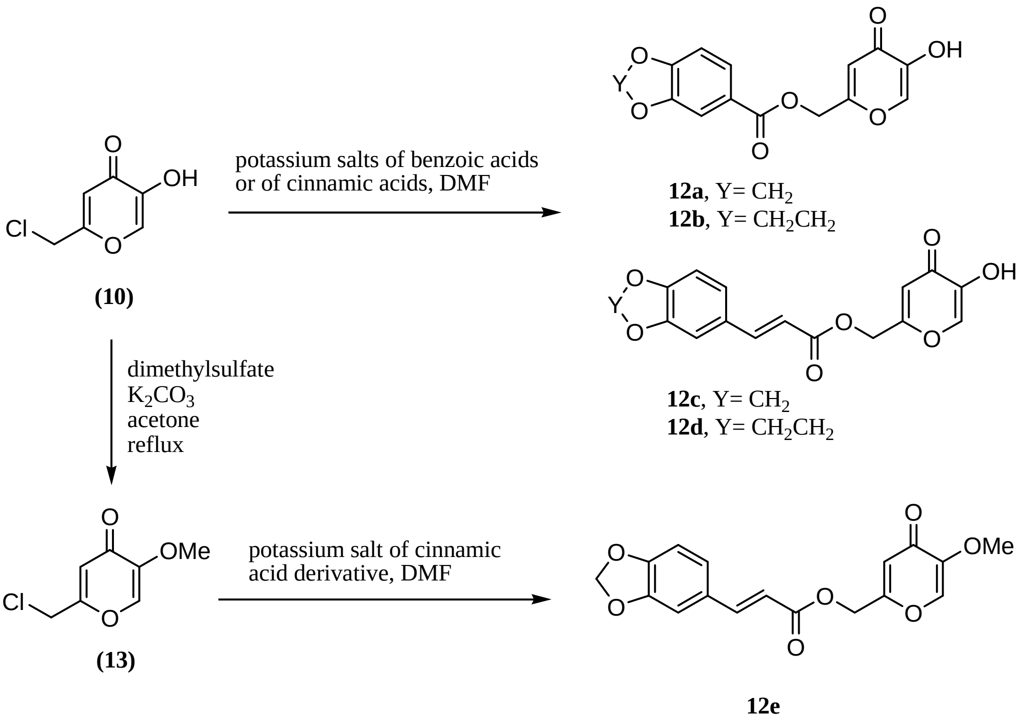


Figure 7 Synthesis of kojic esters **12a-e**

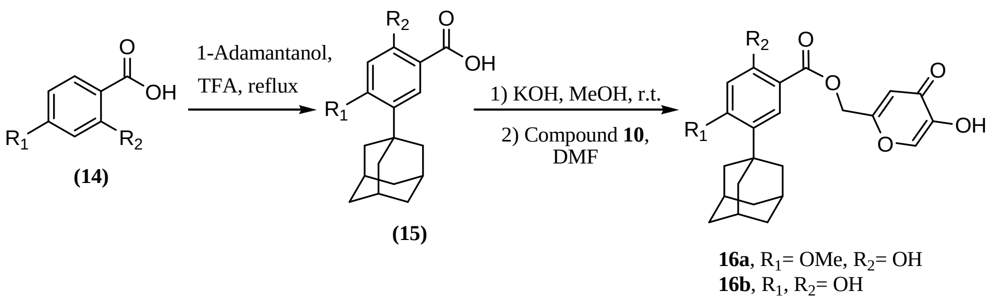
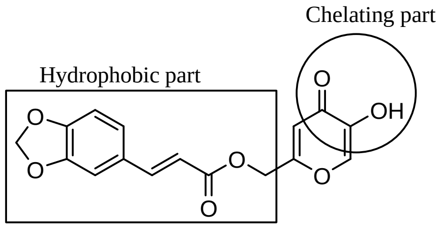


Figure 8 Synthesis of kojic-benzoate esters **16** containing adamantyl moiety



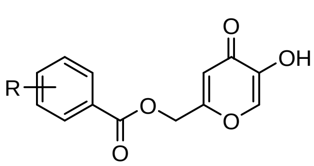
(12c)

Figure 9 Structure of 3,4-methylenedioxycinnamic acid ester of kojic acid (**12c**)

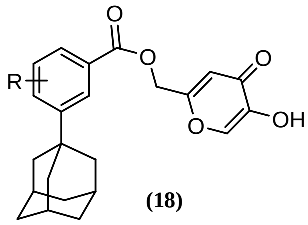
between kojic acid and the 2-hydroxy benzoic acid moiety.

In another study, benzoate ester derivatives of kojic acid, with and without adamantyl moiety were synthesized (Fig. 10).

