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Evidence on the biomodulatory potential of oral collagen peptide supplementation for skin aging: composition, mechanism of action, bioavailability, eficacy and toxicity

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ABSTRACT

Dietary and nutraceutical supplements based on hydrolyzed collagen are capable of modulating the biomechanical and functional properties of the skin tissue. Hydrolyzed collagen (CH) is a group of low molecular weight (3-6 KDa) peptides that can be obtained through the enzymatic hydrolysis of native collagen derived from animal (commonly bovine, porcine or marine) or vegetable sources. The stimulation of the synthesis of structural elements of the dermis has been proposed as one of the main factors associated with its promising biological effects however, its cellular and molecular mechanisms are still unknown in the literature. Thus, this literature review summarizes the most relevant clinical and experimental scientific studies on the composition, action mechanisms, absorption, bioavailability, effectiveness, toxicity and its therapeutic potential response on dermal aging.

Keywords: hydrolyzed collagen, bioactive collagen peptides, gly-pro-hyp, pro-hyp, oral nutraceuticals, skin aging.

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INTRODUCTION

Collagen is one of the most abundant structural proteins, representing about 20-30% of the total isolated proteins in mammals[1]. Furthermore, it is present in different connective tissues, including skin, cartilage, bones, tendons and ligaments[2].

Collagen is a large fibrous protein formed by a complex quaternary structure of triple helix conformation. It consists of peptide chains rich in amino acids glycine, proline, lysine, hydroxylysine, hydroxyproline and alanine[3]. In epithelial tissue, this protein participates in the constitution of the extracellular matrix along with hyaluronic acid and other matrix fibers, for example, reticulin and elastin. In addition, it forms a support network for cells such as fibroblasts, keratinocytes, melanocytes and specialized cells of the cutaneous immune system[4]. It is also responsible for several functions including maintaining hydration, tissue regeneration and healing, cellular stabilization (regular cell organization), mechanical resistance, strength and elasticity properties[5–9].

During skin aging, the cellular metabolism of fibroblasts becomes less active and collagen production also decreases. From the beginning of adulthood, between 18-29 years old, the human body begins to lose collagen.By the age of 40, is estimated that 1-1.5% of collagen is lost per year. And by age of 80, this reduction in overall collagen biosynthesis can reach up to 75% compared to young-adult individuals[6,8].

This gradual decrease in collagen production is accompanied by fragmentation and disorganization of collagen and elastin fibers, loss of water-holding capacity, alteration of the epidermal barrier and mechanical fragility. These structural and functional chronological changes result in more aged-looking skin[3].

Over the last few years, there has been a growing number of scientific studies that seek to develop a variety of anti-aging therapies, including the use of oral supplements. Scientific evidence reports that several food supplements and nutraceuticals have the ability to modulate cellular and skin metabolism, being potentially effective in smoothing and delaying the signs of skin aging[10,11].

Among these nutraceutical compounds is hydrolyzed collagen (CH) that have been described for its biological potential benefits on human skin physiology[11].

Hydrolyzed collagen (CH) is a combination of low molecular weight, water-soluble, bioactive peptides that is able to be directly absorbed into the blood through digestion, becoming more bioavailable than whole collagen proteins. These peptides offers faster absorption, better digestibility, bioavailability and nutritional functionality[12].

According to the Food and Drug Administration (FDA) and the Brazilian Health Regulatory Agency (ANVISA), collagen peptides are classified as GRAS (generally recognized as safe), wherefore, they are safe products with low allergenic risks. And because they are only made of protein, there are no described contraindications related to their use[13].

In Brazil, CH is approved by Anvisa in the food/new ingredient category. In this case, a collagen food supplement as a source of protein must meet the minimum limit, according to the daily recommendation of minimum consumption

established for the indicated population group. For adults, recommendation is 8.4g daily comsumption of the referred product. However, this amount does not need to be met exclusively by collagen, so the resolution allows the manufacturer to combine different protein sources to meet the established minimum limit[11,14].

Another normative act classifies type II collagen food supplements as a bioactive substance, with the purpose of helping to maintain joint function. In this case, it is established that the minimum amount of total collagen is specifically 10mg and for undenatured type II collagen is 1.2mg[14].

