catalysts

Editorial

# Enzymes in Biomedical, Cosmetic and Food Application

**Chia-Hung Kuo [1,2,*], Hui-Min David Wang [3,*] and Chwen-Jen Shieh [4,*]**

1 Department of Seafood Science, National Kaohsiung University of Science and Technology, Kaohsiung 811, Taiwan
2 Center for Aquatic Products Inspection Service, National Kaohsiung University of Science and Technology, Kaohsiung 811, Taiwan
3 Graduate Institute of Biomedical Engineering, National Chung Hsing University, Taichung 402, Taiwan
4 Biotechnology Center, National Chung Hsing University, Taichung 402, Taiwan
* Correspondence: kuoch@nkust.edu.tw (C.-H.K.); davidw@dragon.nchu.edu.tw (H.-M.D.W.); cjshieh@nchu.edu.tw (C.-J.S.); Tel.: +886-7-361-7141 (ext. 23646) (C.-H.K.); +886-4-2284-0733 (ext. 651) (H.-M.D.W.); +886-4-2284-0450 (ext. 5121) (C.-J.S.)

Enzymes play an important role in biomedical, cosmetic and food applications, and their effects are mainly related to their specific reactions and catalytic activity [1]. Enzymes are important catalysts for promoting the formation of complex organic molecules during the synthesis of medical compounds [2]. Enzymes can be used to synthesize various drugs, including antibiotics, hormones, anticancer agents, etc. In chiral synthesis, enzymes are essential for the preparation of drugs with high chirality, as they can specifically catalyze the synthesis of chiral molecules [3]. Moreover, the highly selective catalysis of specific functional groups and bonding sites by enzymes reduces the occurrence of side reactions and helps improve the purity of a product [4]. Through protein engineering technology, the structure of an enzyme can be modified to improve its activity or give it new catalytic properties to adapt to specific synthetic reaction conditions. Enzyme-mediated synthetic reactions are generally greener and therefore considered more environmentally sustainable than conventional chemical approaches. This is attributed to the ability of enzymes to operate under milder conditions, resulting in less energy consumption and a reduced generation of toxic by-products [5]. High yields are frequently attained in synthesis processes when enzymes serve as catalysts, thereby enhancing overall efficiency. The selective catalysis of an enzyme allows for the generation of high-purity products, reducing the need for subsequent purification steps [4]. Various enzymes are known for their applications in cosmetics and the food industry. Proteases can remove a keratin layer attached to the skin's surface and make the skin softer. Catalase and superoxide dismutase can be used to inhibit oxidation reactions in cosmetics and slow down the aging process of products [6]. In food processing, amylase is used to break down starch, for example, in the making of syrup, bread and beer [7,8]. Protease can hydrolyze proteins to produce functional peptides [9], make cheese and tenderize meat [10]. Enzymes can also be used to increase the extraction efficiency of functional compounds in food. This Special Issue investigates enzymes for biomedical, cosmetic and food applications.

Shih's article (contribution 1) discusses three enzyme hydrolysates, Dur-A, Dur-B and Dur-C, derived from *Durvillaea antarctica* biomass using viscozyme, cellulase and α-amylase, respectively. Through $^{1}$H-NMR analysis, these extracts were found to contain fucose-containing sulfated polysaccharides with distinct structural qualities. Notably, Dur-A, Dur-B and Dur-C demonstrated significant antioxidant activities, as confirmed by DPPH, ABTS and ferrous ion-chelating analyses. Moreover, these extracts displayed the ability to inhibit key enzymes associated with metabolic syndrome, including angiotensin I-converting enzyme (ACE), α-glucosidase, α-amylase and pancreatic lipase. In particular, Dur-B exhibited superior antioxidant and antimetabolic syndrome effects compared to the other extracts. These findings suggest that these enzyme hydrolysates, especially Dur-B,



**Citation:** Kuo, C.-H.; Wang, H.-M.D.; Shieh, C.-J. Enzymes in Biomedical, Cosmetic and Food Application. *Catalysts* **2024**, *14*, 162. https://doi.org/10.3390/catal14030162

Received: 2 February 2024
Accepted: 19 February 2024
Published: 22 February 2024

are promising natural antioxidants and antimetabolic syndrome agents for applications in various health-oriented products like food, cosmetics and nutraceuticals.

