

**Universidade de Lisboa
Faculdade de Farmácia**



**Exploration of the hair combing forces
resulting from different cosmetic
treatments**

Mariana Vieira da Silva e Vaz de Medeiros

Mestrado Integrado em Ciências Farmacêuticas

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**Monografia de Mestrado Integrado em Ciências Farmacêuticas
apresentada à Universidade de Lisboa através da Faculdade de Farmácia**

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Resumo

O cabelo é um dos aspetos mais importantes do ser humano devido às suas características fisiológicas, culturais, psicológicas e estéticas. O cabelo é sujeito a diversos cuidados, incluindo a lavagem, o pentear, a escovagem, a secagem, a modelagem e os tratamentos químicos e físicos. Naturalmente, os cabelos ficam danificados, especialmente se não for feita uma rotina de cuidados capilares adequada. Ao pentear, uma das técnicas de cuidado realizadas diariamente, é importante avaliar se novos produtos podem facilitar este processo, evitando a quebra dos fios de cabelo.

Neste trabalho foram estudadas as forças necessárias para pentear os fios de cabelo. Inicialmente avaliou-se qual seria a força necessária para manter uma velocidade constante do pente de 40mm/s. Ao ser aplicada mais força, esta demonstrava que o pente encontrava mais nós ou nós mais fortes, inviabilizando o pentear. Foram realizadas experiências em cabelo seco e molhado para determinar se o inchaço dos fios capilares, com os tratamentos utilizados, teria influência no processo de pentear. Para além destes ensaios, foram aplicados nos cabelos tratamentos com várias substâncias, nomeadamente o ácido málico e um péptido. O uso de um alfa-hidroxiácido, ácido málico, foi aplicado em cabelos virgens, cabelos que não foram submetidos a nenhum tratamento químico permanente e em cabelos descolorados. Para além disto, num segundo ciclo de testes, foi adicionado um péptido para verificar se a interação com o ácido málico beneficiaria ou prejudicaria a ação do ácido málico isoladamente. Os resultados demonstram que geralmente se aplica mais força ao processo de pentear, mostrando que o uso dos tratamentos não facilitou o processo de pentear.

Além disto, foi também estudada a força de tensão e extensão dos cabelos virgens e descolorados. Para isto, os fios capilares molhados foram colocados em molduras com cortes longitudinais para permitir a extensão do fio capilar até ao ponto de rutura. Foi possível observar um aumento significativo da extensibilidade nos cabelos virgens e descolorados, enquanto que os dados da força de tensão foram inconclusivos.

Este tema necessita de ser mais aprofundado no futuro, dado que o número de amostras foi reduzido e as amostras testadas eram de apenas cabelos caucasóides, sendo importante aumentar o número de amostras testadas e os tipos de cabelos testados.

Palavras-chave: Cabelo; Pentear; Ácido Málico; Péptido; Forças de Tensão e de Extensão.

Abstract

Hair is one of the most important aspects of the human being due to its physiological, cultural, psychological and aesthetically characteristics. Hair is subjected to grooming, including washing, combing, brushing, drying, styling, chemical and physical treatments. Naturally, hair will become more damaged especially if not appropriate hair care routine is made. Being combing one of the most grooming techniques done daily, it is important to assess if new hair care products can facilitate the combing experience, avoiding hair breakage.

In the combing experience, it was studied what force would be necessary to apply in order to keep a constant velocity of 40mm/s. If more force was applied, it explained that the comb encountered either more tangles or stronger tangles, thus the hair was not easier to comb. Experiments were conducted in both dry and wet conditions to determine if the hair swelling, carrying the treatments applied, would play a crucial role. It was analysed the use of an alpha hydroxy acid, Malic Acid, on both virgin hair, hair that was not subjected to any permanent chemical treatment, and bleached hair. Also, in a second cycle of testing, a peptide was added in order to see if the interaction with the Malic Acid would benefit or impair the action of Malic Acid alone. The results showcase that more force for the combing process was generally applied, pointing out that the use of the treatments did not ease the combing process.

Furthermore, it was studied the tensile strength and extensibility of the virgin and bleached hair. For this, hair fibres were placed in cases with longitudinal cuts to allow the extensibility of the hair fibre until the breakage point, under wet condition. It was possible to see a significant increase of the extensibility in both virgin and bleached hair whilst the tensile force data was inconclusive.

The topic needs to be further developed in the future since the number of samples was small. Also, as all tested tresses were Caucasoid hair, it is important to increase both the number of tested hair tresses and the types of hair tested.

Key words: Hair; Malic Acid; Combing; Peptide; Tensile Force and extensibility.

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1. Introduction

Hair nowadays plays a crucial role in several aspects. It serves as a protection to our scalp and has a significant importance aesthetically. Physiologically, hair helps to prevent insulation, offers protection against the sun and provokes an enhancement of the cutaneous sensation. Aesthetically, it is considered an ornamentation, can be a fashion statement and in some civilizations, it is associated to cultural practices. Thus, the hair is subjected to several types of grooming, from washing, combing, drying and treatments. All of these activities contribute to damage more the hair, making it necessary to discuss new products and practises to minimize the damage caused. With this study, it was investigated how different hair treatments could improve the combing experience, both in virgin and bleached hair.

1.1. Hair Structure, Morphology and Chemistry

Hair covers the majority of the scalp. The scalp differs from the remaining body skin as it has a large number of large hair follicles producing long, coarse hair fibres with large sebaceous glands attached to each follicle. Also, there is a complex network of blood vessels and nerves connected with the follicles. (1)

There are four stages for hair growth known as anagen (growth phase), catagen (transition or regression phase), telogen (resting phase) and exogen (shedding), becoming a cyclical event. (2) Every few year hairs are replaced by the hair cycle, functioning independently in each hair follicle. Each phase is characterised by specific timing, depending on the body location. On the scalp, anagen has a duration of more than three years, catagen usually lasts three weeks while telogen lasts three months. Anagen accounts for 90% of scalp hair while telogen and catagen account 10% and less than 1% respectively (3). The duration of the anagen phase will determine the hair length. (2)

The hair shaft is composed of three separate regions. The cuticle forms a thick sheath by the superimposition of several scales. It wraps the cortex that constitutes the most voluminous part of the fibre. In the cortex are located the fibrous proteins characteristic of hair, the α -keratins. The third zone, the medulla, is found close to the centre part of the hair. (1)

Hair's composition is mostly comprised of keratin with a small percentage of lipids, protein-bound sulphur, sugars and some trace elements. Keratin is a kind of protein that is formed when amino acids are linked together by polypeptide chain. Hair keratin is made up of 20 different types of amino acids being cystine the amino acid with the highest percentage.

Cystine is a sulphur-containing amino acid, having the highest proportion in all amino acids that are present in the hair fibre. The existence of disulphide bonds within the cystine provides resistance to the hair providing the chemical, mechanical and physical stability of the hair. The disulphide bonds in keratin can be irreversibly lost due to chemical processing and sun radiation since cystine is converted to cysteic acid. (4)

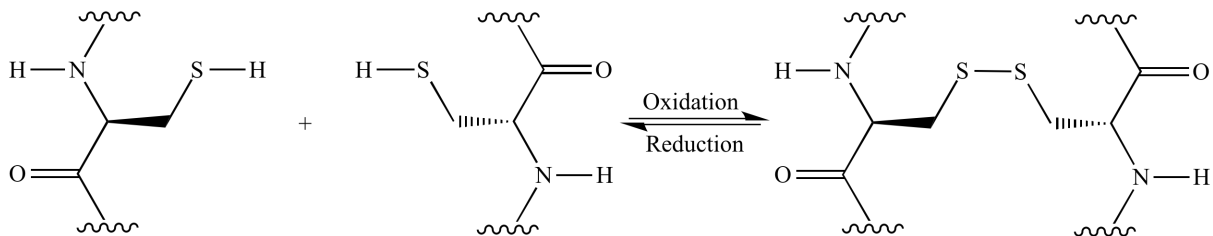


Figure 1: Disulphide bridge between two cystine residues (5)

Fully formed hair fibres are made up of three structures, the cuticle, cortex and medulla. The cuticle is the peripheral layer of the fibre and is responsible for the appearance and tactile perception of hair. It is a series of overlapping tough and rigid scales collectively whose purpose is to provide a resistant outer layer that surrounds the cortex. Individual cuticle scales are approximately 0.5 μm thick and up to six of these concentric layers are typically present in healthy hair. (6)