In this way, the reference values for the labeling of products containing collagen may vary according to the manufacturer[14]. In addition, as it is considered a food supplement, this product does not require therapeutic bioavailability tests; these tests would be responsible for ensuring the quality standard of the hydrolyzed collagen dosage forms[15,16].

The studies have shown that oral ingestion of hydrolyzed collagen on nutraceuticals leads to a transcient increase in the levels of bioactive peptides in the blood. But whether these peptides reach the skin, whether they are capable of depositing in tissue or even how they exert their biological activity, still parameters that are not yet consensually elucidated by the scientific community[17].

Some important particularities that need further studies are the clinical efficacy of CH and wich patients the administration is necessary.

We will present the main scientific evidence of collagen-based oral supplementation about its absorption, bioavailability, action mechanisms and its clinical effectiveness related to skin aging.

MATERIALS AND METHODS

The objective of this study was to demonstrate the main scientific evidence on oral collagen supplementation including its nutraceutical forms, composition, action mechanisms, absorption, bioavailability, effectiveness, toxicity and its therapeutic potential response in modulating skin aging. This is a literature review based on clinical and experimental scientific research. It was consulted through electronic databases including PubMed, MEDLINE/Bireme, ScienceDirect, SciELO, LILACS, ClinicalTrials.gov and Portal Periódicos Capes. Articles published in Portuguese, English and Spanish from 2015 to 2021 were searched, and consulted using the descriptors: “hydrolyzed collagen”, “bioactive collagen peptides”, “Gly-Pro-Hyp”, “Pro-Hyp”, “oral nutraceuticals” and “skin aging”.

In this study, we explore the most controversial aspects in dermatology related to oral collagen intake and the most recent published evidence on the subject.

Inclusion criteria were original articles or bibliographic and/or systematic review, case reports, theses, conference proceedings and scientific books that evaluated collagen administration mainly in dermatological effects, which include tests with experimental models (*in vitro* and *in vivo*) and clinical trials of healthy subjects of any age group.

Initially, a total of 103 publications were identified, but after removing duplicates and screening the articles abstracts, only 26 were eligible for the inclusion criteria in this literature review.

Publications that were not available in full, works published before the year 2015, duplicate articles and subjects that were not related to the inclusion criteria were excluded.

RESULTS

Collagen structure

Collagen has a triple helix structure consisting of three polypeptide alpha chains (Figure 1)[12]. This collagen structure is composed of approximately 1400 amino acids and can have a diameter of 10 to 500nm and an approximate molecular weight of 300kDa[18,19].

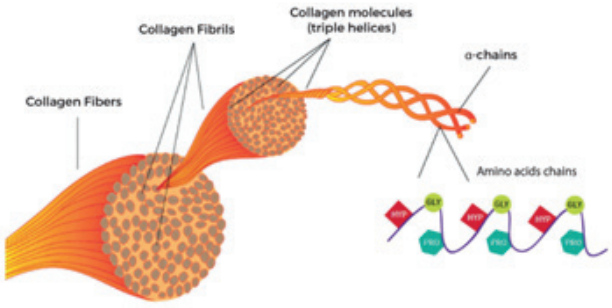

Figure 1. Molecular structure of native collagen and low molecular weight bioactive peptides (Adapted from Reilly & Lozano, 2021).

Each polypeptide alpha chain is composed of a repeated amino acid sequence (Gly-X-Y)n, in which the X position is usually occupied by the amino acid proline (Pro), while the Y position is typically occupied by hydroxyproline (Hyp) (Figure 2)12,20. These three polypeptide chains are arranged parallel to an axis and joined by hydrogen bonds, thus forming collagen fibers (Figure 1)[1,4]. This protein arrangement is responsible for the physical and biological properties of collagens, such as: rigidity, solidity and stability[4].