Benzoate derivatives **17** that did not contain an adamantyl moiety showed potent tyrosinase inhibitory activities. In contrast, compounds **18** showed potent depigmenting activity without tyrosinase inhibitory activity. This is the first study showing the depigmenting activity of kojic acid derivatives without tyrosinase inhibitory activity (43). Cho and co-workers have synthesized cinnamate derivatives of kojic acid by various esterification methods, for use as depigmenting agents. In this report, to obtain the cinnamate ester of kojic acid (compound **12c**), the nucleophilic addition of the potassium salt of cinnamic acid to kojyl chloride was carried out (Fig. 11). Interestingly, the side product (**20**) showed



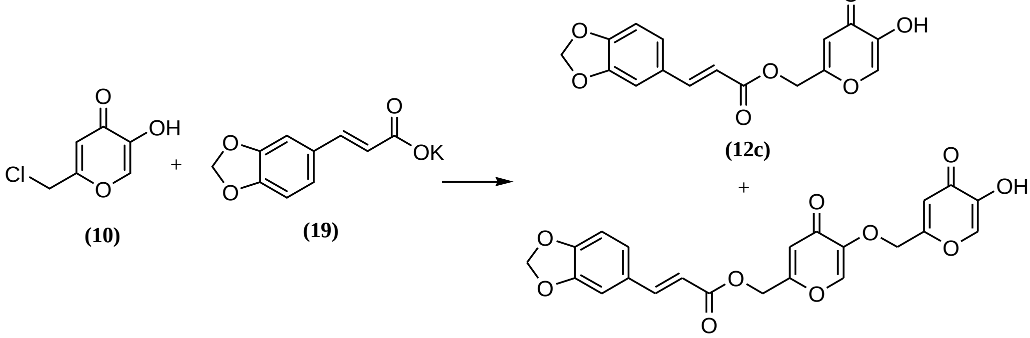
(17)



(18)

R= 4-OMe; 4-OH; 2,4-$(OH)_2$; 2-OH, 4-OMe; 2,4-$(OMe)_2$

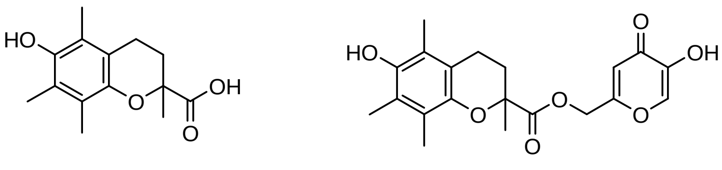
Figure 10 Structure of benzoic acid esters of kojic acid (**17** and **18**)



(10) **(19)** **(12c)** Byproduct **(20)**

Figure 11 Preparation of cinnamate derivatives of kojic acid (compounds **12c** and **20**) more potent depigmenting activity (IC_{50} = 23.51 μM) than compound **12c** (IC_{50} > 100 μM) which is the parent compound of the side product. However, it has no tyrosinase inhibitory activity (44).

A novel kojic acid derivative containing trolox (**21**), namely (±)-5-hydroxy-4-oxo-4*H*-pyran-2-yl-methyl 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylate (**22a**, Fig. 12), was synthesized (45). Indeed, the two biologically active compounds, kojic acid and trolox, were conjugated via an ester bond as they are expected to have dual action. The antioxidant activity and the tyrosinase inhibitory activity of kojic acid derivative **22a** on melanogenesis were evaluated. Compound **22a** exhibited potent tyrosinase inhibitory activity and radical scavenging activity. Limited structure–activity relationship (SAR) investigations indicated that the tyrosinase inhibitory activity may originate from the kojic acid moiety, and the radical scavenging activity may be due to the phenolic hydroxyl group of trolox. Compound **22a** also exhibited potent depigmenting activity in a cell-based assay. The limited SAR investigations revealed that the depigmenting activity of **22a** may be due to the synergistic activities of kojic acid and its trolox moiety (45). As presented in Fig. 13, kojyl chloride derivatives **10** or **13** reacted with potassium salts of trolox, 4-hydroxybenzoic acid or 6-hydroxynaphthoic acid in DMF at 110–120 ºC to give the corresponding ester derivatives **22a–d** (45).



(21), Trolox **(22a)**

Figure 12 Structures of antioxidant compound trolox (**21**) and kojic acid-trolox conjugate (**22a**)