The cuticle is made up of several thin layers of overlapping cells, sub-sheets, which are each separated by an even thinner layer of cell membrane complex (CMC). The sub-sheets are known as epicuticle, A-layer, exocuticle and endocuticle, as shown in Figure 1. (7)

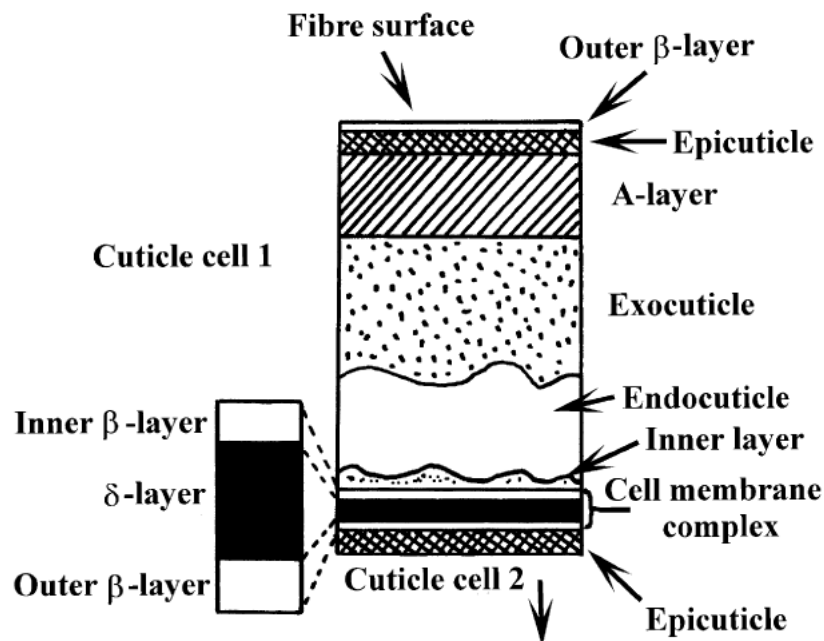


Figure 2: Schematic diagram illustrating the laminated subcomponents of the first cuticle cell in a transverse section (8)

The A-layer has a proteinaceous component and is located in the outer facing part of each cell, with a constant thickness of 110nm. Cystine disulphide crosslinks and iso-peptide crosslinks are constituents of the A-layer and the majority of the cystine residues is engaged in intermolecular crosslinking which was found to have a role in the protection of the hair surface against damaging agents, particularly chemical damage. Also, due to this characteristics, small water swelling can be expected while a high elasticity and hardness can be anticipated, especially when comparing with other structures, such as the hair cortex. Tensile stress when applied to the A-layer is anticipated to fracture more easily than other hair components becoming an advantage as it ensures that only smaller parts are shed from the cuticle scale, improving in the end, the cuticle's lifetime. (7) Since the hair cuticle is a multilayer structure, although there is shedding of small fractions of the cuticle cells, they leave a fresh and new uneroded surface belonging to the cuticle cell below. This allows for hair to continuously look appealing even though it is on a living head for several years. (9)

According to Swift (7), the exocuticle is, similarly to the A-layer, a proteinaceous sheet-like component being between 100nm and 300nm thick and having a rich cysteine content. Although it has a higher concentration of cysteine comparing to the cortex, the A-layer has even a higher concentration of cysteine.

Contrasting to the A-layer and the exocuticle, the endocuticle has a low concentration of cysteine, even though it showcases occasional pockets of cysteine-rich content, becoming a porous structure. This sheet-like subcomponent can vary its thickness between 50 to 300nm and contains higher levels of both acid and basic amino acids than other cuticle components. This, attached with the practically non-existent intermolecular crosslinks, such as disulphide bonds, points out that the component is mechanically soft and susceptible to substantial water swelling, becoming a harsh contrast with the A-layer and exocuticle. (7) When water penetrates the hair and causes swelling, it is lifting the hair cuticles. This allows components of different hair treatments and its materials to enter and exit out of the hair. (10)

The epicuticle involves a layer of lipids, which is mainly composed of 18-methyl eicosanoic acid (18-MEA) attached by thioester bonds to the thin proteinaceous layer. When exposed to ultraviolet and chemical treatments, the fatty acid layer can be deteriorated and therefore inhibiting its ability, as a semi-permeable membrane and lubricant, for reducing inter-fibre friction. (7) The epicuticle is able to adhere to neighbouring cuticle cells due to the intercellular cement. (11)

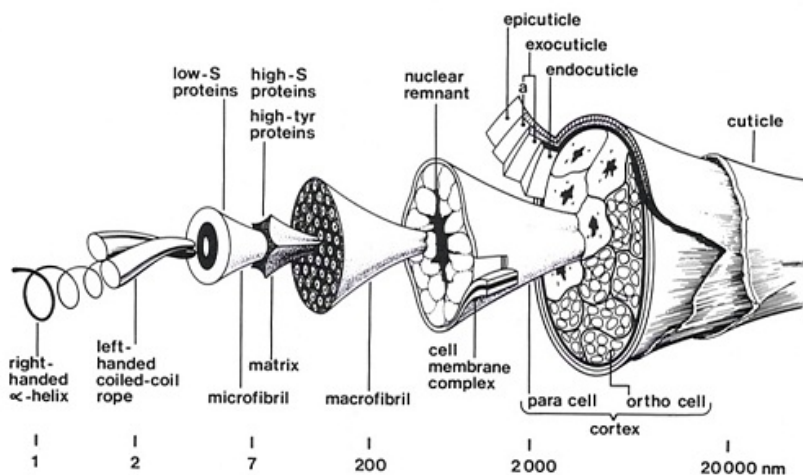


Figure 3 : Schematic diagram of the hair fibre (12)

The cortex represents approximately 80% of the hair and constitutes the core of the fibre, which its fundamental properties are present. It is the specific structure and the preferential orientation along the fibre axis of the subunits arranging the cortical cells that confer on the hair the mechanical properties. (1)

The cortex has on its constitution spindle shaped cortical cells, which are formed of microfibrils, containing the intermediate filaments and are encircled by a cystine-rich matrix. The intermediate filaments are formed of proteins that are twisted into α -helices, which have approximately three amino acids per turn and are stabilised by hydrogen bonds. (1)

The medulla is often completely absent, and, when present, it is sporadic and occupies part of the fibre, leaving large empty spaces between them. (1)

The shape of the hair is more evident when obtaining a cross section cut. Cylindrical shaped hair develops straight fibres (Mongoloid hair) while very elliptical shaped hair leads to curly or frizzy hair (Negroid hair). Negroid and Mongoloid hair have a cross-section noticeably larger than that of Caucasoid hair. (1)

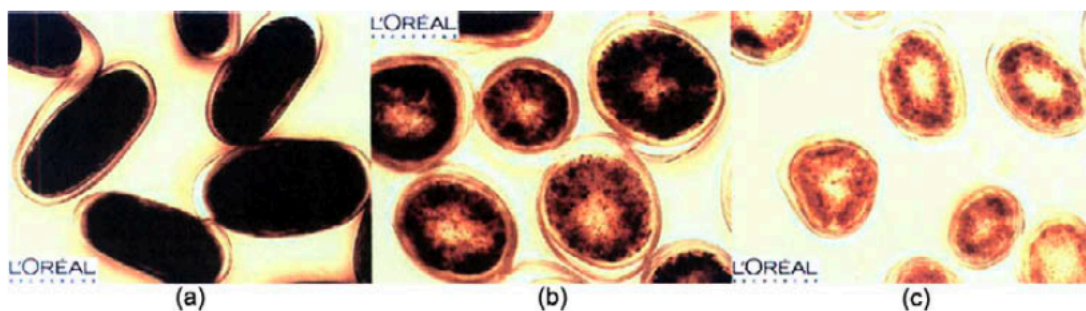


Figure 4: Cross-sectional view of hair from the three major ethnic groups: (a) Negroid hair; (b) Mongoloid hair; (c) Caucasoid hair (1)

Mongoloid hair when compared to Caucasoid hair showcases a rounder and thicker size. Also, it has a greater occurrence of medulla. Regarding the cuticles, Mongoloid hair has more cuticles, thicker cuticles with a narrow interval between the cuticles than Caucasoid hair. (13)