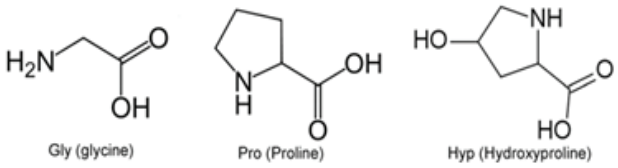

Figure 2. Molecular formula of the main amino acids in collagen structure (Adapted from Knez et al., 2019).

Collagen triple helix stabilization is also influenced by small chemical interactions, including the Van der Waals, hydrophobic, and electrostatic interactions[1].

Collagen can be described as a family of approximately 29 distinct isoforms of proteins found in connective tissues, with collagen types I, II and III being the most abundant in the body[1,10]. Type I collagen is found primarily in the skin, tendon and bone; type II, in cartilage and vitreous humor (found in the eyes); and type III, in skin and muscles[10].

Collagen sources

Collagen is obtained from animal and plant sources, originating from bovine, poultry, porcine, human and marine organisms (fish and other invertebrates). But the main sources of commercial collagen currently used are animal by-products, such as skin and bone from pigs and cattle. This preference is due to its similarity with the human protein[4] and for being a raw material easily obtainable from the food industry[19]. On the other hand, the extraction of hydrolyzed collagen obtained from these traditional sources has limitations due to imminent health problems such as swine flu and bovine spongiform encephalopathy[6].

Marine collagen sources are similar to those from bovine and porcine origin

in terms of biocompatibility and amino acid content, but they have advantages when compared to these sources, such as their greater absorption capacity (low molecular weight), negligible biological contaminants (reduction of toxins and few inflammatory effects) and more affordable commercial extraction[18,22].
There are also synthetic alternative sources of collagen peptides commercially known as "KOD" or collagen mimetic peptides. They are recombinant collagens that replicate multihierarchical portions of natural collagen and act to reduce immune reactions. They are produced from mammals, insects, yeast and plant cell cultures[18].

Nutraceutical forms

Non-hydrolyzed or native collagen

Native (tropocollagen) or non-hydrolyzed collagen is composed of large triple helical structures of long, insoluble amino acid chains - due to the presence of large amounts of hydrophobic amino acids[17,23,24]. In its native form, collagen has low absorption by the body due to its large polypeptide chain and high molecular weight (300-400kDa)[19,23].

Partially hydrolyzed collagen (gelatin)

Gelatin is a soluble mixture of polypeptides obtained through the partial hydrolysis of collagen (16-150kDa)[25,26]. From type I collagen it is possible to obtain partially hydrolyzed collagen (gelatin) and hydrolyzed collagen (HC)[27]. However, its gelatin structure differs from native collagen due to its ability to form a gel by trapping water between the polypeptide chains[25,27].
And just like the oral administration of native collagen, partially hydrolyzed collagen in the form of gelatin is also not efficiently absorbed by the human body[4].

Hydrolyzed collagen

Hydrolyzed Collagen (CH) is a safe food and nutraceutical supplement that consists of a mixture of small, low molecular weight peptides (3-6kDa) generated by the total hydrolysis of native collagen on the animals skin (usually bovine, porcine or fish)[6,11]. The process of obtaining CH is complex and has multiple steps, the main ones being: (1) thermal degradation, (2) chemical degradation (acid or alkaline) and (3) enzymatic hydrolysis (Figure 3)[11].

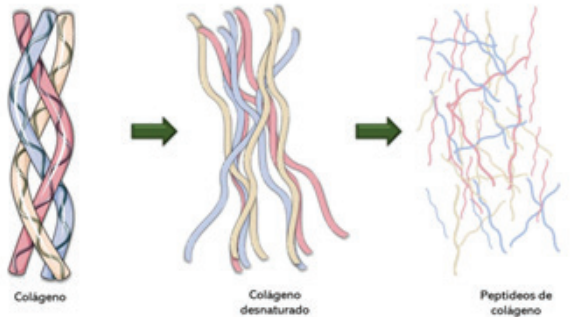

Figure 3. Process of denaturing native collagen into low-weight bioactive molecular peptides (Adapted from da Costa et al., 2020).