Weng's article (contribution 2) reports the immobilization of recombinant endoglucanase (CelA) on regenerated cellulose (RC) membrane modified using two different approaches, one to generate the immobilized metal ion affinity membranes RC-EPI-IDA-$Co^{2+}$ (IMAMs) for coordination coupling and another to develop the aldehyde functional group membranes RC-EPI-DA-GA (AMs) for covalent bonding. A recombinant endoglucanase (CelA) originating from the cellulosome of *Clostridium thermocellum* was expressed in *Escherichia coli* and then immobilized on RC-EPI-IDA-$Co^{2+}$ (IMAM) and RC-EPI-DA-GA (AM) membranes. The characteristics of the immobilized enzyme, along with its preliminary purification, were assessed in comparison to a free enzyme. Moreover, an appreciable enzyme activity till 5 cycles of reusability achieved in this study paves a way for developing an economical process.

Sharma's article (contribution 3) provided a detailed overview of the production of industrial enzymes from microbes using agro-industrial food waste. Enzymes are versatile biocatalysts with immense potential to transform the food industry and lignocellulosic biorefineries. Microbial enzymes offer cleaner and greener methods of producing fine chemicals and compounds. The review highlights novel strategies for food enzyme immobilization and their potential applications in the food industry. Moreover, deeper insights into the development of engineered enzymes for sustainably processing waste biomass into diverse bioproducts are highlighted. In short, this review discusses recent developments in the production of essential industrial enzymes from agro-industrial food waste and the application of inexpensive immobilization and enzyme engineering approaches for sustainable development.

Thirametoakkhara's article (contribution 4) explored the production and applications of two endoxylanases, Xyn45 and Xyn23, derived from the bacterium *Bacillus halodurans*. Xyn45 was obtained through recombinant *E. coli*, while a mixture of nonrecombinant Xyn45 and Xyn23 was acquired from *B. halophilus*. The study revealed that combining these enzymes led to enhanced catalytic activity compared to using Xyn45 alone. Furthermore, the researchers investigated the impact of xylooligosaccharides (XOS) derived from oil palm empty fruit bunches on the growth and metabolism of probiotic strains, *Bifidobacteria* and *Lactobacilli*. The findings demonstrated that XOS, particularly xylobiose, effectively induced the secretion of endoxylanases in *B. halophilus*, highlighting their potential for industrial applications. This research offers valuable insights into the synergistic effects of endoxylanases and the utilization of XOS to promote the growth of beneficial bacteria.

Cunha's article (contribution 5) prepared a new heterogeneous biocatalyst by applying the cross-linking enzyme aggregates (CLEAs) technique to non-commercial β-glucosidase from *Aspergillus niger* produced via solid-state fermentation. The effects of relevant factors on the immobilization process, such as the soy protein isolate and glutaraldehyde concentrations, were evaluated using a central composite rotatable design (CCRD). The influence of certain factors on the hydrolytic activity of the immobilized enzyme (pH, temperature and thermal stability) was evaluated, and it was compared with its soluble form in order to contribute to the advancement of enzyme immobilization technology and consolidate the results of utilizing β-glucosidase in bioprocesses.

Ma's article (contribution 6) presented a significant advancement in biocatalysis by establishing a bienzymatic, parallel cascade that combines aryl alcohol oxidases with peroxygenases for the selective oxidation of benzylic alcohols to corresponding aromatic aldehydes. Aromatic aldehydes are important aromatic compounds for the flavor and fragrance industries. A parallel cascade combining aryl alcohol oxidase from *Pleurotus eryngii* (PeAAOx) and unspecific peroxygenase from the basidiomycete *Agrocybe aegerita* (AaeUPO) was used to convert aromatic primary alcohols into high-value aromatic aldehydes. In a partially optimized system, up to an 84% conversion rate of 50 mM veratryl alcohol into veratryl aldehyde was achieved through a self-sufficient aerobic reaction. The findings offer insights into industrial applications in flavor compound synthesis.

