

Keratinases Applications and Biovalorization of Keratin as an Eco-Friendly and Efficient Management Biotechnological Applications

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Abstract:- Keratin-rich wastes generated from different anthropogenic activities especially during animal products processing, accumulate and cause serious environmental and human health problems. Different chemical and physiological treatment methods to remove these wastes generate other pollutants causing further degradation of the environment. Bio-valorisation of the wastes by some microorganisms using keratinolytic enzymes, keratinases, have shown to be safe, economic and eco-friendly method of converting keratin-rich wastes into value added products with numerous biotechnological applications. The fields of tissue medicine, pharmaceuticals, cosmetics and food industries have immensely profited from the products of keratinolysis. Recombinant technology and protein engineering further aided in keratin production and purification for future applications. Combination of valorised keratin with other components improves their quality and usage for different purposes. This article reviews the different procedures involved in bulk reduction and utilization of keratin-rich wastes for their effective management and applications.

Keywords:- Keratin, Keratinase, Keratinolysis, Bio-valorization, Appendageal, Nano-composites.

I. INTRODUCTION

The need for food security and balanced diet led to increase in livestock production, slaughter and consumption of meat or meat products in different parts of the world (Sans and Combris, 2015). According to Alao, *et al.* (2017), products of slaughter houses are separated into edible and inedible parts which serve different purposes. These edible parts (meat and offal) are consumed as foods to prevent malnutrition while the inedible parts (such as bones, hides and skin, feathers, hooves, horns, hair, bristles and rumen digest) are re-processed into other valuable products for industrial and biotechnological applications.

Keratin is a protein-rich fibrous and recalcitrant structural biomolecule which associate as intermediate filaments forming bulk cytoplasmic epithelia and structural epidermal appendages such as hair, wool, horns, hooves and nails (Rouse and Van Dyke, 2010). These wastes from animal processing are discarded yearly without proper re-use (Kornilowicz-Kowalska and Bohacz, 2011), they are good sources of protein (90%), with nitrogen (15–18%) and

sulphur (2-5%) contents respectively (Kunert, 2000; Onifade *et al.*, 1998). According to McKittrick *et al.* (2012), keratin exist as structural component of skin, hair, feather, horns, hooves, cloves, nails, beaks, reptilian osteoderm, fish slime and teeth, protecting the animals against biotic and abiotic stresses. Keratin is grouped as soft (contains 1 % sulphur) or hard (contains 5% sulphur and above) and the percentage of sulphur (present as cysteine molecules) determines their level of recalcitrance due to disulphide bonds between its polypeptide chains. These disulphide bonds form firm linkages between these polypeptide chains, responsible for its mechanical stability and resistance to biochemical degradation (Imtiaz and Rehman, 2018). The processing of these wastes reduce accumulation, cost of disposal and other environmental problems associated with these wastes, but improves their utilization in the sustainability of livestock production, making raw materials available for other purposes.

The disposal of these wastes was initially done by chemical treatments, incineration or in landfills (Paisley and Hostrup-Pedersen, 2005), but these steps are not environmentally friendly because they generate eco-destructive secondary pollutants and they are not cost effective (Ghaffar *et al.*, 2018; Wu *et al.*, 2017). The combustion of keratin-rich substrates releases volatile greenhouse gases such as nitrogen, sulphur and carbon containing oxides into the atmosphere as well as methane from anaerobic digestion of these wastes (Staronet *et al.*, 2011). According to Zhang *et al.* (2013), the disposal of these protein-rich wastes by incineration or in landfills also results in loss of essential amino acids (methionine, lysine and tryptophan), production of non-nutritive amino acids (lysinoalanine, lanthionine) and environmental pollution. Some landfills in later years generate a lot of non-volatile leachates which finds their way into water body systems and also make the soil unsuitable for planting of crops.

Although keratin-rich animal wastes are generated in million tonnes annually as the third most abundant macromolecule after cellulose and chitin, it does not accumulate in nature because of their degradation by some specific keratinolytic microorganisms but at a slower rate than chemical proteolysis. Keratinolytic microorganisms are widely distributed within the three domains of life namely bacteria, fungi and archaea (Intagun and Kanoksilapatham, 2017; Jayalakshmi *et al.*, 2012). Keratinolytic bacteria were

found amongst bacterial genus (*Bacillus*, *Clostridium*, *Stenotrophomonas*, *Streptomyces* and *Feravidobacterium*), Actinomycetes (*Streptomyces* sp. and *Thermoactinomyces*) and keratinolytic fungi (*Trichophyton*, *Absidia*, *Microsporum*, *Aspergillus*, *Onygena*, *Chrysosporium* and *Myceliophthora*) (Kornilowicz-Kowalska and Bohacz, 2011). Thermophilic and hyperthermophilic bacteria and archaea have also been reported to degrade keratin as well as prions, the etiologic agent of spongiform encephalopathies improving their digestibility and nutritive values (Intagun and Kanoksilapatham, 2017).