CH is the product of the enzymatic hydrolysis of collagen in order to obtain biologically active, easily digestible and highly bioavailable collagen peptide fragments for the human body[3,23].
It is important to emphasize that depending on the collagen source and the enzymatic hydrolysis technology used in the process, it can generate combinations of peptides with different biological activities. Alcalase, for example, is a non-specific enzyme capable of cleaving the native collagen molecule random-

ly generating different peptides. While specific enzymes and sequenced enzymatic hydrolysis cleave specific sites on the collagen molecule resulting in specific bioactives[11].

Regarding its biological activity, some authors report that CH and its derivatives have antioxidant, antihypertensive and metabolism-stimulating activities in different connective tissues such as cartilage, bones and the skin connective tissue[11].

The latest scientific evidence indicates that hydrolyzed collagen is absorbed both in the form of free amino acids (building nutrients) and in the form of collagen peptides (biologically active)[11]. After digestion, they are metabolized into oligopeptides (di- and tri-peptides) in the gastrointestinal tract and then distributed and accumulated in different tissues, such as skin and cartilage, forming new local collagen fibers[4].

Nutraceuticals based on hydrolyzed collagen are rich in amino acids such as hydroxyproline, proline and glycine[4]. And numerous studies demonstrate that the dipeptides prolyl-hydroxyproline (Pro-Hyp) and hydroxyprolyl-glycine (Hyp-Gly) are directly absorbed in this form and deposited in skin tissue. These peptides promote the chemotaxis of dermal fibroblasts and increase the proliferation and bioactivity of these cells. It is also suggested that Pro-Hyp stimulates the synthesis of hyaluronic acid (HA) and, therefore, also acts to improve the hydration of the skin tissue[4].

Biological properties of hydrolyzed collagen (CH)

Several beneficial effects on the oral administration of hydrolyzed collagen have been reported in recent years. These include improvements in acti- vity-related joint pain, tendinopathies, chronic joint instability, osteoarthritis (OA), improved wound healing, antihypertensive function, and inhibitory activity of cardiovascular damage to endothelial cells (ACE inhibitory action and regulation of nitric oxide and ICAM-1), increased glucose tolerance (inhibits intestinal glucose uptake partially related to GLP-1 and increases insulin secretion), modulation of biomechanical and functional properties of the skin such as moisture (hyaluronic acid synthesis), elasticity (dermal fibroblast chemotaxis and cell proliferation), epidermal barrier function (reduces transepidermal water loss (TEWL), restoration of UVB-induced skin quality (increases dermal hyaluronic acid content and prevents water loss on the stratum corneum), as well as a improvement on facial wrinkles (biosynthesis of procollagen I, elastin and fibrillin)[11,20,28].

And many studies suggest a wide distribution of oral collagen among several tissues including liver, brain, kidney, cartilage, muscles, skin, etc.[11,29]. Cartilage and skin are important retention tissues for collagen bioactives[11].

Although these cellular and molecular mechanisms are not sufficiently described in the literature, it is believed that Pro-Hyp is probably a key collagen-derived peptide that plays its biological properties in human organism[20].

Action mechanism

The action mechanism of hydrolyzed collagen still not fully elucidated. But it is known that CH and its bioavailable forms act basically in 2 different ways:

(1) collagen precursors – the collagen hydrolysis product is used by fibroblasts as collagen synthesis precursors; (2) fibroblast stimulators via receptor-mediated activation pathways – the unique oligopeptide sequences (especially hydroxyproline-containing dipeptides) induce fibroblast receptor-mediated activation pathways to stimulate the synthesis of new collagen. Therefore, although the absolute concentration of bioactive peptides is low compared to amino acids, these peptides are capable of producing very effective biological responses even on low levels[8].

Oral supplementation and collagen bioavailability and its derived peptides

The bioavailability of hydrolyzed collagen obtained through oral supplementation is considered a limiting factor for the effectiveness of the nutraceutical in different biological tissues[11]. This bioavailability can be defined as the relative amount of bioactive compounds obtained in the diet, which crosses the gastrointestinal barrier (absorption), reaches the bloodstream (distribution) and becomes available to participate in metabolic processes or for body storage[4]. Thus, research on the oral bioavailability of collagen peptides to improve skin structure and quality has been described in animal and human models[29,30].