1.2. Combing

It was discovered that the loss of cuticles was detected starting a few centimetres away from the scalp and reduced along the fibre which is due to weathering and mechanical deterioration mainly concerning combing of hair, a grooming practice, that most humans, especially women, conduct daily.(9) Combing is considered a damaging process due to the fact that it is a repetitive movement and has a high combing force, especially in wet state and in the ends of dry hair. (14)

In combing dry hair, the force involved at the root end is low, but the force involved at the distal end is very high. The closer to the hair ends, the greater the combing damage, for both long and short hair. (9)

Brushing the hair while blow drying is more damaging due to the prolonged brushing and not the heat applied. The main effect of excessive hair brushing with the corresponding hair staining and, bending and twisting, is gradually wearing down the cuticle scales as the cuticle scales have smooth surfaces and smooth edges at the beginning of the scalp but its edges become rough and progressively eroded and once the scale is removed, the fibre end may split into two, three, or more longitudinal sections. (14)

Combing wet hair has more interfibre friction than in dry hair. Thus, wet hair is more difficult to comb and the combing force at the root end is higher and remains high throughout the length of the fibre. Since wet hair is weaker, it is more susceptible to damage from high combing forces due to hydrogen bonds being broken whenever hair is wet and only reformed when the hair dries. (14)

At different stages, breakage varied due to the difference in fibre interaction. In dry hair, tangles tend to occur at the end of the tress while in wet hair, they usually occur higher. When a tangle occurred, there was interaction between fibres and comb gave rise to compression, abrasion and tension (15). When tangles were combed out, cuticle disruption and fragmentation occurred (16).

1.2.1. Tensile Force

Consumers value the strength of hair as one of the most important aspects of hair care, considering that strong hair is healthy, beautiful, and vibrant. Hair that is considered fragile, brittle and weak is often associated to poor condition and care and its often judged involving the ease of hair breakage. It is possible to assess scientifically the strength of individual hair fibres through standard mechanical testing approaches, mostly the generation of stress-strain curves using constant-rate extension experiments. A schematic of a typical stress-strain curve and the parameters that can be extracted to provide quantification are shown in Figure 5. (17)

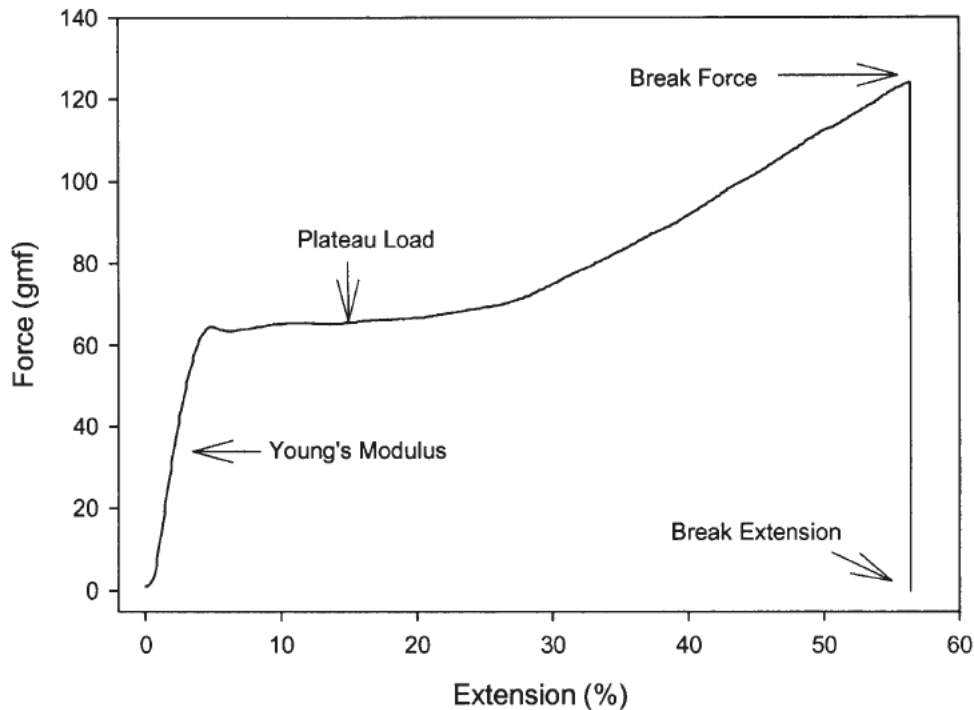


Figure 5: Stress-strain curve for dry hair (17).

This schematic of a stress-strain curve for hair showcases the pseudolinear region that occurs at low strains. Within this region the application and removal of an applied deformation leaves fibres in a recovered state. However, if an applied force exceeds that of the linear-like region, the fibre yields, becoming permanently deformed, and retaining a residual strain upon removal of the applied force (17).

During the extension of keratin fibres, the cuticula and the cortex, the two biggest morphological components, accommodate the stresses imposed on the fibre in different ways. (11)

Regarding the multilayer structure of the cuticle, this stress is accommodated by the various layers within each cuticle cell and by the bonding layers between the cells, ultimately causing the lifting of surface scale edges at higher strain levels. This leads to failure in the weak endocuticular layer and causes the delamination and lifting of the outer layers of the surface cuticle being the damage irreversible.(11) The phenomenon is thought to be due to different extensibilities in the layered cuticle structure and during elongation, the separation of these cuticle layers causes the roughness in the cuticle surfaces and edges. (18)

Regarding the cortex, the elongation is within the hair's elastic region, the hydrogen and ionic bonds were disrupted. When exceeding the elastic region, structure of the intermediate filaments changed as the α -helix starts to unravel into β -helix structure. (18)(19).

Despite cuticle damage, tensile property of hair relies exclusively on the cortex as it was demonstrated by Robbins and Crawford (20) where tensile properties of fibres with cuticle damaged from chemicals, did not differ from the original fibres. The decreases in the wet tensile properties are attributed primarily to the degradation of the disulphide bonds present in the cortex. There were no other significant changes in the wet tensile properties, but gross alterations were visible in the cuticle layer. Whilst in the dry condition, the tensile properties are due to the degradation of the peptide bonds, present mostly in the cortex.

Tensile testing conducted by Ruetsch and Weigmann (11) have shown that under dry conditions, exposing fibres to high extension levels (30 – 35% elongation) and keeping it in the elongated state for 5 minutes resulted in lifting of cuticles due to delamination from endocuticle failure. Immersion in water have enabled most of the cells to return to their original state, but the recovery decreased with repeated elongation at lower extension level. Although the authors suggested that years of exposure to tensile stress from grooming could cause a negative impact on the cohesiveness of cuticle cells, the rate of extension was not mentioned in the paper. Such endocuticle damage could be due to the slow rate and prolonged strain employed which do not occur in real life combing situation.

Damage around the fibre is most probably due to the pressure applied on the cuticle-cortex CMC, by the swollen cortex. When swelling occurs, its pressure causes crack propagation during deformation to extend across the cuticle layers perpendicularly to the hair fibre axis (21).

1.3. Washing

Washing, which comprises soaking hair in water, leathering and drying is a factor contributing to hair cuticles' erosion. Each time hair is washed and dried, the cuticle structure must expand and contract to greater extents to accommodate the increased cortical dimensions since water is absorbed, This repeated habit causes thermal expansion and contraction causing greater levels of swelling when fibres are immersed in water (6). In the case of wet combing, as explained previously, it can cause breakage of hair fibre and erosion of hair cuticles.

1.4. Bleaching

Hair plays a crucial role in the appearance of the human being. Its colour has a deep influence in its appearance, being considerate a reflection of its health state. Throughout several years, different chemical treatments have been applied to change the colour of hair. The natural hair

colour is composed by an insoluble pigment called melanin which can be pheomelanin or eumelanin, red or brown colour respectively. In order to change the hair colour, it is necessary to expose the hair to either of the following treatments, lightening the natural colour through bleaching, adding a new colour to the existing hair colour or do both. (22)

Bleaching is used to achieve a lighter hair colour through the decolourisation and solubilisation of the melanin pigments by oxidants (22). However, the disulphide bonds within and between proteins in the cuticle and cortex are oxidised and cysteic acid is produced which increased significantly the static electricity was produced when the hair was combed (23).