Nabil-Adam's article (contribution 7) investigated the antidiabetic, anti-inflammatory and antioxidant competency of *Jania rubens* polyphenolic extract (JRPE) through its interactions with $\alpha$-amylase, lipase and trypsin enzymes. An HPLC analysis revealed the dominance of twelve polyphenolic compounds. The antioxidant and antibacterial activities demonstrated by *Jania rubens* extract suggest its potential as a natural source for combating oxidative stress and microbial infections. Computational analyses further corroborated the ability of these polyphenolics to form complexes with digestive enzymes. The findings provide a foundation for understanding the anti-obesity and antidiabetes characteristics of *Jania rubens* polyphenolic compounds.

Xing's article (contribution 8) presented several important findings regarding the enzymatic modification of bovine lactoferrin (bLf) and its potential applications. Laccase-mediated pectin–ferulic acid conjugate (PF) and transglutaminase (TG) treatments were studied for their ability to enhance the diversity and abundance of bLf peptide fragments during in vitro simulated gastrointestinal digestion. The encapsulation of bLf by PF led to enhanced diversity and abundance of active peptide fragments, especially for long-chain species. The TG treatment on the lactoferrin–pectin–ferulic acid conjugate (LfPFTG) demonstrated an influence on the final gastrointestinal digest, highlighting the potential of TG-induced crosslinking between bLf chains. The identified peptides, with enhanced abundances in the LfPFG digest, hold promise for various applications in nutraceuticals, clinical therapy or as components in antibacterial vaccines.

Statkevi˘cius's article (contribution 9) identified 11 full-length fold type IV aminotransferases (ATs) that were successfully expressed and used for substrate profiling. Three of them (AT-872, AT-1132 and AT-4421) were active toward (R)-methylbenzylamine. The research showed the high thermostability of specific ATs, such as AT-872 and AT-1132, along with their broad substrate spectrum, particularly AT-872 and AT-4421. The purified proteins showed activity with L- and D-amino acids and various aromatic compounds, such as (R)-1-aminotetraline. The study demonstrated the close relationship between AT-1132 and branched-chain amino acid transaminases (BCATs) through the protein sequence, and its specificity for amino acceptors was similar to that for BCATs, which are most active with $\alpha$-ketoglutarate. AT-872 and AT-4421 were homologous to the D-amino acid transaminases (DAATs), which prefer pyruvate as an amino acceptor. These properties make these enzymes potential biocatalysts for the production of valuable amino compounds. These findings offer valuable insights into the potential of these enzymes in the synthesis of chiral drugs and the environmentally friendly preparation of chiral amines.

Gupta's article (contribution 10) reported the significance of various natural compounds and extracts in the fields of photoprotection and skincare. It extensively covers studies on microalgae, peptides and antioxidants, highlighting their potential applications in inhibiting melanogenesis, reducing oxidative stress and protecting against skin cancer. The role of antioxidants, such as vitamins C and E, in enhancing the photoprotective properties of sunscreens was illustrated. It discusses the relevance of various topical options containing DNA repair enzymes, antioxidants and growth factors in preventing skin aging and cancer. Moreover, the potential of photolyases, which are enzymes that repair DNA damage caused by ultraviolet radiation, and their uses in skincare are discussed.

Tran's article (contribution 11) investigated the metabolic processes and pharmacological effects of the synthetic cannabinoid JWH-019 ((1-Hexyl-1H-indol-3-yl)-1-naphthalenyl-methanone). Through a series of experiments utilizing human liver microsomes (HLMs) and recombinant P450 enzymes, 6-OH JWH-019 was found to be the primary oxidative metabolite in HLMs, and CYP1A2 was the principal enzyme responsible for the monohydroxylation of JWH-019. The research not only enhances our understanding of the metabolic pathways of this synthetic cannabinoid but also provides crucial insights into its pharmacological activity.

de Carvalho-Silva's article (contribution 12) investigated the stability and performance of immobilized tannase, an enzyme derived from *Aspergillus ficuum*, using two different methods: their entrapment in calcium alginate beads and their covalent attachment to magnetic nanoparticles. The kinetic and thermodynamic behavior of the enzyme under various conditions was studied. The findings indicated that the magnetic nanoparticle immobilization method results in the lowest activation energy, suggesting its suitability for cost-effective industrial applications. Additionally, shelf-life tests revealed that the enzyme remained stable for a longer period of time when immobilized on magnetic nanoparticles.