The mechanisms of protein and peptide absorption are relatively well elucidated. These nutraceuticals are mainly hydrolyzed by extracellular and intracellular proteases and peptidases into a mixture of free amino acids and small oligopeptides in the gastrointestinal tract[29].

The gastrointestinal tract consists of a thin layer of epithelial cells that are joined by tight junction complexes capable of forming a selective and polarized barrier. This structure allows the formation of two membrane surfaces with different functions; an upper surface facing the lumen and another basolateral surface facing the circulation. These two compartments are also responsible for protecting the body against harmful xenobiotic agents[29].

Peptide absorption is facilitated by the large membrane surface area formed by the folding of intestinal villi and microvilli. In general, permeation across the intestinal membrane occurs by two types of transport including paracellular and transcellular[29].

Paracellular transport is a passive pathway controlled by tight junction resistance, in which net permeation is factors dependent such as the porosity of the junction itself, the size and charge of peptides and amino acids. Transcellular transport involves different mechanisms such as passive transcellular diffusion, transport-mediated pathway and transcytosis transport[29].

Most amino acids are absorbed by specialized Na^{+-} dependent and Na^{+-} independent transporters, whereas peptides are mediated particularly by proton-coupled oligopeptide (POT) transporters[29].

Once absorbed, the peptides are used as precursors for essential proteins synthesis in the human body. Furthermore, upon entering the circulatory system, peptides may undergo further degradation through enzymatic cleavage by plasma peptidases and esterases during their distribution[29].

It is estimated that the final intestinal absorption is approximately 10%, which

may vary according to the source of CH and the experimental model that was used[29].

Thus, although hydrolyzed collagen is easily digested and absorbed, there are still important factors that can alter its oral bioavailability[29].

Bioavailability studies of hydrolyzed collagen (CH)

Previous evidence of the bioavailability of hydrolyzate collagen in a dynamic simulator of the human digestive system (TIM model) reported that the absorption of CH is 82% after 6h of the oral administration. And later, when evaluated by SDS electrophoresis and HPLC, the results report that in vivo CH absorption increases to 95% after 12h of the oral administration, remaining relatively high for upon 96 h. Furthermore, it was demonstrated that CH peptides of molecular weight between 1 to 6 kDa were able to transit from the mucosa to the intestinal serosa, which suggests the ability of these peptides to be absorbed in their partially intact form[11].

In a later study by Watanabe-Kamiyama et al. (2010) the biodistribution of collagen bioactive peptides when administered orally in rats was investigated. Previously radiolabeled collagen peptides and amino acids revealed that the hydrolyzed collagen was partially absorbed in its peptide form. And another observed result was that the radioactivity in the plasma peaked 3 hours after oral administration, and the maximum value in the skin occurred during the following 3 hours. And even after 14 days of oral administration, it was possible to find 70% of the maximum radioactivity in the skin, suggesting a long-term accumulation of hydrolyzed collagen in the skin tissue[4,11].

According to the later experimental study published by Wang et al (2015) the relative and absolute bioavailability of orally administered collagen in rats was 57.8% and 49.6%, respectively. These data suggest that about half of the collagen administered to rats was reaching the systemic circulation to perform its biological activity. This fact is possibly due to the first-pass effect in which the liver promotes the metabolism of foreign substances before they reach the systemic circulation[31,32].

But when referring to more recent clinical evidence, we can name the publication by Skov et al. (2019) that investigated the postprandial absorption of collagen and elucidated the impact of the enzymatic hydrolysis process on the absorption rate and collagen bioavailability. It was a crossover, randomized, blinded, controlled study conducted in 10 healthy male subjects with oral administration of collagen supplement of 35g of Crude Protein Enzymatically Hydrolyzed Collagen (EHC), 35g of Non-Enzymatically Hydrolyzed Collagen (NC) or placebo (250 mL of water) on three non-consecutive experimental days. The results reported that both EHC and NC demonstrated a significant increase in the plasma concentration of almost all amino acids (AAs) over a period of 4 hours (240 minutes). It also reported that the enzymatic collagen hydrolysis significantly increased the absorption rate and bioavailability of the AAs glycine, proline and hydroxyproline[28].