Keratin also serves homeostatic functions such as cellular integrity maintenance, protects cells from apoptosis, regulation of cell growth and migration by keratin-associated proteins and its post-translational modifications (Jacob *et al.*, 2018).

II. STRUCTURE OF KERATIN

According to Rouse and Van Dyke (2010), keratin is a general term used to refer to insoluble proteins associated with intermediate filaments and form cytoplasmic epithelial and epidermal appendageal structures such as nails, hair, horns wools and hooves (Meyers *et al.*, 2008). These structural proteins are grouped into hard and soft keratin depending on arrangement of the intermediate filaments (IFs) in the matrix of cysteine-rich proteins which contributes to their recalcitrant nature. The grouping into hard (feathers, horns, Hoofs etc.) and soft keratin (skin and callus structures) were determined by the percentage sulphur component of cysteine in the keratin (1% sulphur in soft keratin and 5% and above in hard keratins) as reported by Jinet *et al.* (2017). These sulphur residues form disulphide linkages between the filaments, with hydrogen bonds and other hydrophobic interactions provide recalcitrance to the protein (Abdel-Nabyet *et al.*, 2017; Lange *et al.*, 2016). The self-assembled intermediate filaments of keratin were also classified into α -keratin (made of α -helical-coils that are self-assembled into intermediate filaments as present in horns, hoofs, hairs and nails) and β -keratin (rich in β -pleated sheets constructed from supramolecular fibril bundles as present in bird beaks, scales, claws, feathers, reptile scales and wools) based on their secondary structures (McKittricket *et al.*, 2012; Boddeet *et al.*, 2011; Meyers *et al.*, 2008). Also, Yamada *et al.* (2002) reported that post-translational modifications of keratin (due to disulphide bond formation, phosphorylation and glycosylation) yield different modified filaments of keratin with different bio-accessibility. These modifications confer on keratin insolubility in water, organic solvents, weak acids and alkaline solutions as well as resistance to degradation by

other proteases (trypsin, papain and pepsin) besides keratinases.

Hard keratin-containing biomaterials have been found to contain both α - and β -keratins but at different proportions (Ng *et al.*, 2014; Dariot and Brandelli, 2014; ErRafiket *et al.*, 2004). The molecular arrangement of α -keratin involves two keratin polypeptides with head to tail dimeric helical structure (McKittricket *et al.*, 2012). The dimeric structure couples to self-assemble into 10 nm tetramers that make up the intermediate filaments of the biomolecule. The head domain of keratin contains glycosyl sugar moiety (N-acetyl glucosamine) and phosphoryl functional groups (Upreti *et al.*, 2003; Jacob *et al.*, 2018), and changes to these functional groups due to de-phosphorylation or de-glycosylation changes the charges on the keratin structure as well as disassemble the proteins structure (Herrmann *et al.*, 2004; Silengo *et al.*, 2003). Reports by Sprecher *et al.* (2001) showed that the keratin tail also contributes to its structural organisation. Mutation to the genes related to the tail domain caused a failure in the formation of these intermediate filaments which is a risk factor for some rare disorders such as cell fragility or altered homeostasis affecting internal or surface epithelia and hairs (Jacob *et al.*, 2018; Toivola *et al.*, 2015). But the formed intermediate filaments as suggested by Fraser and Mc Rae (1980) are embedded in a protein matrix with viscous-elastic properties containing high sulphur (cystenyl) or high glycine-tyrosine (glysyl) protein residues. High concentrations of glycine and tyrosine were reported by Tombolato *et al.* (2010) to be present in α -keratin which differs from β -keratin by the amount of disulphide bonds they contain and this determines the level of their rigidity and insolubility. They form corny tissues which differ in their biochemical properties and microarchitecture of intermediate filaments necessary for cell development and regulation of protein degradation (Sanghvi-Shah and Weber, 2017).