Arnold's article (contribution 13) showed the two-step enzymatic synthesis of (S)-norlaudanosoline ((S)-NLS), a crucial precursor for opioids, using lyophilized whole-cell biocatalysts containing ω-transaminase (Tam) and norcoclaurine synthase (NCS) enzymes. The research revealed that the addition of a substantial quantity of cells containing the NCS enzyme resulted in a high yield of an optically pure chiral product. The cells were immobilized to enable their retention in fixed-bed reactors for batch and flow system comparisons. The flow system produced a substantial amount of (S)-NLS, allowing for its long-term practical application for opioid precursor synthesis.

**Funding:** This research received no external funding.

**Conflicts of Interest:** The authors declare no conflict of interest.

**List of Contributions**

1. Shih, M.-K.; Hou, C.-Y.; Dong, C.-D.; Patel, A.K.; Tsai, Y.-H.; Lin, M.-C.; Xu, Z.-Y.; Perumal, P.K.; Kuo, C.-H.; Huang, C.-Y. Production and characterization of *Durvillaea antarctica* enzyme extract for antioxidant and anti-metabolic syndrome effects. *Catalysts* **2022**, *12*, 1284.
2. Weng, Z.-H.; Nargotra, P.; Kuo, C.-H.; Liu, Y.-C. Immobilization of recombinant endoglucanase (CelA) from *Clostridium thermocellum* on modified regenerated cellulose membrane. *Catalysts* **2022**, *12*, 1356.
3. Sharma, V.; Tsai, M.-L.; Nargotra, P.; Chen, C.-W.; Kuo, C.-H.; Sun, P.-P.; Dong, C.-D. Agro-industrial food waste as a low-cost substrate for sustainable production of industrial enzymes: A critical review. *Catalysts* **2022**, *12*, 1373.
4. Thirametoakkhara, C.; Hong, Y.-C.; Lerkkasemsan, N.; Shih, J.-M.; Chen, C.-Y.; Lee, W.-C. Application of endoxylanases of Bacillus halodurans for producing xylooligosaccharides from empty fruit bunch. *Catalysts* **2022**, *13*, 39.
5. da Cunha, T.M.; Mendes, A.A.; Hirata, D.B.; Angelotti, J.A. Optimized conditions for preparing a heterogeneous biocatalyst via cross-linked enzyme aggregates (CLEAs) of β-glucosidase from *Aspergillus niger*. *Catalysts* **2022**, *13*, 62.
6. Ma, Y.; Li, Z.; Zhang, H.; Wong, V.K.W.; Hollmann, F.; Wang, Y. Bienzymatic cascade combining a peroxygenase with an oxidase for the synthesis of aromatic aldehydes from benzyl alcohols. *Catalysts* **2023**, *13*, 145.
7. Nabil-Adam, A.; Ashour, M.L.; Tamer, T.M.; Shreadah, M.A.; Hassan, M.A. Interaction of *Jania rubens* polyphenolic extract as an antidiabetic agent with α-amylase, lipase, and trypsin: In vitro evaluations and in silico studies. *Catalysts* **2023**, *13*, 443.
8. Xing, M.; Ji, Y.; Ai, L.; Xie, F.; Wu, Y.; Lai, P.F. Improving effects of laccase-mediated pectin–ferulic acid conjugate and transglutaminase on active peptide production in bovine lactoferrin digests. *Catalysts* **2023**, *13*, 521.
9. Statkevičius, R.; Vaitekūnas, J.; Stanislauskienė, R.; Meškys, R. Metagenomic type IV amino-transferases active toward (R)-methylbenzylamine. *Catalysts* **2023**, *13*, 587.
10. Gupta, A.; Singh, A.P.; Singh, V.K.; Singh, P.R.; Jaiswal, J.; Kumari, N.; Upadhye, V.; Singh, S.C.; Sinha, R.P. Natural sun-screening compounds and DNA-repair enzymes: Photoprotection and photoaging. *Catalysts* **2023**, *13*, 745.
11. Tran, N.; Fantegrossi, W.E.; McCain, K.R.; Wang, X.; Fujiwara, R. Identification of cytochrome P450 enzymes responsible for oxidative metabolism of synthetic cannabinoid (1-hexyl-1 H-indol-3-yl)-1-naphthalenyl-methanone (JWH-019). *Catalysts* **2023**, *13*, 1008.
12. de Carvalho-Silva, J.; da Silva, M.F.; de Lima, J.S.; Porto, T.S.; de Carvalho, L.B., Jr.; Converti, A. Thermodynamic and kinetic investigation on *Aspergillus ficuum* tannase immobilized in calcium alginate beads and magnetic nanoparticles. *Catalysts* **2023**, *13*, 1304.
13. Arnold, A.H.; Castiglione, K. Comparative evaluation of the asymmetric synthesis of (S)-norlaudanosoline in a two-step biocatalytic reaction with whole *Escherichia coli* Cells in batch and continuous Flow Catalysis. *Catalysts* **2023**, *13*, 1347.