Although bleaching treatments damage the cuticle, subsequent exposure to mechanical stresses was responsible for the removal of cuticles. Due to cuticle degradation and decrease in 18-MEA, bleached fibres are predictable to feel drier, experience a greater fibre swelling under wet conditions and higher friction during combing. (23).

1.5. Treatments

Alpha hydroxy acids are natural acids that can be found in several types of food. They include citric acid (found in citrus fruits, such as oranges and lemons), glycolic acid (found in sugar cane), lactic acid (milk), malic acid (found in apples), and others and are obtained from their natural sources or may be made synthetically. Alpha hydroxy acids are most commonly used for skin conditions such as dry skin, wrinkled skin, or acne. Products containing AHA ingredients may be for consumer use, salon use, or medical use, depending on the concentration and pH (24).

Malic Acid, 2-hydroxybutanedioic acid, is not commonly used in cosmetic treatments, thus, it is not well known its activity, both in skin and hair. With this project, it was intended to shed some light for this component and understand if its use would be beneficial for the combing experience.

The penetration of chemicals occurs mainly through the intercellular diffusion of the hair. Virgin hair has an isoelectric point around 3.67, thus the hair surface carries a negative charge, making the hair care products formulation to include cationic polymers. When hair is damaged, for example, bleached, there is loss in body, lustre and smooth texture, resulting in a poor wet and dry combability since there is an increase of electrostatic charging. Using protein materials in hair care formulations takes place in order to provide shine, strength, softness and good combing properties. With virgin hair, water and chemicals are hardly absorbed and penetrate

into the hair surface. When hair is bleached, the peptide penetration increases since it becomes a self-adjusting system due to the increase in the negative charge of the hair. (25) The protein solution utilized was made with Hydrolysed vegetable protein PG-Propyl Silanetriol Water (HVPPS) in order to see if, together with Malic Acid, the combability experience would be eased.

2. Aim and Objectives

2.1. Aim

The aim of this project was to explore the effect of an alpha hydroxy acid with or without a peptide on the combing forces and the extensibility of hair.

2.2. Objectives

- a. To understand if the use of an alpha hydroxy acid, malic acid, would improve the combing experience in both virgin and bleached hair;
- b. To understand if the use of an alpha hydroxy acid, malic acid, would differ on the tensile force and the extensibility of both virgin and bleached hair;
- c. To understand if the use of a peptide, HVPPS, would improve or inhibit the combing experience after malic acid being used.
- d. To understand if the use of a peptide, HVPPS, would improve or inhibit the Tensile force applied and affect the extensibility after malic acid being used.

3. Materials and Methods

3.1. Materials

3.1.1. Hair Tress

Experiments were conducted on fine European (Caucasoid) brown hair supplied by Banbury Postage UK. Each hair tress was 25cm long, 1,5cm wide and weighed 5g. Tresses were stored in a 35°C oven overnight.

3.1.2. Malic Acid

Malic Acid, or 2-hydroxybutanedioic acid, with a solubility of 55,8g/100ml was utilised to create a 5% acid solution. (26)

Malic Acid was used due to being an alpha hydroxi acid which restores the electrostatic bonds between proteins. In order to have a pH between 3 and 3,5, 20% sodium hydroxide was utilised to achieve the goal pH. The final solution had a pH=3,01, being distributed in different beakers to be used during the experiment. Before each test, the pH was tested having a range between 2,9 and 3,3.

3.1.3. Protein

The protein solution utilised was made with 1% of Hydrolysed vegetable protein PG-Propyl Silanetriol (and) Water (HVPPS).

3.1.4. SLES

Sodium Laureth Sulfate (SLES), or disodium 1-dodecoxydodecane sulfate was utilised to wash all hair tresses according to the Washing Protocol. Its pH was between 6,5 and 7. SLES was utilised to remove any contaminants or interferences as it is an anionic surfactant. A 20% aqueous solution was prepared previously. (27)

3.1.5. Bleaching Chemicals

The Wella Professional Multi-Blondor Powder and Wella Professional Welloxon Perfect Peroxide Developer 12% 40vol were used for the Bleaching Process. Bleaching took place to evaluate if the hair would have potential measurement changes during the experiments.

3.1.6. Comb

Pro Tip small cutting comb PTC01 was used to comb the hair tresses. The comb was washed with sodium laureth sulfate (SLES) between each Combing Process and the different hair tresses.

The ampler side had 31 tines with an interteeth distance of 0.2cm whereas the finer side had 64 tines with an interteeth distance of 0.1cm.

3.1.1. Measuring Devices

For the Combing Experience, the TA.XT Plus Texture Analyser, Stable Micro Systems (United Kingdom) was fitted with a comb and grips. The Exponent software was utilised, being calibrated to a combing speed of 40mm/s.

For the Extensibility Experience, the TA.XT Plus Texture Analyser, Stable Micro Systems (United Kingdom) was used. Cases were placed into the grips and extended 10mm/s until the hair fibre reached the breakage point.

3.2. Methods

3.2.1. Washing Protocol

The hair tresses were washed with SLES (20%) previously to each Combing Protocol in order to guarantee that any residues were removed. Tresses were rinsed under running tap water for 30 seconds, washed during 1,5 minutes with 2.0ml of SLES using a syringe and massaging the hair carefully to avoid creation of excessive tangles, rinsed for 1,5 minutes and repeated once more. Hair tress were squeezed between the middle and forefinger while moving in a downward motion and repeat it for another 2 times to remove the excess water. This allowed avoiding possible contaminations and interferences. From now on, every time wash is mentioned it was conducted accordingly to the Washing Protocol.

3.2.2. Bleaching Protocol

Bleaching was carried with Wella Professional Multi-Blondor Powder and Wella Professional Welloxon Perfect Peroxide Developer 12% 40vol. Each hair tress was bleached with 10 g of the bleaching mix, 4,36g of powder to 7,64g of developer (1: 1,75), being the bleaching mix a total of 100g. Tresses were bleached for 10 minutes wrapped in foil at room temperature

3.2.3. Combing Experience

3.2.3.1. Flow of Treatments and Measurements

The hair treatment sequence has been outlined below. 10 replica tresses were used for each stage of the treatment and the results were averaged.

1) Virgin Hair:

- a. Cycle 1: No active treatment;
- b. Cycle 2: Active treatment with Malic Acid
- c. Cycle 3: Active treatment with Malic Acid and HVPPS

2) Bleached Hair

- a. Cycle 1: No active treatment;
- b. Cycle 2: Active treatment with Malic Acid
- c. Cycle 3: Active treatment with Malic Acid and HVPPS

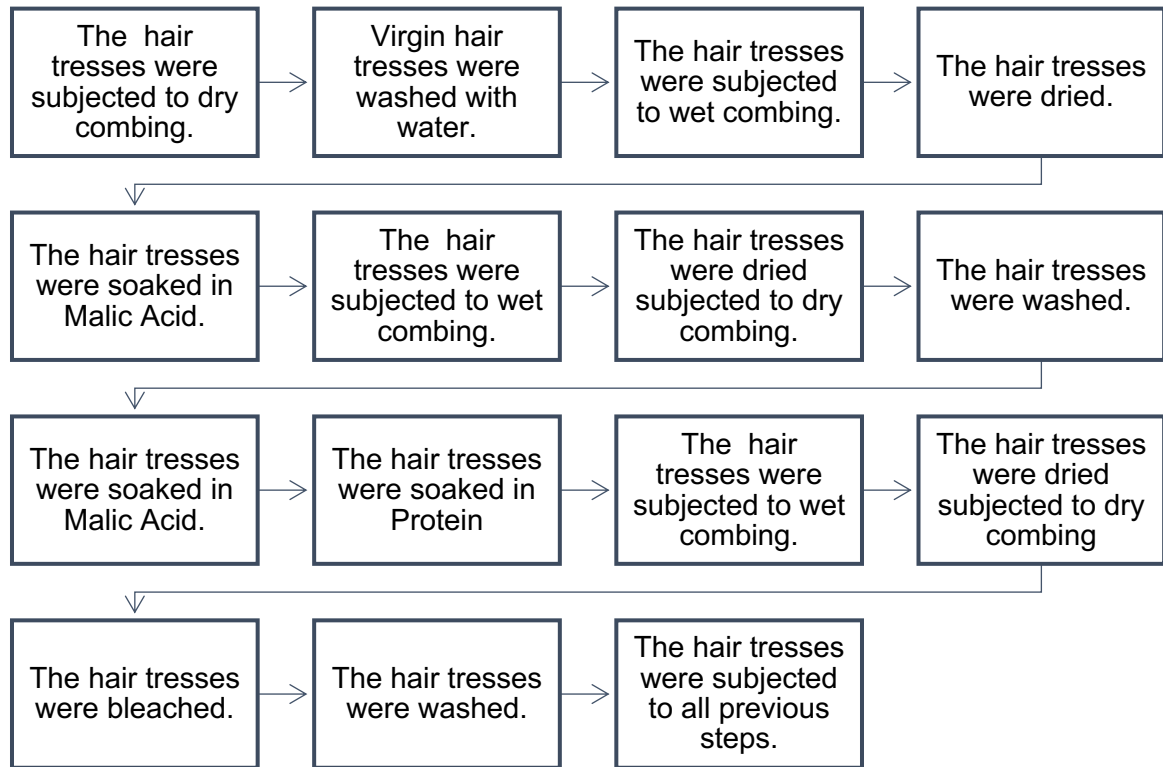


Diagram 1: Treatments and hair combing measurements conducted on hair tresses.