Reports by Bragulla and Homberger (2009) suggested that keratin and keratin filaments function in intracellular vesicle transport and cell signalling, transporting melanosomes from their endocytosis site at the cell periphery to the cells centre (Planko *et al.*, 2007). These filaments can easily assemble or disassemble to provide the cytoskeleton with flexibility, but their polymerisation is controlled by signalling molecules such as various kinases, heat shock proteins and phosphatases (Magain *et al.*, 2007). According to Oshima (2007), keratin and its filaments do not only function in cell signalling and transport but also in compartmentalization and differentiation. These characteristics of keratin and its filaments enables them to contribute effectively to wound healing and control of cell metabolic processes such as cell growth and protein synthesis as stated by Gu and Coulombe (2007).

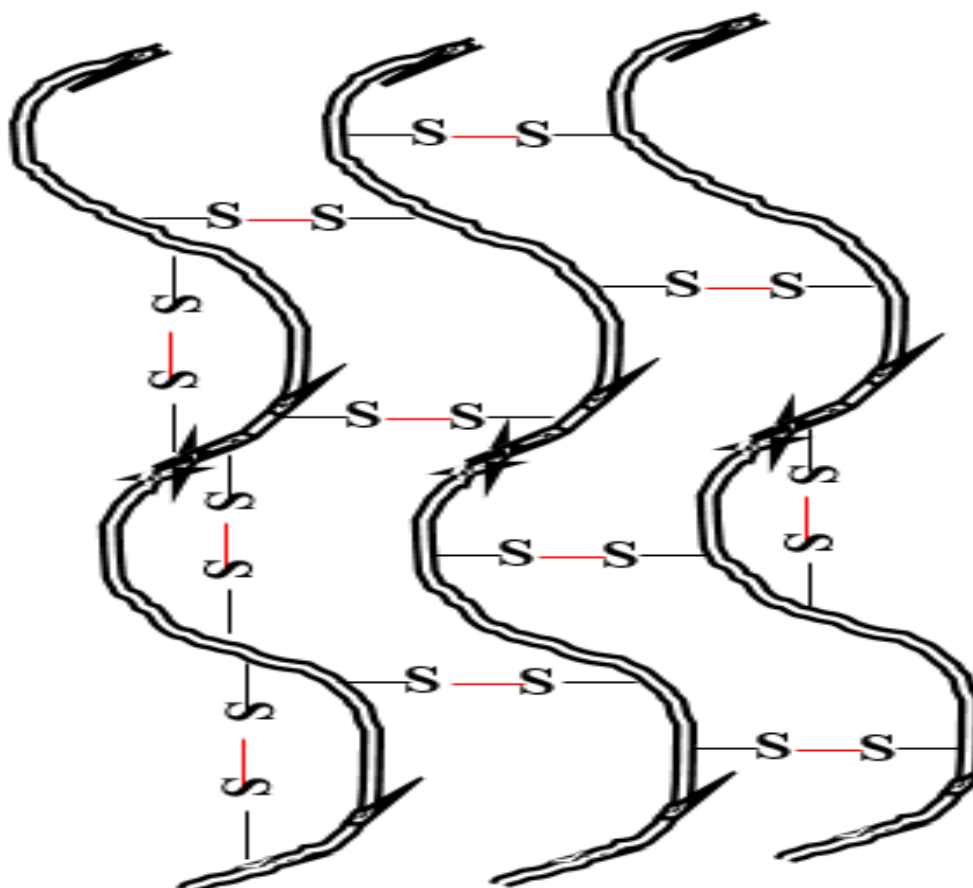


Fig. 1: Keratin structure showing the disulphide bonds which bonds sulphur in cysteine of the same or adjacent keratin strands to determine its recalcitrance

III. KERATIN DEGRADATION

Keratins can be chemically degraded using reducing or oxidizing agents like thioglycolate, hydrogen peroxide, dithiothreitol or mercaptoethanol which cleave the disulphide bonds (Navone and Speight, 2018), but traces of these chemicals persist in the hydrolysates which pollutes the environments. The use of oxidative and reductive chemistries to break down keratin was the earliest methods employed by researchers for this purpose (Rouse and Van Dyke, 2010). Alternatively, microbial digestion of keratin presents an important means for keratin recycling in the environment because of its eco-friendly approach compared to chemical proteolysis and incineration. Reports from literatures suggested that actinomycetes, bacteria and fungi produce keratinases which may sometimes be thermostable and can effectively degrade keratin-rich wastes (Nnolim and Nwodo, 2021; Onifade *et al.*, 1998). These wastes were initially disposed in landfills or by incineration, but the generation of secondary pollutants from these methods resulted to the need for alternative means of disposal. Also, the inactivation of heat-sensitive amino acids (tryptophan, methionine and lysine) into non-nutritive lysinoalanine and lanthionine have been reported by some authors (Bagewadi *et al.*, 2018; Zhang *et al.* 2013), and this reduces the nutritive values of keratin hydrolysates.