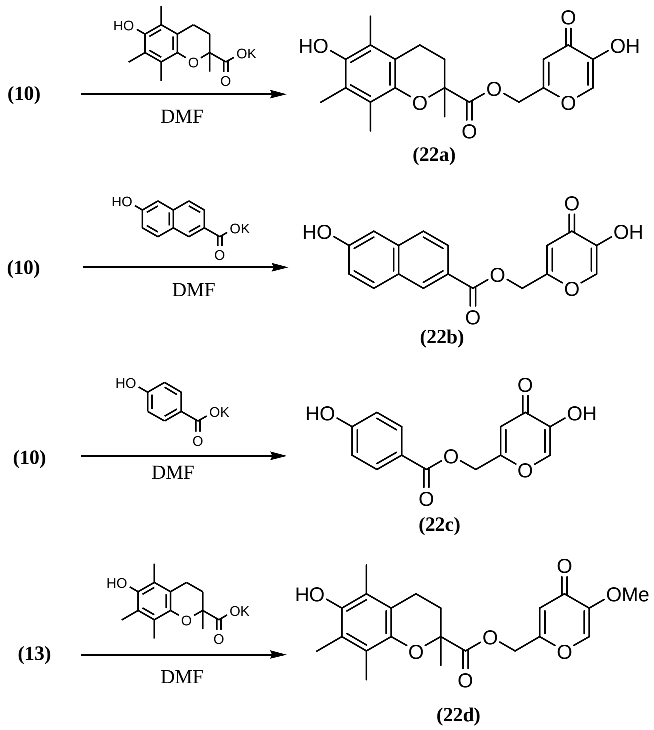


Figure 13 Synthesis of kojic acid-trolox conjugate and related compounds

Kojic acid derivatives conjugated with amino acids

In a report by Noh et al. (46), kojic acid has been coupled with amino acids to obtain kojic acid–amino acid amide conjugates **24** as new stable tyrosinase inhibitors (Fig. 14). Firstly, the primary alcohol of kojic acid was reacted with 1,1′-carbonyldiimidazole (CDI) and then coupled to the resin-bounded amino acids. In this reaction, the kojyl moiety is connected to the amino acid via a carbamate linker. After cleavage of the kojic acid–amino acid amide (**24**, KA-AA-NH_2) from the resin, it was characterized by MALDI-TOF mass spectroscopy. The conjugates of different amino acid amides with kojic acid were evaluated for their inhibitory activity on mushroom tyrosinase. The results showed that most of conjugates had better inhibitory activity than the parent molecule kojic acid. When amino acids such as phenylalanine, tryptophan, tyrosine, and histidine, which possess aromatic side chains, were conjugated to kojic acid, the tyrosinase inhibitory activity was enhanced dramatically. Noh et al. suggested that the aromatic residue of mentioned amino acids may contribute to the binding of the inhibitor to the hydrophobic pocket of the enzyme. Further studies showed that kojic acid–phenylalanine amide conjugate (**24a**, KA-F-NH_2, Fig. 15) showed the strongest inhibitory activity, which was maintained for over 3 months at 50 ºC, and acted as a noncompetitive inhibitor (46).

Kim group synthesized a series of kojic acid-tripeptides by solid-phase parallel synthesis and evaluated them as tyrosinase inhibitors (47). As depicted in Fig 16, the resin-bound tripeptides reacted with activated kojic acid **23**. After cleavage, the kojic acid-tripeptide conjugates **26** were obtained in good yields. Most of the kojic acid-tripeptide conjugates exhibited more potent tyrosinase inhibitory activities than kojic acid. The most potent compound (kojic acid-FWY) was about 100-fold more potent than kojic acid. Furthermore , it was less toxic than kojic acid and its storage

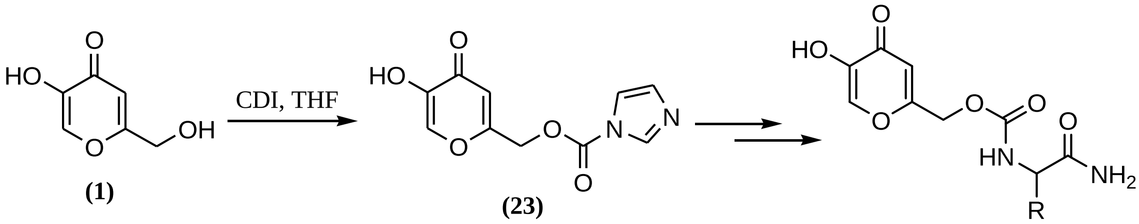


Figure 14 Synthesis of kojic acid–amino acid amide conjugates (**24**, KA-AA-NH_2)

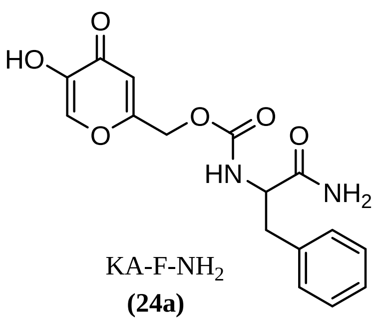


Figure 15 Structure of kojic acid–phenylalanine amide conjugate (**24a**)