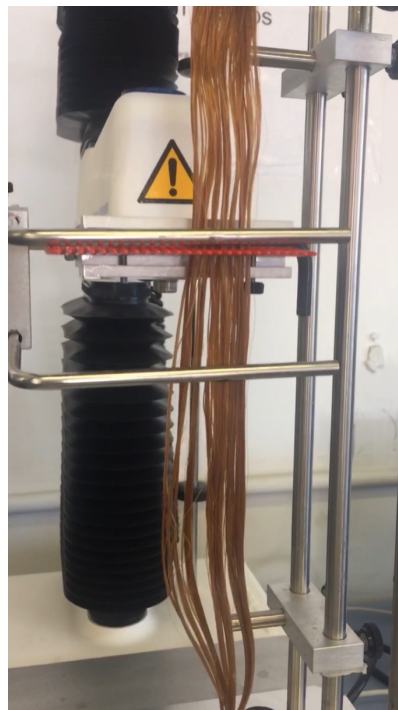


Figure 6: Combing Experience

3.2.3.2. Dry Combing Protocol for Virgin Hair Cycle 1

Table 1: Dry Combing Protocol for Virgin Hair Cycle 1

Activity
1. Ensure that gloves are worn before handling the hair tress.
2. Remove the hair tress from the oven.
3. Place the thermostat-hydrometer next to the set up.
4. Record the date, room temperature and humidity.
5. Remove static with an anti-static gun.
6. With a hairdryer (cool setting and low airflow), tangle the ends of the hair tress and individual fibre for 45 seconds and remove any static with an anti-static gun.
7. Name the file accordingly and start the recording.
8. Set the speed to 40 mm/s and adjust to the calibrated height.
9. Start the combing experiment.
10. If the comb is in contact with a severe tangle, do not force it through. Stop the experiment, detangle the knot and restart from the top of the hair tress.
11. Repeat step 9 to 11 for another 3 times.
12. Upon completion, remove the hair tress and measured fibre and store it back in a 35°C oven.

3.2.3.3. Wet Combing Protocol for Virgin Hair Cycle 1

Table 2: Wet Combing Protocol for Virgin Hair Cycle 1

Activity
1. Ensure that gloves are worn before handling the hair tress.
2. Remove the hair tress from the oven.
3. Place the thermostat-hydrometer next to the setup.
4. Record the date, room temperature and humidity.
5. Soak the hair tress into a 1 L beaker of water for 10 minutes.
6. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
7. Dip the hair tress into a beaker of water for 3 times to create tangles.
8. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
9. Name the file accordingly and start the recording.
10. Set the speed to 40 mm/s and adjust to the calibrated height.

11. Start the combing experiment.
12. If the comb is in contact with a severe tangle, do not force it through. Stop the experiment, detangle the knot and restart from the top of the hair tress.
13. Repeat step 9 to 11 for another 3 times.
14. Upon completion, remove the hair tress and measured fibre and store it back in a 35°C oven.

3.2.3.4. Wet Combing Protocol for Virgin Hair Cycle 2

Table 3: Wet Combing Protocol for Virgin Hair Cycle 2

Activity
1. Ensure that gloves are worn before handling the hair tress.
2. Remove the hair tress from the oven.
3. Place the thermostat-hydrometer next to the set up.
4. Record the date, room temperature and humidity.
5. Soak the hair tress into a 1 L beaker of water for 10 minutes.
6. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
7. Soak the hair tress into a 300ml beaker of Malic Acid for 10 minutes.
8. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
9. Dip the hair tress into a beaker of Malic Acid for 3 times to create tangles.
10. Remove the excess by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
11. Name the file accordingly and start the recording.
12. Set the speed to 40 mm/s and adjust to the calibrated height.
13. Start the combing experiment.
14. If the comb is in contact with a severe tangle, do not force it through. Stop the experiment, detangle the knot and restart from the top of the hair tress.
15. Repeat step 9 to 11 for another 3 times.
16. Upon completion, remove the hair tress and measured fibre and store it back in a 35°C oven.

3.2.3.5. Dry Combing Protocol for Virgin Hair Cycle 2

Dry combing with for Virgin Hair Cycle 2 was conducted after the protocol shown in Table 3 was carried. The hair tresses were dried with the hairdryer for 10 minutes and placed in

the oven until fully dried. Afterwards, the protocol followed was conducted as shown in Table 1.

3.2.3.6. Wet Combing Protocol for Virgin Hair Cycle 3

Table 4: Wet Combing Protocol for Virgin Hair Cycle 3

Activity
1. Ensure that gloves are worn before handling the hair tress.
2. Remove the hair tress from the oven.
3. Place the thermostat-hydrometer next to the set up.
4. Record the date, room temperature and humidity.
5. Soak the hair tress into a 1 L beaker of water for 10 minutes.
6. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
7. Soak the hair tress into a 300ml beaker of Malic Acid for 10 minutes.
8. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
9. Soak the hair tress into a 300ml beaker of HVPPS for 10 minutes.
10. Remove excess water by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times
11. Dip the hair tress into the beaker of HVPPS for 3 times to create tangles.
12. Remove the excess by squeezing the hair tress between the middle and forefinger while moving in a downward motion and repeat it for another 2 times.
13. Name the file accordingly and start the recording.
14. Set the speed to 40 mm/s and adjust to the calibrated height.
15. Start the combing experiment.
16. If the comb is in contact with a severe tangle, do not force it through. Stop the experiment, detangle the knot and restart from the top of the hair tress.
17. Repeat step 9 to 11 for another 3 times.
18. Upon completion, remove the hair tress and measured fibre and store it back in a 35°C oven.
19. Ensure that gloves are worn before handling the hair tress.
20. Remove the hair tress from the oven.

3.2.3.7. Dry Combing Protocol for Virgin Hair Cycle 3

Dry combing with Malic Acid and HVPPS for Virgin Hair was conducted after the protocol shown in Table 4 was carried. The hair tresses were dried with the hairdryer for 10 minutes and placed in the oven until fully dried. Afterwards, the protocol followed was conducted as shown in Table 1.

3.2.3.8. Combing Protocol for Bleached Hair

Combing of bleached hair tresses, both dry and wet, was conducted after each tress being subjected to the bleaching process. Afterwards, each protocol was carried as shown, respectively, in Table 1, Table 2, Table 3, the 3.2.3.5 Dry Combing Protocol, Table 4 and 3.2.3.7 Dry Combing Protocols.

3.2.4. Extensibility Experience

Hair is damaged due to several grooming experiences such as washing. Since during washing hair is pulled, twisted and stretched, the Extensibility test, very commonly used in the industry, was carried to assess if the Active Treatments would improve the Extensibility of the hair.

3.2.4.1. Flow of Measurements

A total of 82 hair fibres were removed, one from each hair tress (2 control tresses, 20 Virgin with Malic Acid, 20 Virgin with Malic Acid and HVPPS, 20 Bleached with Malic Acid, and 20 Bleached with Malic Acid and HVPPS) were mounted individually on paper card as shown below. The fibres were positioned and attached securely to the paper card with strong tape. Care was taken to position the fibres perpendicularly to the short sides of the card and parallel to the long sides of the card, being afterwards placed in the TA.XT Plus Texture Analyser, Stable Micro Systems (United Kingdom) to measure the Tensile Strength and Extensibility of each hair fibre. Each hair fibre was tested while still wet. The procedure stopped when the hair fibre was extended until the breakage point.