## References


1. Deckers, M.; Deforce, D.; Fraiture, M.-A.; Roosens, N.H. Genetically modified micro-organisms for industrial food enzyme production: An overview. *Foods* **2020**, *9*, 326. [CrossRef] [PubMed]
2. Shen, Y.; Xia, Y.; Chen, X. Research progress and application of enzymatic synthesis of glycosyl compounds. *Appl. Microbiol. Biotechnol.* **2023**, *107*, 5317–5328. [CrossRef] [PubMed]
3. Eletskaya, B.Z.; Berzina, M.Y.; Fateev, I.V.; Kayushin, A.L.; Dorofeeva, E.V.; Lutonina, O.I.; Zorina, E.A.; Antonov, K.V.; Paramonov, A.S.; Muzyka, I.S. Enzymatic Synthesis of 2-Chloropurine Arabinonucleosides with Chiral Amino Acid Amides at the C6 Position and an Evaluation of Antiproliferative Activity In Vitro. *Int. J. Mol. Sci.* **2023**, *24*, 6223. [CrossRef] [PubMed]
4. Kuo, C.-H.; Tsai, M.-L.; Wang, H.-M.D.; Liu, Y.-C.; Hsieh, C.; Tsai, Y.-H.; Dong, C.-D.; Huang, C.-Y.; Shieh, C.-J. Continuous production of DHA and EPA ethyl esters via lipase-catalyzed transesterification in an ultrasonic packed-bed bioreactor. *Catalysts* **2022**, *12*, 404. [CrossRef]
5. Baek, Y.; Lee, J.; Son, J.; Lee, T.; Sobhan, A.; Lee, J.; Koo, S.-M.; Shin, W.H.; Oh, J.-M.; Park, C. Enzymatic Synthesis of Formate Ester through Immobilized Lipase and Its Reuse. *Polymers* **2020**, *12*, 1802. [CrossRef] [PubMed]
6. Rinnerthaler, M.; Bischof, J.; Streubel, M.K.; Trost, A.; Richter, K. Oxidative stress in aging human skin. *Biomolecules* **2015**, *5*, 545–589. [CrossRef] [PubMed]
7. Ben Rejeb, I.; Charfi, I.; Baraketi, S.; Hached, H.; Gargouri, M. Bread Surplus: A Cumulative Waste or a Staple Material for High-Value Products? *Molecules* **2022**, *27*, 8410. [CrossRef] [PubMed]
8. Dabija, A.; Ciocan, M.E.; Chetrariu, A.; Codină, G.G. Maize and Sorghum as Raw Materials for Brewing, a Review. *Appl. Sci.* **2021**, *11*, 3139. [CrossRef]
9. Ucak, I.; Afreen, M.; Montesano, D.; Carrillo, C.; Tomasevic, I.; Simal-Gandara, J.; Barba, F.J. Functional and Bioactive Properties of Peptides Derived from Marine Side Streams. *Mar. Drugs* **2021**, *19*, 71. [CrossRef] [PubMed]
10. Mohd Azmi, S.I.; Kumar, P.; Sharma, N.; Sazili, A.Q.; Lee, S.-J.; Ismail-Fitry, M.R. Application of Plant Proteases in Meat Tenderization: Recent Trends and Future Prospects. *Foods* **2023**, *12*, 1336. [CrossRef] [PubMed]