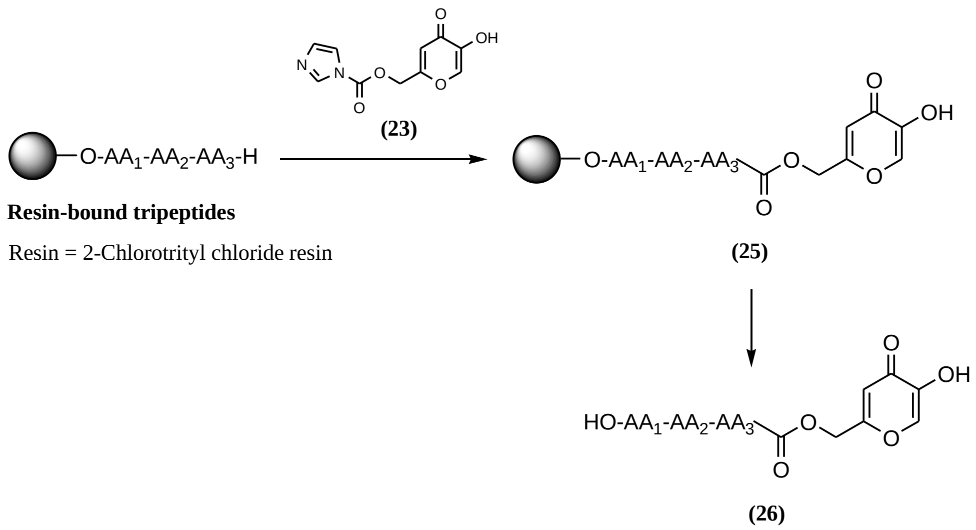


Figure 16 Solid-phase synthesis of kojic acid-tripeptides **26**

stabilities was approximately 15-times higher than that of kojic acid. In addition, this research group demonstrated that kojic acid-tripeptide amides have similar tyrosinase inhibitory relative to kojic acid-tripeptide free acids, while exhibit more favorable storage stability. These findings indicate the importance of C-terminal amide form in kojic acid-peptide conjugates.

Compounds containing two molecules of kojic acid

In a study by Kobayashi et al., various amino acid derivatives of kojic acid (**28**, Fig 17) were synthesized to improve the tyrosinase inhibitory activity of kojic acid (48). The *N*-kojic–amino acid **27** were synthesized starting from kojic acid and appropriate amino acid by using DSC (*N,N*'-disuccinimidylcarbonate) and DMAP (4-dimethylaminopyridine). Subsequent esterification of another molecule of kojic acid with compound **27** gave the target compounds **28**. Almost all synthesized compounds were more active than kojic acid. In general, *N*-kojic–amino acid–kojiate **28** was found to have a higher inhibitory activity than *N*-kojic–amino acid **27**. Among them, the *N*-kojic-L-phenylalanylkojiate was the most potent compound. It was 380 times more potent

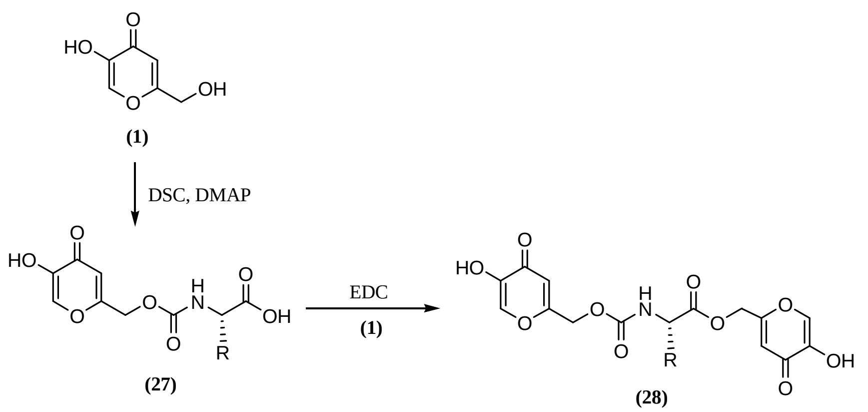


Figure 17 Synthesis of *N*-kojic–amino acid–kojiate (**28**)

than kojic acid. The inhibition mechanism of these derivatives is considered to be non-competitive which is similar to that of kojic acid (48). In another study, two molecules of kojic acid were connected by various linkers containing chemical bonds such as ester, amide and thioether (Fig. 18) (49). Chlorokojic acid (**10**) was reacted with sodium azide in DMF and subsequently converted to kojyl amine HBr **29**. The coupling of compound **29** with succinyl chloride in the presence of triethylamine in THF afforded di-kojylsuccinic amide **30**. The nucleophilic substitution of chlorokojic acid (**10**) with potassium salt of kojyl succinic acid gave di-kojylsuccinate **31**. The reaction of compound **10** with dithiols in the presence of TEA afforded thioethers **32** (Fig. 18).

The synthesized dimmers of kojic acid (**30-32**) were evaluated against tyrosinase enzyme and melanin formation in melan-a melanocytes. Among them, dithioether derivatives (**32a-c**) showed the highest inhibitory activity. The obtained results showed that the dithioether linker and its flexibility are important for improving anti-melanogenic activity. The propylene thioether compound **32b** with IC_{50} value of 1.97 µM was the most active inhibitor against tyrosinase enzyme. It was about 25-fold more potent than kojic acid. In malan-a cell based assay, butylene dithioether derivative **32c** exhibited superior inhibitory activity of melanin synthesis, being approximately 1000 times more potent than kojic acid (49). Moreover, compound **32b** exhibited the most potent inhibitory activity of NO production in LPS activated macrophages (50).