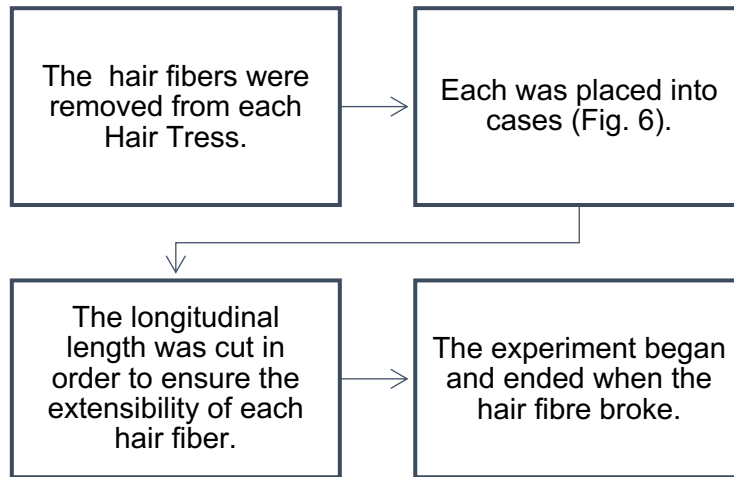


Diagram 2: Measurements conducted on hair tresses.

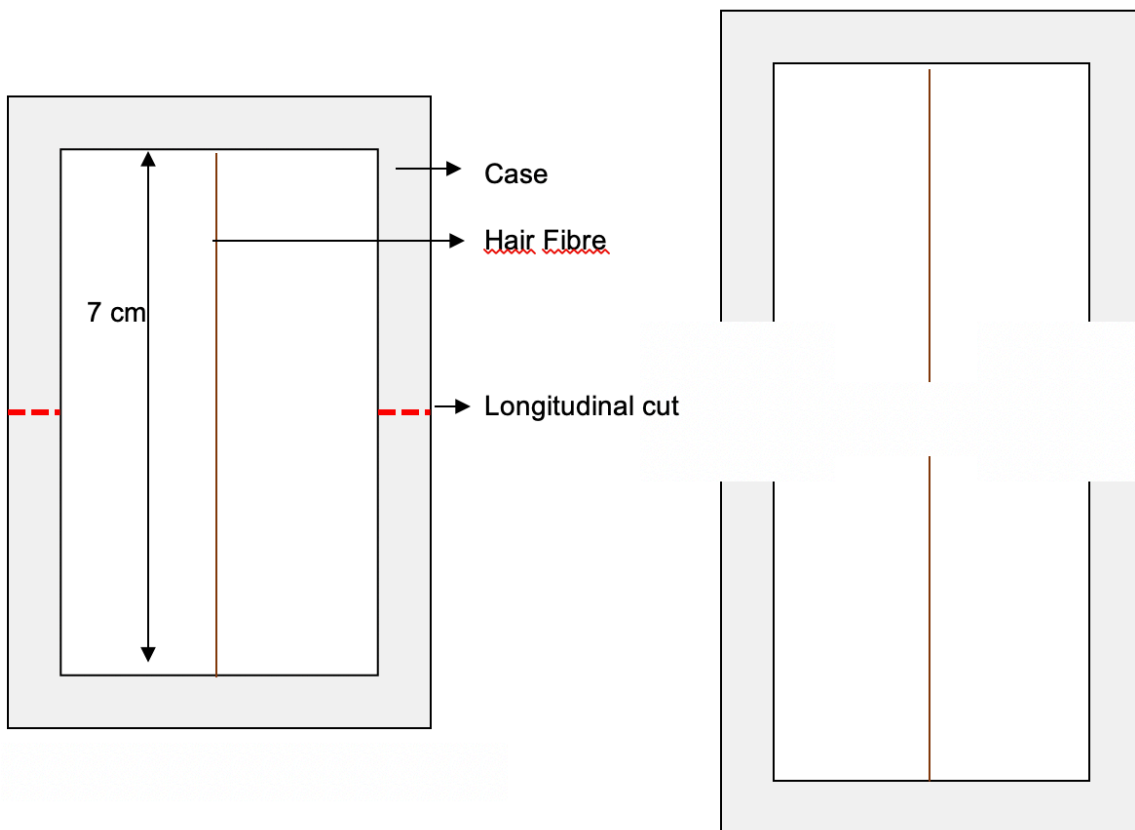


Figure 6: Scheme of the Hair case on the initial phase and after the breakage point

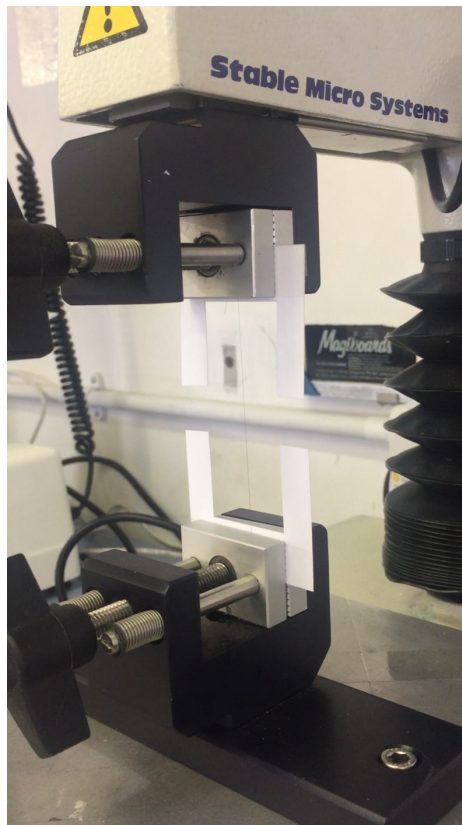


Figure 7: Hair fibre being stretched

4. Results

To analyse the results, a code system was used to name the Hair Tresses.

Table 5: Hair Tresses code name system

Name	Code
Virgin	V
Bleached	B
Dry	D
Wet	W
Control	C
Malic Acid	MA
Malic Acid and HVPPS	AP

4.1. Combing Experience

Ten hair tresses were utilised, each experiment consisted of 4 measurements, being the first one discarded. Three experiments were carried on each Hair Tress. All results showcase the average force (N).

Table 6: Combing Experience results for Virgin Hair

Force (g)	Cycle 1		Cycle 2		Cycle 3	
	Wet	Dry	Wet	Dry	Wet	Dry
V1	45,310	30,126	157,27	90,786	62,824	51,408
V2	45,699	48,686	149,42	59,725	78,423	40,849
V3	36,457	35,599	158,22	47,823	46,634	63,097
V4			207,28	80,573	57,859	93,193
V5			172,60	62,191	57,534	64,176
V6			126,27	53,815	52,523	53,405
V7			177,70	134,50	66,235	62,464
V8			253,28	129,98	68,261	41,997
V9			178,53	150,60	73,351	76,598
V10			194,63	54,368	62,985	129,41
Average	42,489 ± 5,2272	38,137 ± 9,5367	177,52 ± 33,381	86,436 ± 38,386	62,663 ± 9,5217	67,660 ± 26,769

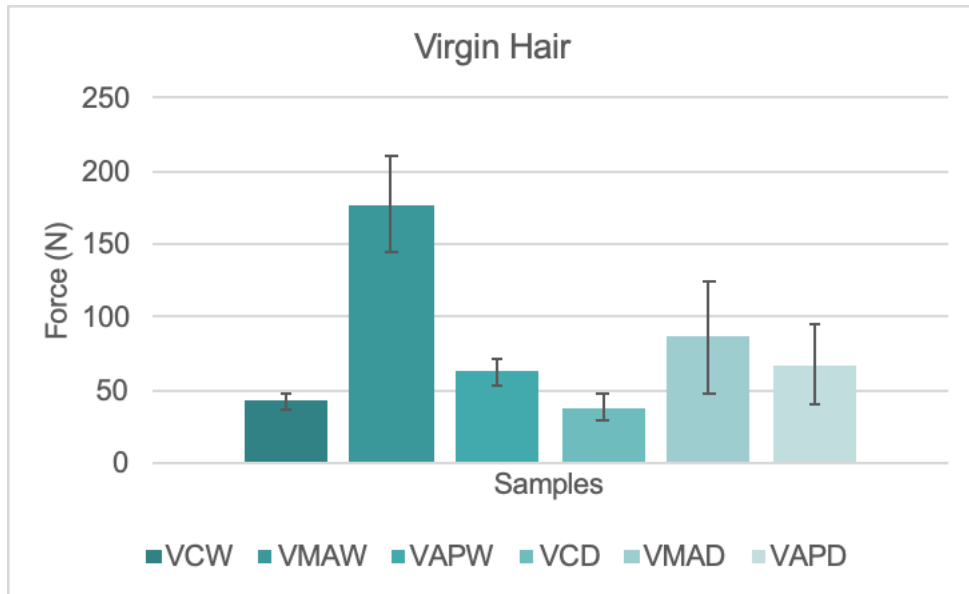


Figure 7: Graph demonstrating the Force used in Virgin Hair Tresses

The same experience was carried on Bleached Hair.