Eukaryotes were also reported to degrade keratin via ubiquitin proteasome pathway (UPP) by modifying the protein with ubiquitin molecules which is present in almost

all eukaryotic cells and can be recovered after the process. The four-step process as reported by Sahniet *al.* (2015) initiates by activating the ubiquitin to highly reactive ubiquitin thioester catalysed by ubiquitin activating protein (Pickart, 2001) which is then transferred to sulfhydryl group of the carrier protein containing cysteine molecule that will form thioester linkage with the activated ubiquitin. Furthermore, a ligase that acts as a scaffold and catalyst the transfer of activated ubiquitin to lysine residue in the target protein (keratin) and then to the lysine molecules in the ubiquitin to yield substrates anchored on ubiquitin molecule (Ciechanover and Schwartz, 1998). Finally, the 26S proteasome present in cytosol and nucleus of eukaryotic cells then catalyses the rapid degradation of the ubiquitin attached proteins (Roge *et al.*, 2010; Jaitovich *et al.*, 2008; Lecker *et al.*, 2006). However, Călin (2017) and Moreira *et al.* (2007) was of the opinion that fungal keratinolysis may be initiated by mechanical attack of the substrates by mycelial pressure or penetration which separates the substrates into simpler units (polypeptides) that will be further broken down by other proteases.

In bacteria and archaea, keratinases (a name given to collection of enzymes capable of keratin degradation) are employed and this group of enzymes are mostly produced in the presence of keratin containing substrates (Lange *et al.*, 2014; Bockle *et al.*, 1995). Keratinases are naturally proteases classified as proteinase with EC number 3.4.11.25 (Riffel and Brandelli, 2006), but easily grouped as serine

proteases (serine endopeptidases) or metalloproteases (Sahniet *al.*, 2015). Proteases have broad spectrum of activity and more effectively hydrolyses soluble proteins like casein than insoluble proteins like keratin (Suntornsuket *al.*, 2005; Syed *et al.*, 2009) and their biochemical properties differs according to the producing organism and thus display different activities on various substrates (Jayalakshmi *et al.* (2012). Some authors however, reported that purified keratinases are often ineffective in keratin hydrolysis due to high levels of disulphide bonds in keratin (Gupta and Ramnani 2006; Riffe *et al.*, 2007; Xie *et al.*, 2010). So, this hydrolysis process does not solely depend on protease production but release of thiol functional groups (Dariot *et al.*, 2009), intracellular or extracellular disulphide reductases production (Ghosh *et al.*, 2008), thiosulfate and sulphite release (Ramnani *et al.*, 2005) and the presence of cell bound redox system (Ramnani and Gupta, 2007) which aid sulfitolysis, the first step of keratin degradation process (Navone and Speight, 2018; Rahayu *et al.*, 2012). The sulfitolysis step is followed by proteolysis which involves breakdown of the separated keratin polypeptides into amino acid components by other protease complexes produced by the sekeratinolytic organisms.

The natural degradation of keratin has been shown to involve at least three enzymes (an endo-acting, an exo-

acting and an oligopeptide acting keratinase) working synergistically to degrade the substrate (Inada and Watanabe, 2013; Ramnani and Gupta, 2007). The breakup of disulphide bonds in keratin work hand in hand with keratinases to unwind its molecular structure and give the protease access to the substrate using extracellular endoproteases (S8), exoproteases (M28), oligopeptidases/metalloproteases (M3) and sulfite/disulfide reductases as reported by Lange *et al.* (2016). Laba and Rodziewicz (2014) also reported that none of the purified keratinase fractions have completely degraded keratin into bioavailable and bio-accessible amino acids and peptides, but complete keratinolysis usually follow complex pathways in a step-wise manner. This microbial keratin degradation was also suggested to be similar to microbial lignocellulose degradation (Li, 2019; Huang *et al.*, 2015; Yamamura *et al.*, 2002), where lytic polysaccharide monooxygenases LPMO protein complexes associated with polysaccharides decomposition are also present in the genome of keratinolytic organisms (Busk and Lange, 2016). Difference in the types of keratin (α - and β -keratins) also affects their rate of degradation. The β - keratin substrates are easily accessible to keratinase degradation because of their densely stacked structure and contains lesser amount of disulphide bonds (Gupta and Ramnani, 2006; Coulombe *et al.*, 2000).