Rho et al. further investigated the structure–activity relationship of kojic acid thioethers by preparing mono-kojyl thioethers **33**, sulfoxides **34**, and sulfones **35** (Fig. 19) (51). Kojyl thioethers **33** were prepared by the reaction of kojyl chloride **10** with potassium salts of thiols. Mono-oxidation of the thioethers **33** with MCPBA (*m*-chloroperbenzoic acid) in CH_2Cl_2 produced sulfoxide derivatives **34**. The treatment of thioether derivatives **33** with oxone in a mixture of MeOH/H_2O afforded sulfone derivatives **35**. In the tyrosinase inhibition bioassay , the

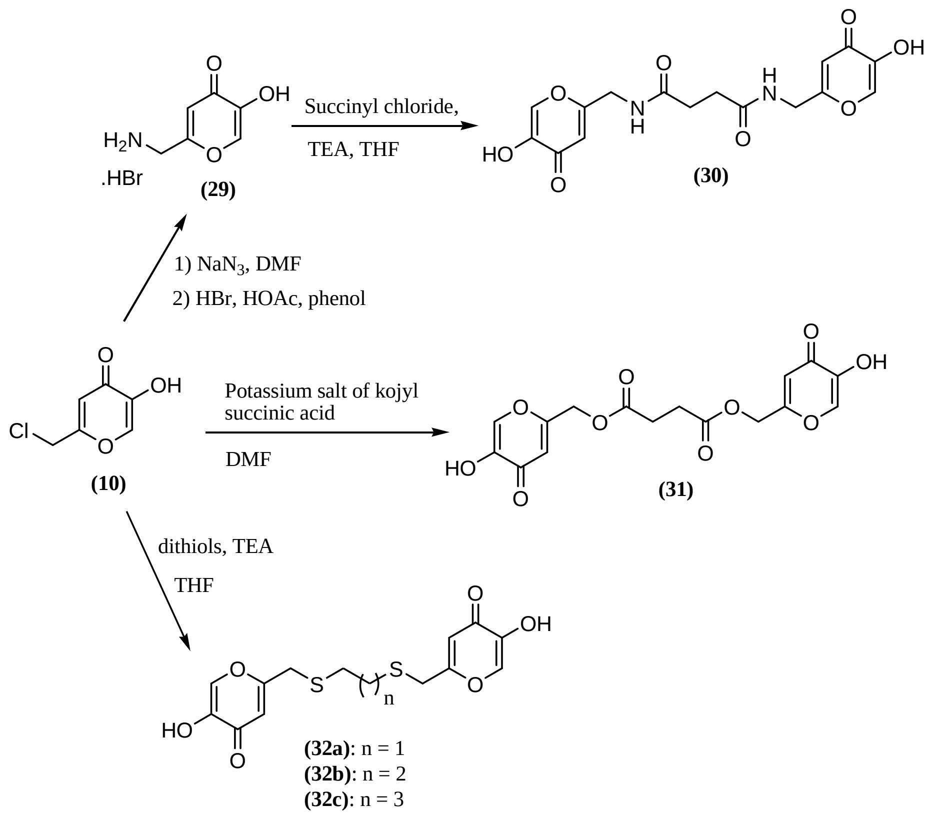


Figure 18 Synthesis of kojic acid dimmers **30-32**

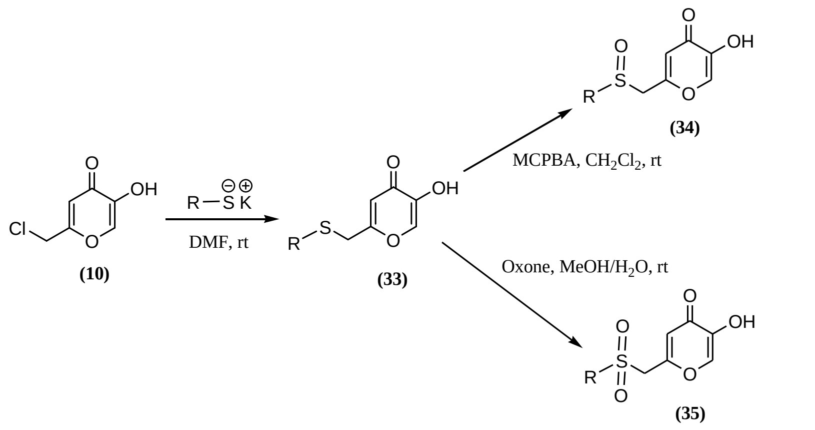


Figure 19 Synthesis of mono-kojyl thioethers **33**, sulfoxides **34**, and sulfones **35**

pentyl, hexyl, and cyclohexyl thioethers exhibited potent inhibitory activity. In contrast, sulfoxide and sulfone derivatives **34** and **35** showed decreased activity. The pentyl thioether derivative with IC_{50} value of 0.097 µM was the most potent compound against tyrosinase. The obtained results for inhibitory activity of NO production were similar to those of tyrosinase inhibition. Stilbene-like kojic acid derivative **42** was synthesized by joining two pyrone rings through an ethylene linkage by Horner-Emmons reaction of the protected aldehyde **37** with phosphonate **40** (Fig. 20). Both