Table 7: Combing Experience results for Bleached Hair

Force(g)	Control		Malic Acid		Malic Acid and HVPPS	
	Wet	Dry	Wet	Dry	Wet	Dry
B1	155,97	41,716	136,73	47,209	114,93	64,229
B2	168,26	68,896	121,13	38,022	126,39	55,552
B3	84,44	45,028	116,63	79,457	231,59	98,179
B4			64,335	72,655	179,39	104,68
B5			67,115	64,110	157,95	39,016
B6			50,810	111,26	123,99	110,86
B7			188,48	61,956	136,24	194,70
B8			94,796	74,819	91,742	187,03
B9			65,545	43,196	70,217	56,229
B10			68,416	59,545	77,466	112,96
Average	136,22 ± 45,269	51,880 ± 14,829	97,398 ± 43,228	65,223 ± 21,248	130,99 ± 49,039	102,34 ± 53,425

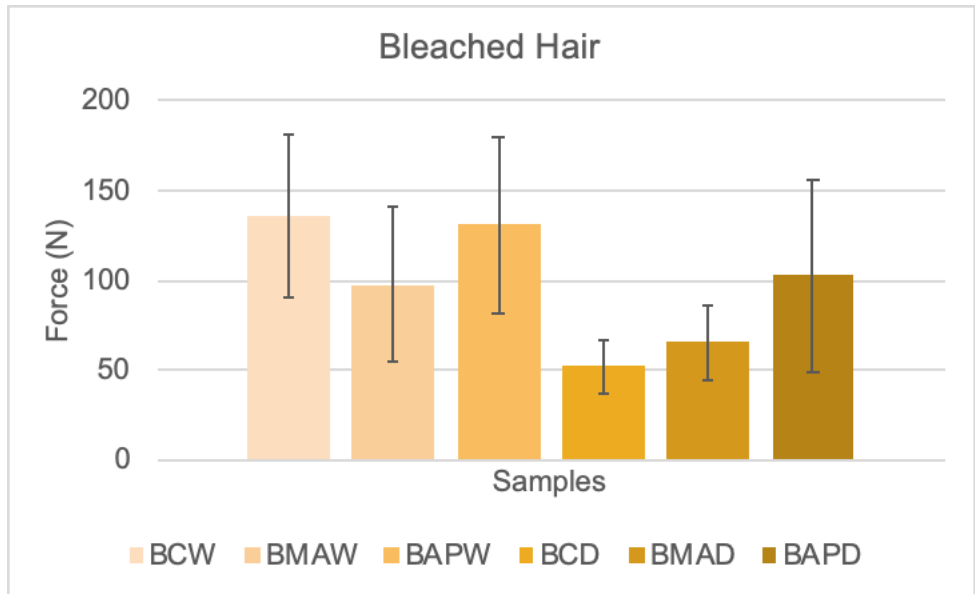


Figure 8: Graph demonstrating the Force used in Bleached Hair Tresses

4.2. Extensibility Experience

Table 8: Results of the Extensibility Experience in Virgin Hair

	Tensile Strength N Force	Extensibility mm Distance		Tensile Strength N Force	Extensibility mm Distance
VC	0,7600	22,32	VC	0,7600	22,32
V1MA	0,7300	21,33	VAP1	1,220	27,05
V2MA	0,9600	31,09	VAP2	1,020	24,57
V3MA	0,8900	30,62	VAP3	0,6600	27,92
V4MA	1,380	28,11	VAP4	0,9600	30,61
V5MA	1,080	34,85	VAP5	1,270	31,56
V6MA	1,220	31,58	VAP6	1,070	27,85
V7MA	2,050	0,8400	VAP7	1,090	30,08
V8MA	1,050	33,12	VAP8	0,6600	24,04
V9MA	1,470	30,38	VAP9	1,000	28,71
V10MA	1,200	30,60	VAP10	1,210	28,79
V11MA	0,9100	31,05	VAP11	0,7300	34,85
V12MA	0,5500	12,04	VAP12	0,8800	29,08
V13MA	0,8500	31,52	VAP13	1,330	31,25

V14MA	1,070	30,39
V15MA	0,9200	19,83
V16MA	1,430	30,31
V17MA	1,390	33,33
V18MA	0,7600	27,93
V19MA	1,670	32,78
V20MA	1,420	28,92
Average	1,150 ± 0,3556	27,53 ± 8,161

VAP14	0,7000	26,68
VAP15	1,140	30,20
VAP16	1,540	34,69
VAP17	1,030	31,17
VAP18	1,120	30,41
VAP19	0,9200	29,05
VAP20	1,040	28,93
Average	1,030 ± 0,2281	29,38 ± 2,711

The same experience was carried on Bleached Hair.

Table 9: Results of the Extensibility Test in Bleached Hair

	Tensile Strength	Extensibility
	N	mm
	Force	Distance
BC	1,220	17,9
BMA1	0,8200	24,75
BMA2	0,8900	21,43
BMA3	1,150	33,10
BMA4	0,9300	28,70
BMA5	1,110	26,91
BMA6	0,9100	20,86
BMA7	1,060	30,39
BMA8	1,020	31,71
BMA9	1,120	31,28
BMA10	1,080	32,41
BMA11	0,7500	23,94
BMA12	0,8000	29,91
BMA13	0,9400	33,69
BMA14	1,210	33,46
BMA15	0,8900	33,54
BMA16	0,9800	26,49

	Tensile Strength	Extensibility
	N	mm
	Force	Distance
BC	1,220	17,9
BAP1	0,9300	24,13
BAP2	1,460	27,07
BAP3	0,8500	30,24
BAP4	1,030	23,86
BAP5	1,250	29,36
BAP6	0,6600	27,82
BAP7	0,9900	29,40
BAP8	0,9100	25,18
BAP9	0,7100	30,32
BAP10	1,040	31,34
BAP11	1,050	29,24
BAP12	0,8400	25,23
BAP13	0,8800	29,17
BAP14	0,5000	29,72
BAP15	0,7900	28,70
BAP16	0,7900	29,66

BMA17	1,060	30,96
BMA18	0,8800	30,06
BMA19	0,9300	35,21
BMA20	1,170	29,30
	0,9850 ±	29,40 ±
Average	0,1295	4,080

BAP17	1,160	30,10
BAP18	1,140	30,45
BAP19	0,9700	32,26
BAP20	0,9600	31,33
	0,9455 ±	28,73 ±
Average	0,2125	2,398

The Tensile Force test was conducted but the data that was collected was inconclusive. Thus, it indicates that a larger number of the fibre replicas might be required to have conclusive and trustworthy results.