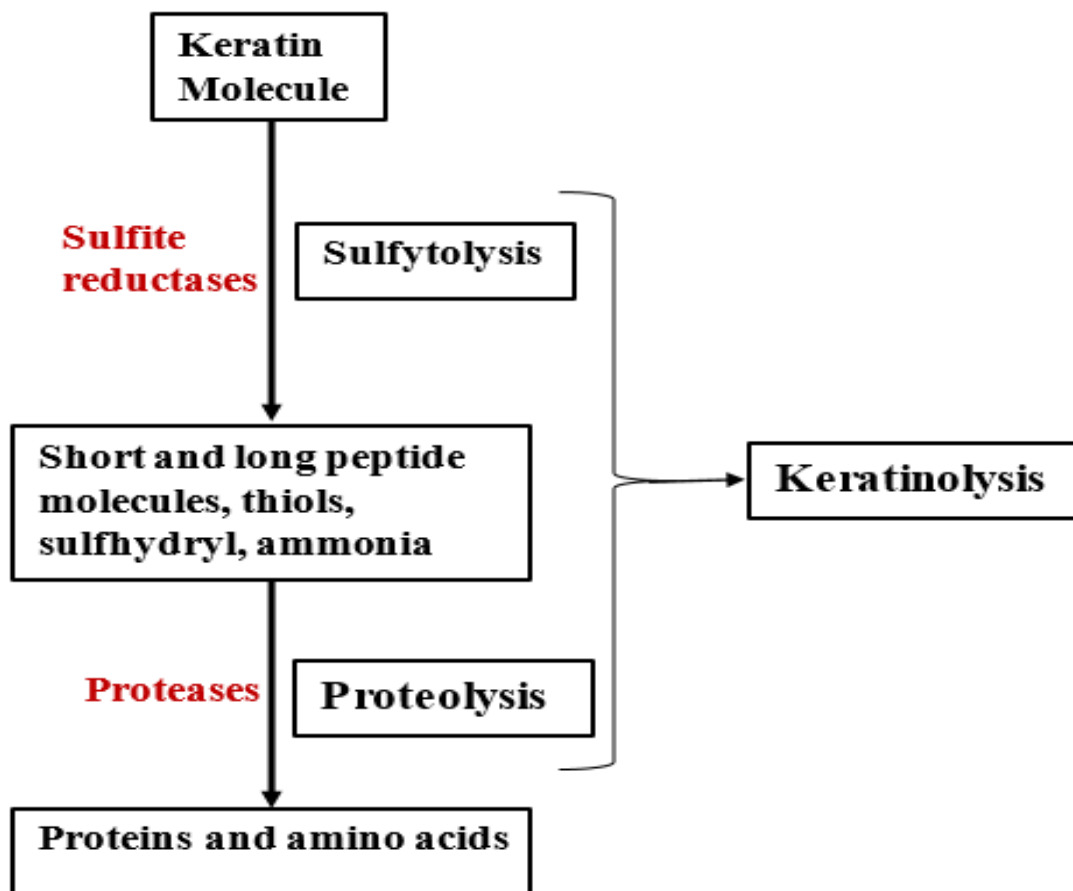


Fig. 2: Different stages involved in keratin degradation and their enzymes

IV. KERATIN AND KERATINASE APPLICATIONS

Keratin has been used for different purposes depending on the characteristics it displayed. It has inundated biological activity and biocompatibility like other naturally-derived biomolecules capable of self-assembled structures when degraded to regulate cellular recognition and behaviour. Choung and Homberger (2003) opined that cells and tissues are materials with distinctive physical properties subjected to regulated systems due to environmental interactions which make them adapt gradually to new conditions and uses. Protein-based materials have been of importance in the formation of extracellular scaffolds and matrix that mimics living cells and aid in various ex-situ biological interactions (Rouse and Van Dyke, 2010). Some of these qualities are possessed by keratin molecules which allow it to be used directly or indirectly to create other important materials such as biomaterials for wound healing, tissue engineering, medical devices, trauma and carriers for drug delivery. Human hair keratins as naturally-derived biomaterials possess good cellular interaction between some specific amino acid sequences within the keratin molecules and cellular integrins, and also show good biodegradability and biocompatibility properties when recovered under mild conditions. These characteristics enables them to elicit low immune reaction when used as implants in biomedical engineering applications such as nerve conduit filler for peripheral nerve regeneration, homeostatic agents, hydrogels or films for wound healing and other tissue regeneration procedures (Lee *et al.*, 2014). It has also been reported to be effective in pharmaceutical industries as carriers in drug delivery system (Tsfaye *et al.*, 2018). Because of human hair's keratin characteristic of spontaneous polymerisation and self-assembly, it has been applied in the development of various biomaterials such as films and porous scaffolds (Xing *et al.*, 2011; Lee *et al.*, 2014; Mohamed *et al.*, 2021). These keratin-based scaffolds expose a good number of functional groups in highly folded network of amino acids modified to improve their adsorbent ability have been used as bio-sorbents in waste water treatments (Saha *et al.*, 2019). Furthermore, Adelere and Lateef (2016) also suggested that keratinases can be used in the treatment of abattoir or slaughter house effluents to degrade keratin components and prions. Also, thermoplastics have been developed from keratin bio-composites and have its various applications in food packaging and biomedical procedures.

