Natural molecules to fight Asian hair exposome

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Abstract

Various hair treatments and environmental factors such as ultraviolet and pollution induce hair shaft damage. All these parameters refer to exposome. In this study, we evaluated the effect of a natural complex extract, AC, which associates the antioxidant activity and photo-protective power of *Zingiber officinale* roots and *Magnolia officinalis* bark with the Pracaxi oil (*Pentaclethra macroloba*), an original oil rich in long chain fatty acid to reinforce lipid layer and to refill cuticle gap. Permeation, protein carbonylation, mechanical resistance and hair strengthening and gloss were evaluated in Asian hair. The active ingredient named AC (trade name Zoryalys®) penetrates to the heart of the hair. Thanks to this, it prevents protein oxidation induced by pollution and UV exposure. As a result, the hair structure integrity is protected, inducing strength and resistance of the hair, and finally gloss.

Hair has always been considered as a symbol of beauty. The appearance of hair is one of the main factors that contributes to our sense of attractiveness and can therefore have a strong influence on one's self-esteem and self-confidence. However, every day, hair is exposed to exposome, leading to external and internal damage of the hair shaft and therefore impacting its beauty. This term exposome was first introduced by Wild,[1] who described it as totality of exposures to which an individual is subjected from conception to death. A redefined definition was provided by Miller and Jones,[2] who proposed that the exposome should be considered as a cumulative measure of environmental influences and the subsequent associated biological responses of an individual throughout their life. More precisely, three broad exposome domains, often overlapping with each other are proposed to classify environmental exposures within the exposome. These domains are as follows:

- The general external (wider influential factors, such as social capital, urban-rural environment, and climate);
- The specific external (chemical contaminants, infectious agents, occupation and lifestyle);
- The internal exposome includes internal chemical environments determined by internal processes (e.g. metabolic and inflammatory).

More precisely, stress and health, pollution, nutrition, physical and chemical manipulation, climate and radiation, and microbiota are included in Hair Exposome. Among them, exposure to sunlight and urban pollution are considered as the main unavoidable stressors that impact physicochemical properties of hair and lead to cuticle and cortical damages to the hair fibre affecting its strength and its beauty.[3,4] The lipid layer, mainly composed of covalently bound fatty acids whose major component is 18-MEA (18-methyleicosanoic) and covering the outermost surface hair fibre cuticle, is the most vulnerable to external damages. 18-MEA creates a hydrophobic mantle around the hair fibre surface that acts as a boundary lubricant to reduce friction resistance between hair fibres.[5] With the loss of this natural protection, the hair is more hydrophilic and proteins of the underlying cuticle and then of inner cortex, which are the key component of hair, become more exposed to harmful factors and more prone to be degraded.[6,7] The deleterious effect of exposome reaches the inside of the cuticle, inducing photodegradation of the cysteine groups and peroxidation of the cell membrane complex, composed by fatty acids, cholesterol and ceramides. As the cysteine disulphide bonds are broken by UV light, they are oxidised and thus produce cysteic acid. This increase in cysteic acid is considered to be a major cause of hair integrity damage and reduced mechanical strength. Oxidation of the amide carbon of polypeptide chains also occurs, producing carbonyl groups. It is important to note that protein carbonylation is induced either by the direct impact of ROS or by reactions with secondary products of oxidative stress such as reactive aldehydes produced by lipid peroxidation, forming adducts with side chains of cysteine, histidine and lysine residues.[8,9] As hair fibre cannot be repaired, protecting and maintaining the hair fibre architecture are essential to prevent daily