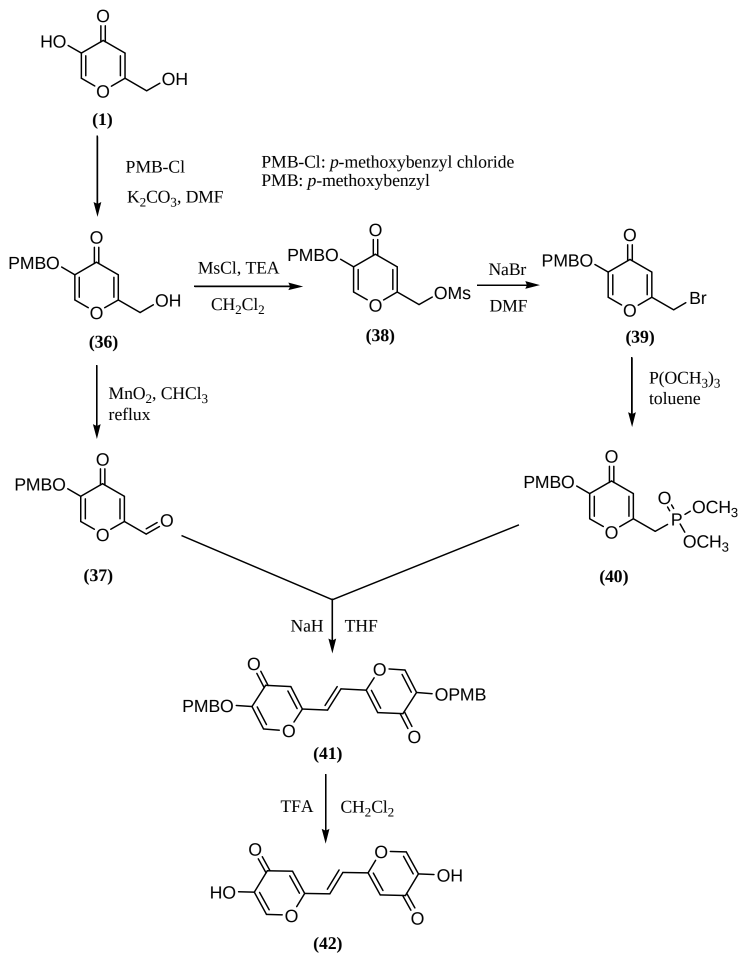


Figure 20 Synthesis of stilbene-like kojic acid derivative **42**

intermediates **37** and **40** were derived from kojic acid as depicted in Fig. 20. The enzymatic assay revealed that the tyrosinase inhibitory activity of compound **42** was about 8 times more potent than that of kojic acid. This compound also exhibited significant melanin synthesis inhibitory activity in cell-based assay. The obtained results for dimeric compound **42** compared to kojic acid indicate that the connection of two pyrone rings of kojic acid through a suitable linker can be an useful strategy for finding new potent tyrosinase inhibitors (3).

C-2 Side chain-modified kojic acid derivatives

Chemically, the 2-(hydroxymethyl) side chain in the kojic acid structure is a good site for oxidation to related aldehyde. With a key intermediate aldehyde in hand, diverse derivatives can be prepared. As shown in Fig. 20, for oxidation of primary alcohol in kojic acid structure, the enolic OH should be protected with a suitable group such as *p*-methoxybenzyl. The oxidation of protected kojic acid **36** with MnO_2 resulted in protected aldehyde **37**. Kang et al. used this intermediate for synthesis of pyronyl-acrylic acid esters **44**, which share structural features of kojic acid and hydroxylated cinnamic acid (caffeic acid, **43**, Fig. 21) (52). Horner-Emmons condensation of aldehyde **37** with compound **45** gave methyl ester **46**, which hydrolyzed with LiOH in aqueous THF to afford acid **47**. Compound **47** was *O*-alkylated with appropriate alkyl iodides or alkyl tosylates. The target compounds **44** were obtained by removing of the PMB-protecting group using trifluoroacetic acid (TFA) in CH_2Cl_2 (Fig. 22).

The abilities of synthesized compound **44** to inhibit tyrosinase and melanin production were evaluated by Kang et al. Among the tested derivatives, compounds derived from diethylene glycol were found to inhibit melanin production at 20 µg/ml. It should be noted that in this test, kojic acid inhibited melanin production at 200 µg/ml (52).

Yi et al. have synthesized thiosemicarbazone analogs of kojic acid (**52**) as tyrosinase inhibitors (53). The synthesis of the target compounds **52a,b** was outlined in Fig. 23. Firstly *O*-methyl kojic acid (**49**) was oxidized to aldehyde **50** by using MnO_2. The *O*-methyl group was removed by $AlCl_3$ to give compound **51**. The condensation reaction of carboxaldehydes **50** or **51** with the thiosemicarbazide in anhydrous ethanol in the presence of acetic acid afforded corresponding thiosemicarbazone analogs **52**. The inhibitory evaluation of