Horny or corny tissues of animals such as hooves and horns have some economic and emotional values namely use of mammalian fur for clothing, animal horns for drinking vessels, feathers for clothing and bed materials, skins for leather products, tortoise shells for combs and decorations, hooves and horns for slow decaying fertilizers and mammalian hairs for weaving and knitting yarns (Thys and Brandelli, 2006; Gupta and Ramnani, 2006). The economic importance of keratin-rich materials in cosmetics, dermatology and wool industry were also studied (ErRafiket *et al.*, 2004) and showed to be important in tissue repairs and wound healing. The enzyme keratinase has also been used as skin lightening agents in body creams and in topical treatment of hyperkeratosis.

As opined by Raju and Divakar (2013), processed poultry feathers can be used as fertilizers, animal feed, films glues as well as source of rare amino acids like cysteine, serine and proline. Reddy *et al.* (2017) on the other hand suggested that bioconversion of keratin-rich animal wastes yield hydrolysates rich with important amino acids such as methionine, lysine, tryptophan, threonine and isoleucine. These hydrolysates of keratin-rich substrates are used as carbon and nitrogen sources for microbial growth, animal feed supplements and in the amendment of sulphur deficient soils. The thermophilic anaerobic digestion of these animal wastes converts them into methane gas, and so these substrates serves as raw materials for biogas production as well as in hydrogen production using *Thermococcus litoralis* (Sahniet *et al.*, 2015). The thermophilic bioconversion of animal wastes also eliminates pathogenic organisms from the digestate which will be further used for soil amendment (Sugiyama *et al.*, 2005) or employed as composting material. This procedure can substitute for antibiotics use in feed stocks and possess many other environmental and industrial applications.

Feather efficiency and digestibility were improved as reported by Shih (1993) through application of keratinases to the substrate transforming it to valuable source of animal feedstock protein. Keratin induced enzyme, keratinases have shown the ability to degrade accumulated proteasome resistant prion proteins (PrP^{Sc}), the causative agent of transmissible spongiform encephalopathy (Kucziuset *et al.*, 2004; Langeveld *et al.*, 2003) and can be employed in the treatment of the disease. Prions are highly resistant to conventional methods of pathogens inactivation by chemical and physical methods and a small amount of these hard-to-degrade isoforms amyloid proteins pose serious health risk to humans and animals. Nagal and Jain (2010) also suggested that keratinases possess the ability to hydrolyse keratin-rich wastes into valuable products such as glues, films, feed stuffs fertilizers and amino acids. Their bioconversion ability on keratin-wastes made the enzyme a suitable agent for the bioremediation of keratin wastes based polluted environments (Imtiaz and Rehman, 2018; Jayalakshmi *et al.*, 2012).

Microbial keratinases have other industrial application like serving as detergent additives in the removal of stubborn stains of protein origin due to their proteolytic abilities (Reddy *et al.*, 2017; Paul *et al.*, 2013). In the treatment of appendageal infections of the toes and nails, the enzyme helps to digest the cornified cell layers for the drug to penetrate and attack the aetiological agents causing the infection. The delimiting process using sodium sulphide and bate during leather processing according to Sahni (2015) have been substituted with keratinase which yields hair of good quality. Srivastava *et al.* (2019) stated that microbial keratinases exhibit wide range of applications in food industry, modification of fibre, keratin-rich wastes bioremediation, drug delivery, biopolymers, detergents and leather industries. Furthermore, keratin based composite biopolymers produced by a combination of keratin and other polymers such as starch serve as raw material for hygiene products (diaper products and super absorbent materials), edible and non-edible food packaging materials, tissue

engineering materials (heart valve, artificial heart, hip and finger joints), fashion industry (breast implants and artificial lens), artificial skin replacement, pharmaceutical industries (drug delivery system components) and in wound dressing (Tesfayeet *et al.*, 2018). Further research on composites of keratin and other biopolymers with their applications will help alleviate the threat of keratin wastes accumulation and pollution of the environment.