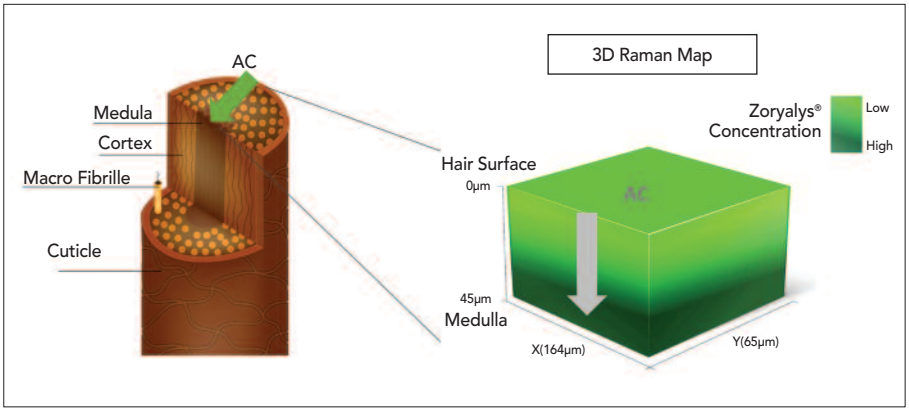

Figure 1: Penetration of the active complex (AC) at the heart of the hair. 3D Raman map illustrating the penetration profile of the active from the cuticle surface of hair fiber (0 µm) to the medulla (45µm) 1-hour after application.

damages and to retain both mechanical and cosmetic hair properties. This protection requires an adequate combination of bio-active components. The use of natural antioxidants from plants such as polyphenols have been shown to protect hair from lipid peroxidation and protein degradation induced by UV exposure.[10] More precisely, shogaol and gingerol (6-gingerol, 8-gingerol, and 10-gingerol), which are the major polyphenols of *Zingiber officinalis* roots, possess high antioxidant activity.[11] Detailed literature survey has pointed out that *Zingiber officinalis* roots described for treatment of many diseases and herbal medicine play a major role in the preparation of many Ayurvedic medicine, the science of life.

Other polyphenols such as honokiol and magnolol, two major components of the genus Magnolia are bioactive constituents of the traditional Chinese medicine that have anti-oxidative properties.[12] As previously mentioned, UV and pollution exposure can induce peroxidation and loss of the hair protective lipid layer. Vegetable oils produced by oleaginous plant species from Amazonia have unique compositions with interesting physicochemical and nutritional properties, including high content of unsaturated fatty acids, especially ω6 and ω9 fatty acids.[13] Among them, Pracaxi oil (*Pentaclethra macroloba*) is notorious for providing natural long chain fatty acids in particular behenic acid (C22), representing about 20% of fatty acid composition. Oleic acid (C18) and lignoceric acid (C24) represent also about 50% and 13% of Pracaxi oil.

After having proposed solutions for skin protection against exposome, an active ingredient was developed to fight against hair exposome, using a biomimetic approach. Aware that both morphological and biochemical differences across ethnic groups exist in hair, and aware that exposure to exposome is important in Asia, a focus on Asian hair was realised in this investigation. Ex vivo tests were carried out on hair fibres with Active complex (AC) formulated at 1%. This AC associates the antioxidant activity and photo-protective power of *Zingiber officinale* roots and *Magnolia officinalis* bark with the Pracaxi oil (*Pentaclethra macroloba*), an original oil rich in long chain fatty acid to reinforce lipid layer and to refill cuticle gap.

Material and methods

Firstly, to ensure potential efficacy of AC in the whole hair fibre, a permeation profile was evaluated. For that, Raman spectroscopy was used. The confocal Raman spectra were collected before and after 1-hour application of the 70 µL of AC 1%, from the surface of hair fibre to 45 µm depth with 2 µm steps (cuticle, cortex and medulla). After sample preparation, the hair fibres were placed into calcium fluoride (CaF2) windows for Raman evaluation. The hair fibres were attached by means of an adhesive tape, preventing their movement during the analysis process. Measurements were made with the laser positioned on the surface and centre of the fibre.