Experimental studies of the action mechanism and the CH effectiveness on the skin

Initially, an in vitro assay conducted by Postlethwaite et al. (1978) quantified the collagen chemotactic activity and its collagen-derived peptides in human dermal fibroblast cell culture. The results indicated that all three native human collagens (types I, II and III) had a chemotactic effect for fibroblasts. He even demonstrated that peptides derived from the digestion of collagen types I, II and III through collagenases and synthetic di-and tripeptides containing hydroxyproline also had chemotactic effects on fibroblasts. These results suggest that the degradation of native collagen generates bioactive peptides capable of stimulating the chemotactic activity of fibroblasts in vivo and attracting these cells to repair tissue damage[11].

Other studies conducted with fibroblast cultures demonstrate that the presence of collagen induces these cells to synthesize more collagen and other tissue components such as proteoglycans and hyaluronic acid. Further evidences report that this effect is dose-dependent, and does not occur with non-hydrolyzed collagen or even with collagen peptides[30].

In the work of Liang et al. (2010), they evidenced the long-term beneficial effects that oral ingestion of CH promoted on the chronological animals aging. The results revealed the potential of CH to: (1) inhibit collagen loss and fragmentation; (2) increase collagen I and III protein expression levels and procollagen I and III gene expression through the Smad/TGFβ signaling pathway; (4) inhibit collagen degradation by attenuating MMP-1 expression and increasing MMP-1 inhibitor (TIMP-1) expression and (5) reducing oxidative stress in aged skin[11].

A study conducted by Ohara et al. (2010) in human fibroblast culture showed that collagen peptides such as proline-hydroxyproline (Pro-Hyp) were able to stimulate cell proliferation (1.5 times) and the synthesis of hyaluronic acid (3.8 times). In addition, Pro-Hyp stimulated cellular mitotic activity and hyaluronic acid synthesis mediated by transcriptional activation of the hyaluronate synthase 2 (HAS2) gene. These data indicate the positive effects of collagen-derived peptides on the synthesis of dermal extracellular matrix components and on fibroblast cell proliferation[11].

The most recent scientific evidence in experimental models published by Lapi et al. (2021) reports that mice fed a 5% diet supplemented with supplements enriched with marine collagen exhibited an increase in the expression levels of chemokines in the wound region, an increase in expression of vascular endothelial growth factor-A (VEGF-A) and increase in collagen deposition. Thus, promoting tissue repair in the experimental model of skin wound healing through chemokine induction[33].

Clinical studies on the effectiveness of CH for the skin

When referring to its clinical efficacy, two double-blind, randomized, placebo-controlled clinical trials conducted by Proksch et al. (2014) can be reported. The first one was performed in 69 women aged 35-55 years, administering 2.5g or 5.0g of specific collagen peptides or placebo, once a day for 8 weeks. The results showed that women

in both groups who received specific CH showed significant improvement in skin elasticity and that these effects were maintained even after 4 weeks of stopping treatment. These positive effects were even more pronounced in women over 50 years old, in which they also revealed the positive influence of CH on hydration and transepidermal water loss in these same subgroups. In the following clinical study, 2.5g of specific collagen bioactive peptide or placebo was administered once daily for 8 weeks to 114 women aged 45-65 years. The data reported that after 8 weeks of treatment with CH there was a reduction of more than 30% in the volume of wrinkles and an increase of around 60% in the content of procollagen I. Furthermore, these positive effects were also long-lasting, remaining even 4 weeks after the end of the treatment[11].

In a study proposed by Asserin et al. (2015), the authors report that women between 40 and 59 years old who received oral CH supplementation demonstrated a significant increase in skin hydration and dermal collagen density. In addition, there was a significant reduction in dermal collagen fragmentation after 4 weeks of treatment. And these effects were maintained after 12 weeks[6].

In the same year, the publication by Schunk et al. (2015) demonstrated that women with oral CH supplementation for 8 weeks had an improvement in skin elasticity and a decrease in transepidermal water loss. And these positive effects were still maintained for another 4 weeks[4].