V. RECENT ADVANCES AND PROSPECT IN KERATIN AND KERATINASE APPLICATION

Due to keratin's eco-friendly, non-abrasive, biodegradable nature consistent in quality, renewability, and biocompatibility, with lowest density value compared to other natural and synthetic fibres as well as sustainable resource, it has been used as a suitable material for high performance fibre reinforcement in polymer composites (Arshad *et al.*, 2016; Saha *et al.*, 2019). Keratin as organic compound with interesting properties, such as biocompatibility, can be possibly used as organic linker for the production metal organic frameworks (MOFs), which has numerous applications. Composite of chitosan and keratin nanoparticles have also been reported to serve as biomimetic scaffolds applied in bone tissue engineering and drug delivery systems (Nayak *et al.*, 2019; Râpă *et al.*, 2020).

Hybrid nano-biomaterials development by *in-situ* modifications of keratin resulted to green products with enhanced properties which can be used in nano-structured biomaterial composites, textiles and other bio-products (Khosa and Ullah, 2013). In order to improve the fibre properties, nano-engineering techniques through nanostructures incorporation have been used to improve their quality and applicability. This resulted to nanostructured materials prepared from nano-clays and POSS nano-cages feed stocks. These feed stocks are known to improve barrier properties, thermal stability, flame resistance, scratch resistance and UV transmissions in synthetic polymers if well dispersed in the matrix (Paul and Robeson, 2008). Barone *et al.* (2005) showed that the incorporation of 1-3 nm POSS molecules (which is eco-friendly, odourless and very thermally stable) into reinforced keratin fibre improved its moisture resistance, thermal stability, oxidation resistance, and mechanical properties, as well as reduces its flammability.

The isolation, purification and application of microbial keratinases induced by the presence of keratin-rich substrates for the valorisation of keratin-rich wastes, supplements for the improvement of animal feed nutritional value, degradation of infectious prions, removal of skin

corns and calluses, treatment of acne and nail fungal infections as well as incorporation into depilatory and skin peeling cream have been studied by different scientists (Lange *et al.*, 2016; Sharma *et al.*, 2018). It can also be applied in the cleaning and unblocking of drainage pipes and septic tanks for easy flow of sewage sludge (De Oliveira Martinez *et al.*, 2020). However, De Oliveira Martinez and colleagues further suggested the need for standardized experiment that will avoid pre- and co-treatments of substrate for accurate protease activity on keratin substrates and reduce experimental biases. The need for in-depth knowledge of the phylogenetic analysis of keratinases and their position within the phylogenetic trees of peptidases will further enhance the search for other peptidases with keratinolytic ability. Also, there is need for sequence analysis with the identification of specific domains and conserved sites on the genes that contribute to their keratin hydrolysing abilities they reported. Recombinant keratin of human hair origin have been produced and showed to be of high purity as well as used to accelerate healing of dermal wounds (Gao *et al.*, 2019; Konop *et al.*, 2021).

Keratin hydrolysis generates a lot of keratin-derived bioactive peptides that have been applied as antimicrobial (Sundaram *et al.*, 2015), antioxidant (Sundaram *et al.*, 2015; Zeng *et al.*, 2013), anti-inflammatory (Kelly *et al.*, 2009; Kelly *et al.*, 2007), antihypertensive agents (Ohba *et al.*, 2003), early stage amyloid aggregation inhibitors (Jones *et al.*, 2010), anti-aging (Yeo *et al.*, 2018; Jin *et al.*, 2018; Jin *et al.*, 2019) as well as antidiabetic (Fontoura *et al.*, 2014) agents depending on preparation method and source of the keratin (De Oliveira Martinez *et al.*, 2020). Keratin hydrolysates have been efficiently applied for different purposes and they yielded good results in their applications.

Further research on keratin/starch composites by Tesfayeet *et al.* (2018) produced innovative, transparent and biodegradable plastic films that can be used in food packaging industries as an alternative to petroleum based plastic materials. Starch produced from avocado seeds were processed and blended with keratin extract from chicken feathers to produce the film. They also suggested that the film can be used in the manufacture of hygiene products such as super adsorbent diaper products and manufacture of artificial skin products and drug delivery systems. In another work, Sanchez-Ramirez *et al.* (2017) produced a transparent citric acid-containing plastic film from wool hydrolysate with biocidal activity for food packaging. Its biocidal activity helps to improve the foods shelf life by reduction or inhibition of microbial growth within the environment.