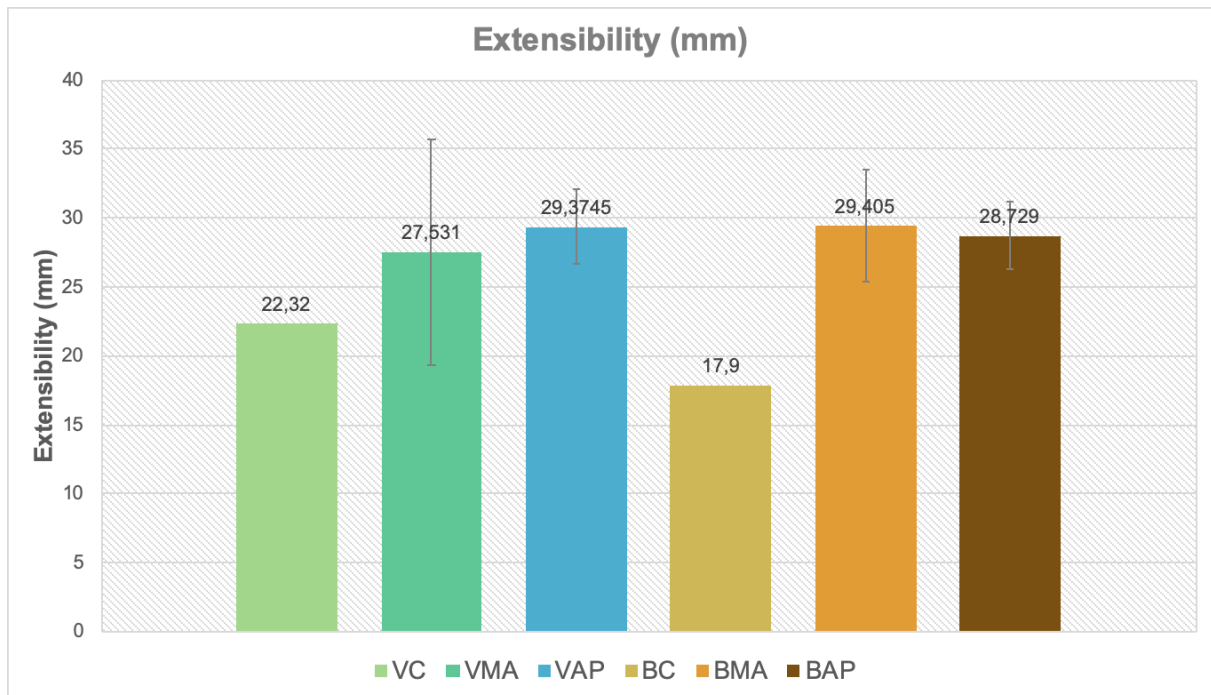


Figure 9: Hair Extensibility under wet condition

5. Discussion

5.1. Combing Experience

5.1.1. Virgin Hair

Under wet condition, treatment with Malic Acid results rose sharply when comparing to Control results. The use of HVPPS (polymer) reduced the stiffness of the hair, showing that it was not necessary as much force to maintain the velocity (40mm/s), thus fewer tangles were

experienced. Although its use, it was still necessary to increase the force when comparing to treatment with Malic Acid results.

Under dry condition, an increase of force utilised was observed in both treatments with Malic Acid and treatments with Malic Acid and HVPPS. As observed under wet condition, the use of HVPPS reduced the force utilised when comparing to treatment with Malic Acid results.

5.1.2. Bleached Hair

Under wet condition, there was a decreased utilisation of force after Treatment with Malic Acid. This illustrates that the use of Malic Acid in Bleached hair decreased the tangles and interactions, which was observed during the experiments as the hair tresses were softer and more malleable. After Treatment with Malic Acid and HVPPS, the force increased, showcasing that the use of the HVPPS almost cancelled the benefits of using Malic Acid as both values of Control and Treatment with Malic Acid and HVPPS are similar.

The results go against the expected since in damaged hair, peptides increase more easily the hair, improving the shine and smoothness. (25)

Under dry condition, an increase of force utilised was witnessed. A clear increase was observed especially after the Treatment with Malic Acid and HVPPS, demonstrating that the use of HVPPS increased, even more, the stiffness of hair, as predicted.

5.1.3. Comparison

When comparing these results, it is important to point out that the only occasion when the active treatments had a result that showcased improvement was in Bleached Hair under wet condition. This could be due to the significant damage present in the Bleached Hair, and as Malic Acid is an alpha hydroxy acid it helped to restore the electrostatic bonds between proteins, making the hair easier to comb. Also, since Control had no active treatment it was expected that, under wet condition, the combing force increased as there was hair swelling and more interfibre friction. The addition of water created more surface tension since it was acting as glue between the fibres.

As expected, the HVPPS increased the force utilised in the majority of the cases, confirming the claim that it provides both stiffness and volume to hair.

The standard deviation is higher in the Bleached Hair measurements, showcasing that more tests have to be conducted to confirm the results.

5.2. Extensibility Experience

5.2.1. Tensile Strength

Regarding Virgin Hair, there was an increase of the Tensile Force when using both active treatments. When adding the HVPPS, a minor decrease is witnessed.

Regarding the Bleached Hair, there is a decrease in both treatments.

5.2.2. Extensibility

Regarding Virgin Hair, it is clear to state the Extensibility increased when using Malic Acid. In other words, this states that the hair tresses elongated more before breakage. When adding the HVPPS the Extensibility increased slightly, but since the sample was small, it is not possible to know for a fact if the HVPPS played a crucial role.

Regarding the Bleached Hair, a significant increase of Extensibility was present, being even higher than the Virgin Hair Control results. This demonstrates that the use of Malic Acid provided more strength to the hair fibres, allowing them to stretch more before reaching its breaking point. The results of the Treatment with Malic Acid and HVPPS showcase a slight decrease in the Extensibility.

When comparing virgin and bleached hair, and since bleached hair is stiffer than virgin hair, once the treatments were applied, the bleached hair's expansibility increased and was similar to the results of the virgin hair, showcasing that the Malic Acid played an important role.

6. Conclusion

This study was conducted in order to consider and evaluate the usage of an alpha hydroxy acid, in this case Malic Acid, as a substance that would facilitate the combing experience and increase the hair strength and extensibility. Since proteins are used in hair care formulas, it was also conducted a round of experiments using both Malic Acid and HVPPS.

The aim set for this project was mostly achieved as conclusions can be made regarding the use of Malic Acid both in the Combing Experience and the Extensibility Experience. It was not possible to reach any conclusions regarding the Tensile Force due to the data being inconclusive. The project was very useful to understand the benefits and disadvantages of the use of Malic Acid together or not with the HVPPS.

Since bleached hair is severely damaged, it was important to understand if the use of an alpha hydroxi acid would help to improve the combing experience. It is possible to conclude that the use of Malic Acid in wet hair improved the combability as less force was required, meaning that less tangles were encountered when combing the hair. When adding the HVPPS, there was an increase of force applied, both in dry and wet conditions, suggesting that using both treatments would not be beneficial for the consumer. Regarding the use of Malic Acid in dry bleached hair, there was a slight increase of force utilised, but no conclusions can be made since the standard deviations are significant.

While in bleached hair there were positive results, with Virgin hair, there was a significant increase of force when using Malic Acid, making the hair much more difficult to comb, especially under wet condition. After using HVPPS, the force applied decreased dramatically in wet hair while in dry hair, a less significant decrease was noticed.

Regarding the Extensibility, it was noticed a clear increase when using Malic Acid in both Virgin and Bleached Hair. The Extensibility rose sharply when Malic Acid was used in Bleached Hair. When adding the HVPPS, the effect caused by the Malic Acid decreased.

Virgin Hair extensibility also improved with the use of Malic Acid. Together with HVPPS, the Extensibility was even more significant.

Malic Acid appears to give more strength to the hair fibres both in Virgin and Bleached hair under wet condition. This conclusion is very important has while washing hair, blow drying and styling, damage is caused due to the pulling, stretching and rubbing. Since the use of Malic Acid increases the Extensibility in both hair types, the hair can be more elongated before reaching its breakage point.

In conclusion, the use of Malic Acid in Bleached Hair under wet condition was beneficial. The use of HVPPS made the effect of Malic Acid worse thus they should not be utilised together to reach a better combing experience.

Since bleached hair is damaged, there is loss of strength and hair becomes more difficult to comb, thus, overall, the use of Malic Acid is recommended to be included in formulations specialized for bleached hair has it improve the combability and extensibility.

In summary, the outcomes reached are useful as they help to mitigate problems seen in previous projects. It is concluded that Malic Acid is crucial to restore the electrostatic bonds between proteins lost during bleaching.

More assessments are required to determine if the results obtained are accurate and representative. Tensile testing is necessary to take place with more hair tresses in order to assess if the use of Malic Acid and/or HVPPS will reduce or increase the measurements.

To increase statistical power, the sample size should be increased, thus more tresses per treatment are required as well as more types of hair should be included, as ethnicity plays a crucial role on the hair fibre, changing the combing experience from person to person. Overall, this investigation has revealed some new information on the use of Malic Acid and HVPPS in hair regarding to combability, extensibility and showcased the importance of developing the topic more in the future.

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