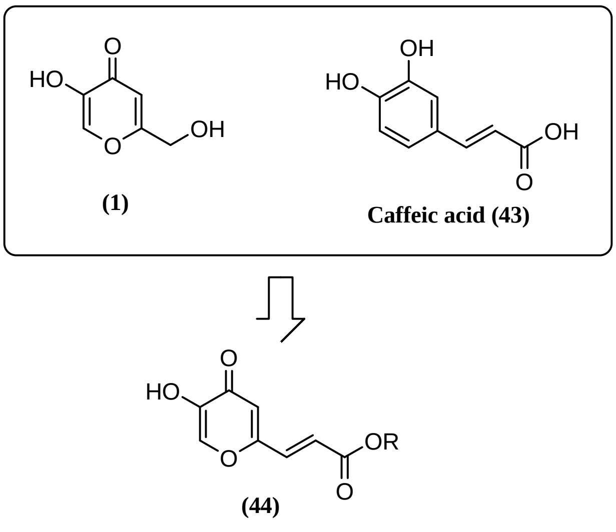


Figure 21 Design of hybrid compounds pyronyl-acrylic acid esters **44**

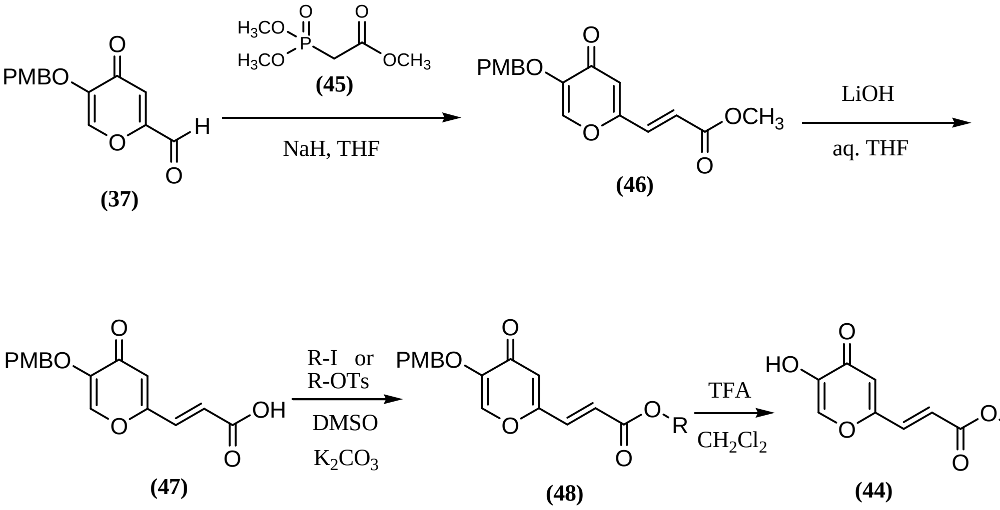


Figure 22 Synthesis of hybrid compounds pyronyl-acrylic acid esters **44**

compounds **52a,b** against commercial mushroom tyrosinase revealed that *O*-methylated compound **52a** showed no inhibitory activity, while thiosemicarbazone analog **52b** bearing a free enolic group exhibited high activity against mushroom tyrosinase (IC_{50} = 11 µM). The latter compound was about 9-fold more potent than the parent compound kojic acid (53).

Miscellaneous derivatives

The topical formulations of kojic acid are used as skin-lightening agent. However, it is hardly absorbed through the lipid membrane of its target sites, the melanocytes due to its hydrophilic character (54). In some investigations, it has been attempted to connect the kojic acid to a suitable carrier.

Kim et al. synthesized a kojic acid analog

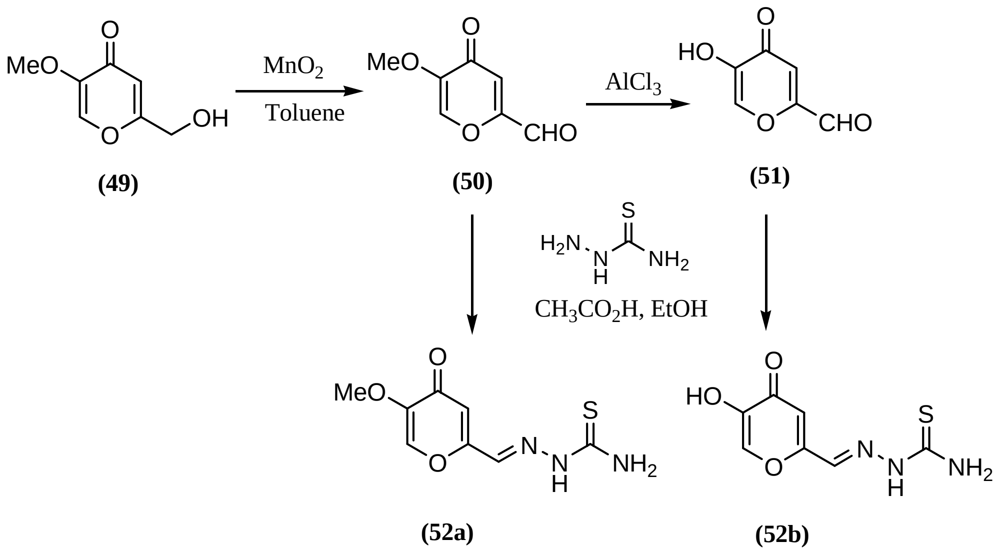


Figure 23 Synthetic pathway to thiosemicarbazone analogs of kojic acid (**52**)