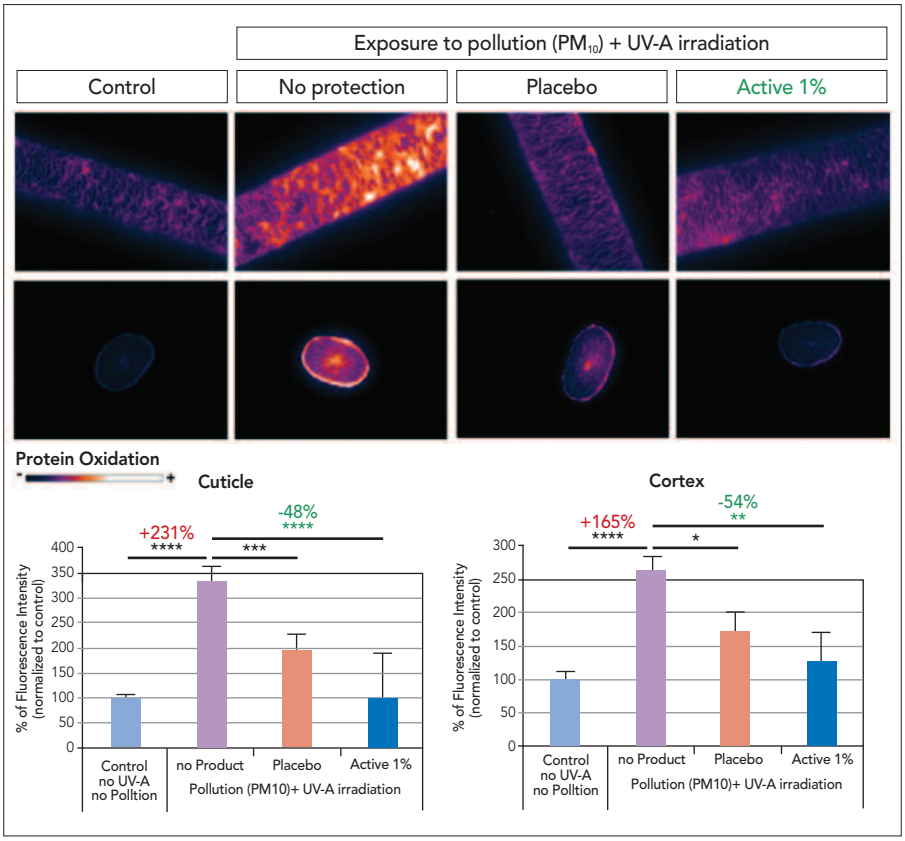

Figure 2: Effect of the active complex (AC) on protein carbonylation induced by pollution and UV-A. Hair shafts were exposed to fine particulate matter (PM10) and UV-A with or without previous 3 day-application of Placebo or AC. In the upper panel: *in situ* visualization of oxidized proteins on Asian hair shaft: lateral and sagittal (cortex and cuticle) views . In lower panels, corresponding quantification of (PM10+UV-A)-induced carbonylation levels in cuticle and cortex compartments. Mean values ± SD are presented.*p<0.05, **p<0.01 ***p<0.001, ****p<0.0001.

Secondly, since hair cannot be repaired once damaged, the protection of hair proteins from carbonylation is an efficient approach for hair protection against daily stresses including urban pollution and UV radiation. Thus, the protective efficacy of AC 1% against hair protein oxidative damage induced by urban pollution and UV-A was assessed through the analysis of protein carbonylation in Asian hair fibres. Natural Asian hair shafts (Dark hair) were incubated with the active complex or a placebo during 3 cycles of 24h followed by a washing step for the first two. Fine particulate matters of ambient air (PM10; Particulate Matter Hazardous Air Pollutants from European Reference Material; Ref. CZ100) were applied over 6 hours on the hair shafts that were concomitantly subjected to UV-A irradiation (Emission peak at 365 nm, total dose: 84 $J.cm^{-2}$). The control hair group was not treated with PM10 and was not irradiated. The UV-A dose of 84 $J.cm^{-2}$ was equivalent to the UV-A fraction received during a day of outdoor exposure (7h.day^{-1}) in summer day. In situ detection of protein carbonylation was performed using fluorescent probe on sagittal sections and on entire hair shafts.

Thirdly, the protective effect of the active complex on hair components was assessed by its capacity to limit UV-induced tryptophan degradation and disulfide bonds breakage.

Light brown natural Asian hair were incubated with the active complex during 3 cycles of 24h followed by a washing step for the first two. Hair tresses were then exposed to the entire spectrum of sunlight, including ultraviolet (UV-A and UV-B), visible light and infrared (IR) using the Q-Sun Xenon Test Chambers with a daylight filter producing a spectrum similar to direct sunlight. The received irradiation were equivalent to 30 days of outdoor exposure (6h.day^{-1}) in summer day.