In another study, Alvim (2015) proposed the oral administration of a nutritional supplement (collagen peptides, vitamin C and Hibiscus sabdariffa) for 90 days in female patients between 35 and 65 years old. The results showed a significant increase in dermal thickness. In addition, it revealed a positive perception in a subjective questionnaire about the improvement of the patients skin firmness, elasticity and hydration[4].

The following year, the publication by Inoue et al. (2016) evaluated the effects generated by oral supplementation of dipeptides (Pro-Hyp and Hyp-Gly) on the skin. The analysis showed that hydrolyzed collagens with greater amounts of bioactive peptides had superior effects on the skin. In addition, it showed that the higher concentration of Pro-Hyp and Hyp-Gly in collagen promoted an improvement in elasticity, moisture and facial wrinkles[4].

These beneficial effects were confirmed in the publication by Kim et al. (2018) which demonstrated the significant improvement in hydration, wrinkles and skin elasticity in women between 40 and 60 years old who received oral CH supplementation for 12 weeks[6].

Addor et al (2018) evaluated the association between collagen peptides, vitamin C and extracts of Hibiscus sabdariffa and Aristotelia chilensis in women aged 35 to 65 years. Data demonstrated an increase in dermal thickness, skin firmness and elasticity after three months of treatment[6].

These previously reported clinical effects can be corroborated through the study by Czajka et al. (2018). In which, they evaluated the influence of oral marine collagen supplementation in 120 patients for 90 days. The findings demonstrate that in addition to the im-

provement in skin texture and elasticity, there was also a protective effect on the joint health of these patients[6].

In a study published by Campos et al. (2019), the clinical efficacy of a dermocosmetic formulation containing di-and tripeptides was compared to oral supplementation based on hydrolyzed collagen combined with vitamins A, C, E and zinc. The result showed that the topical formulation increases significantly the water content of the stratum corneum and the skin elasticity after a period of 28 days and acts on the echogenicity of the dermis after 90 days, while the oral supplementation acts on the skin elasticity and has effects more pronounced in dermis echogenicity (reducing skin pores) within the same 90-day period. These findings reveal that oral supplementation and topical application of hydrolyzed proteins act in a complementary way on the skin tissue[6].

Further scientific evidence was published by Bolke et al (2019), in which they demonstrated that supplementation of collagen peptides combined with fruit extract, Vitamin C, E, zinc and biotin administered to 72 healthy women, with 35 years old, during 12 weeks. This protocol was able to improve the hydration, elasticity, softness and skin density of the treated group[6].

Mödinger et al. (2021), in a recently published study, confirmed some of these effects of hydrolyzed collagen on the skin that were previously described in the literature. In this work, the authors reported that oral collagen peptide-based nutraceutical supplementation (2.5 g of collagen peptides and selected micronutrients) in healthy women aged 40-65 years for 12 weeks was able to reduce facial wrinkles. More pronounced effects were reported especially in elderly women, as well as a slight improvement in skin hydration in middle-aged women[34].

DISCUSSION

Possibly the most important aspect associated with oral collagen supplementation is its clinical efficacy on skin aging. This is mainly due to the existence of a limited number of quality studies that can robustly demonstrate the effects of collagen for this purpose. Even then, these few studies have revealed the potential effects of hydrolyzed collagen (CH) and its derivatives in terms of their ability to modulate cell functions and human skin metabolism. Among these effects are included increased skin hydration, antioxidant properties, enhanced photoprotection, increased synthesis of extracellular matrix components, decreased transepidermal water loss, improved wound healing, increased dermal collagen synthesis, smoothed wrinkles and improvement of skin texture and elasticity. Regarding the action mechanism, scientific evidence reports that hydrolyzed collagen and its derived peptides act basically in two different ways: (1) they provide amino acids as building nutrients for collagen synthesis and (2) the peptides act as regulators of cellular collagen synthesis activity through fibroblast receptor-mediated activation pathways. Thus, hydrolyzed collagen is responsible for modulating the chemotaxis and cell proliferation of fibroblasts and stimulating the synthesis of matrix components such as proteoglycans and hyaluronic acid.