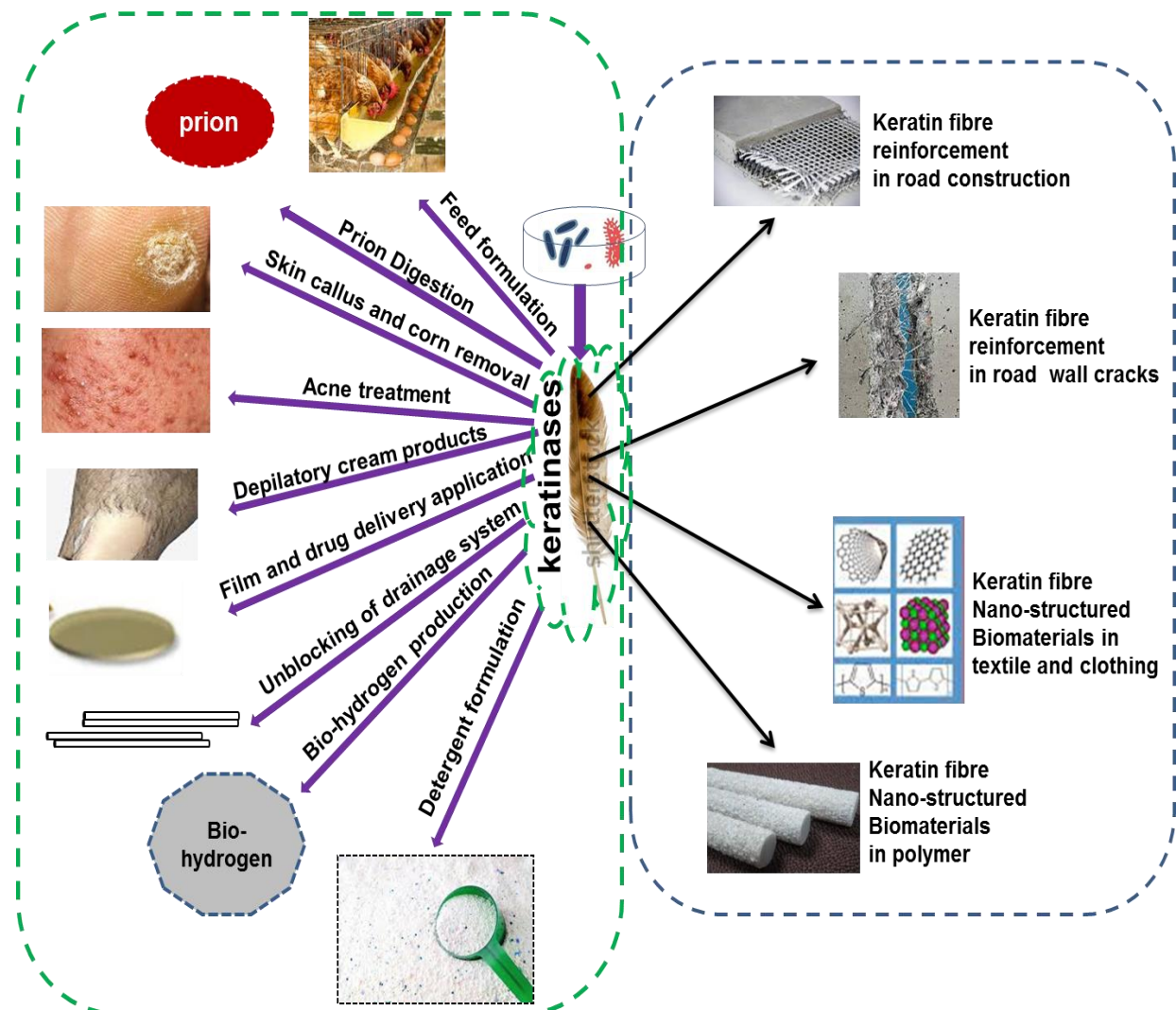


Fig. 3: Recent advances and prospect of keratin and keratinases applications

VI. CONCLUSION

Keratin is a complex and recalcitrant protein with numerous applications. Effective management of keratin-rich wastes provides several bioactive molecules that can be applied in different biotechnological and industrial processes. But their mismanagement results to its accumulation leading to serious environmental deterioration. Leachates from its accumulation and landfills pollute underground waters and make them unfit for other needed usages. It is therefore important to utilize these complex wastes in a way that will improve lives and harness its other numerous potentials biotechnological potentials. Microbial valorisation of these recalcitrant wastes proved to be an eco-friendly methods for their improved digestibility and applicability in various fields. This process will not only reduce environmental pollution but provide an important source of active biomolecules used to improve their structural stability.

• **Conflict of Interest:** The authors declare no conflict of interest in carrying out this research.

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