(kojyl-APPA, **56**) containing 5-[(3-aminopropyl)phosphinooxy]- moiety as stable derivative of kojic acid. Compound **56** was prepared by the reaction of kojic acid with 2-chloro-[1,3,2] oxazaphosphinane 2-oxide (**54**) in the presence of TEA in $CHCl_3$/EtOH, followed by hydrolysis in acidic medium of H_2O/MeOH (Fig. 24). Interestingly, the alcoholic OH group of kojic acid did not take part in reaction with compound **54**. The effects of compound **56** on tyrosinase activity and melanin synthesis were evaluated by Kim and co-workers. The masked form of kojic acid **56** displayed higher stability than kojic acid. Also, its permeation through the skin was about 8-times more than kojic acid. Compound **54** showed no tyrosinase inhibition effect compared with kojic acid *in vitro*, however displayed the same inhibitory effect as kojic acid on melanin synthesis in mouse melanoma and normal human melanocytes. It seems that compound **56** is converted to kojic acid in living cells (55). In another study, Manosroi and co-workers have investigated the entrapment of kojic acid and its oleate ester. Kojic oleate (**57**) was prepared starting from kojic acid and oleic acid in CH_2Cl_2 by using DCC (*N*,*N*'-dicyclohexylcarbodiimide) and DMAP (4-(*N*,*N*-dimethylamino)pyridine) (Fig. 25). In this study, the entrapment efficiencies of kojic acid and kojic oleate in the vesicles were investigated by dialysis and column chromatography, respectively. The obtained results indicated that kojic oleate could be intercalated in the bilayer structure of the vesicles composed of amphiphile (Span 60, Tween 61 or DPPC)/cholesterol/dicetyl phosphate at molar ratio of 9.5:9.5:1.0. In general, they concluded that the esterification of kojic acid improved its entrapment in the vesicles (56).

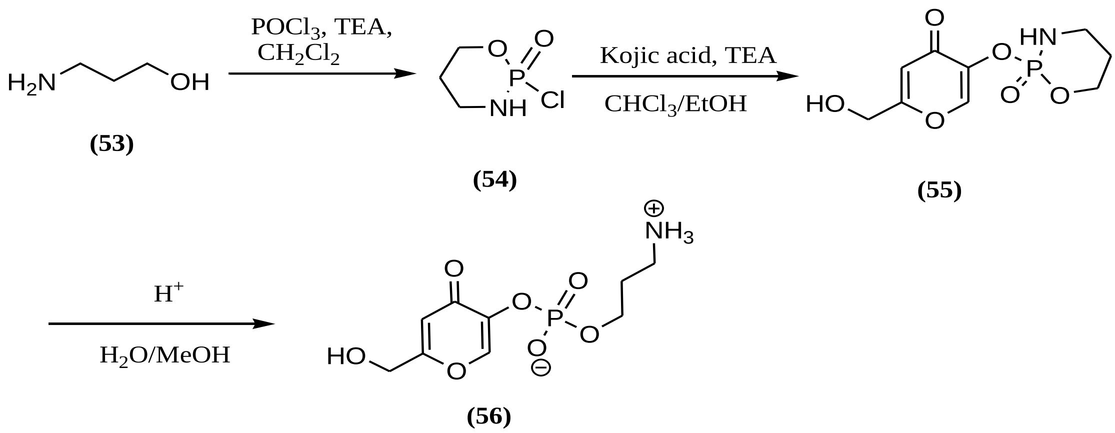


Figure 24 Synthesis of 5-[(3-aminopropyl)phosphinooxy]-kojic acid (**56**)

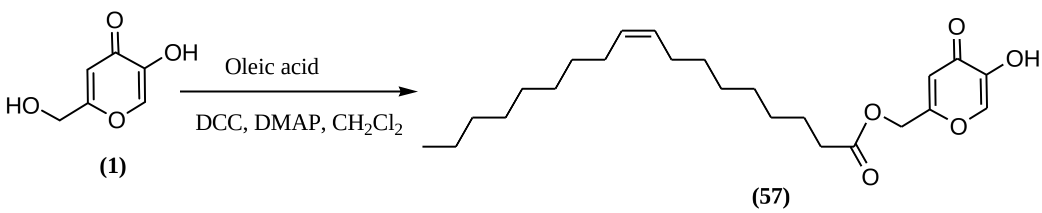


Figure 25 Synthesis of kojic oleate (**57**)

Conclusion

Kojic acid is a small-molecule with tyrosinase inhibitory activity, which has been used a skin-lightening agent. This agent is the most intensively studied inhibitor of tyrosinase; however it has unsatisfactory inhibitory activity, insufficient stability band unwanted side effects. To overcome these disadvantages, researchers have attempted to design new analogs of kojic acid with higher potency, satisfactory stability and safety. Diverse modifications on this small-molecule have been made to find new tyrosinase inhibitors. The main modifications were conversion of the γ-pyranone to 4-pyridinone, esterification of 2-(hydroxymethyl) group, C-2 side chain-modification, and conjugation of kojic acid with amino acids.

Conflict of interest statement

The authors claim that they have no conflicting interest in this study.

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