More recent evidence suggests new action mechanisms such as inhibition of

collagen loss and fragmentation through overexpression of the metalloproteinase inhibitor (MMP-1), induction of procollagen I and III gene expression through the Smad/TGFβ signaling pathway, stimulation of hyaluronic acid synthesis mediated by the activation of hyaluronate synthase 2 (HAS2) gene transcription and attenuation of oxidative stress in skin tissue.

Regarding the bioavailability, hydrolyzed collagen demonstrates fast gastrointestinal absorption, passing into the bloodstream partially in its intact form and the rest in its oligopeptide form. And they report a bioaccumulation of these peptides in the skin for approximately 96 hours after ingestion, thus revealing their long-lasting effects. In particular, specific peptides such as prolyl-hydroxyproline (Pro-Hyp) and hydroxyprolyl-glycine (Hyp-Gly) exhibited better effects on dermal cell metabolism and proliferation. Concerning to the biological activities of peptides derived from hydrolyzed collagen, their relationship to the structure and molecular weight of these peptides becomes evident, which in turn are strongly affected by the processing conditions of these supplements.

It was shown that not all collagen hydrolysates are the same, as the composition of collagen hydrolysates or peptides may vary according to the enzymatic hydrolysis process used to obtain them (enzymatic cleavage at specific or random sites). This ensures the manifestation of dissenting clinical effects. It was also emphasized that the exemption of bioavailability tests for food supplements allows a variation of the reference values for labeling of the products containing collagen according to the manufacturer. Studies indeed demonstrate that oral collagen supplementation is generally safe, with no associated adverse events. Another important aspect is that topical administration and oral supplementation of hydrolyzed collagen exhibit complementary effects, which suggests that cosmetic administration did not replace oral intake.

Another point explored is the inconsistency of evidence in dermatology that supports the indication of ingesting these bioactives orally and which patients actually need this supplementation in their routine. However, to date, it is suggested that hydrolyzed collagen supplementation is indicated for people over 30 years old or who have joint problems. Supplementation is also recommended for people with a protein deficit in their diet, which can potentially exacerbate joint problems or accelerate the loss of the skin's structural capacity. Among the classic clinical signs that point to the need for oral supplementation, include a decrease in skin firmness and elasticity, the appearance of wrinkles and expression lines, skin dehydration and peeling, hair loss and structural alteration of the strands, in addition to weak and brittle nails.

Over time, more evidence demonstrates the skin benefits associated with oral collagen supplementation. However, these works have factors that can be questioned, especially at the methodological level of clinical trials when compared with each other. Since the collagen supplements used in these studies have different compositions as they contain, in addition to collagen, vitamins, minerals and food concentrates, which

can directly influence the effects of supplementation. And another factor considered, is the variability in the biotechnological process to obtain the bioactive peptides that change the amino acid levels of the formulations, leading to data inconsistency.

And many studies are still considered limited by the scientific community because they only involve patients from specific geographic regions, sex and age groups. In addition, they are confronted with respect to their objective evaluation methodology when determining the results in the clinical appearance of patients and how in fact supplementation causes these effects in individuals.

Finally, the lack of consistent evidence suggesting that orally digested collagen is mainly deposited locally in the dermis instead of other regions of the body, makes the need for oral collagen administration controversial in patients with a balanced protein diet.

Thus, further studies are still needed to better elucidate the role of oral supplementation in photoprotection, in epidermal barrier diseases and even in skin aging. These additional studies should standardize the evaluation methods, establish ideal dosages, include a larger individuals range, using the methodology for longer studies. And most importantly, define in which patients collagen supplementation is necessary according to age, sex, skin type, ethnicity, clinical condition and skin comorbidities.

CONCLUSION

Based on the limited studies available in the literature, it can be concluded that oral supplementation of bioactive collagen peptides has demonstrated several promising short-and long-term effects on human skin health. Thus, CH supplementation becomes a potential adjuvant therapy in the prevention and/or treatment of pathologies and skin changes.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

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