Tryptophan degradation upon solar exposure was determined via Fluorescence Spectrometry and changes in disulfide bonds were assessed through the measurement of cysteic acid formation using infrared spectroscopy. Finally, specular reflection of hair was measured to evaluate hair gloss.

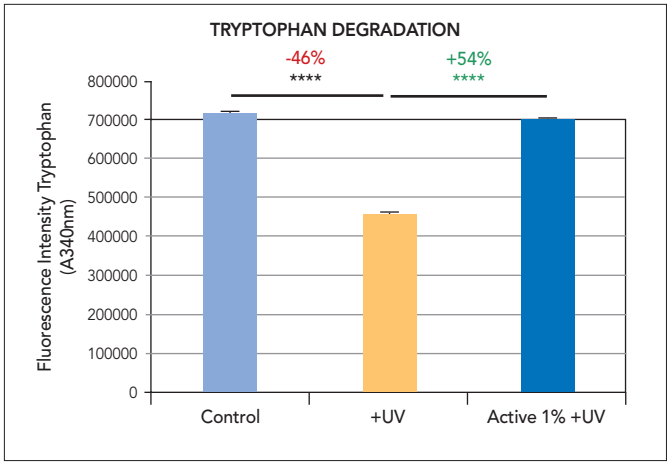

Figure 3: AC protects hair from tryptophan degradation induced by sunlight exposure. Application of the active complexe on Asian hair during 3x24h before full-spectrum sunlight exposure (UV). Mean values ± SD are presented. ****p<0.0001

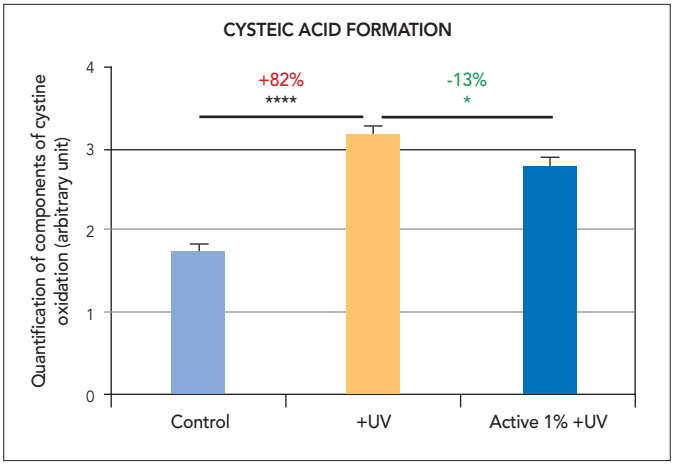

Figure 4: AC limits formation of cysteic acid induced by sunlight exposure. Application of the active complexe on Asian hair during 3x24h before full-spectrum sunlight exposure (UV). Mean values ± SD are presented. *p<0.05, ****p<0.0001

As UV-induced photochemical damage on hair fibre could consequently alter mechanical properties of hair, tensile properties and strength of hair in response to solar exposure (UV-A, UV-B, IR and visible light) were evaluated after AC 1% treatment. Nine tresses of natural light brown Asian hair were prepared, weighing 2.5 g each and 25 cm long. All tresses underwent a standard pre-cleaning process with 10% Sodium lauryl ether sulfate (SLES) solution for 1 minute then rinsed with running water. Tresses were dried in a standardised environment at 55 ± 5% relative humidity and 22 ± 2°C, during 24 hours before tests. Tresses were first submersed in AC 1% for 24 hours (10 g of product.g^{-1} of hair), rinsed 30 seconds and dried in a controlled environment for 10 hours. This process was repeated two times more, completing 3 cycles of application corresponding thus to a chronic application during 3 days. At the third application, tresses were not rinsed. Then, tresses were exposed a first time to the full-spectrum of solar radiation (UV-A, UV-B, IR and visible light), equivalent to about 5.6 days (6h.day^{-1}) of sunlight exposure in Brazil or in summer day in Paris using a sunlight radiation simulator (Sun Xenon Test Chambers, model Xe-1-BC equipped with a 1800W xenon lamp). After the first cycle of radiation, the tresses were washed with SLES 10%, for 60 seconds and then rinsed for 30 seconds. The tresses were dried, in a controlled environment for 10 hours. Then, a new application of AC 1% was made, leaving the tresses submerse in product for 2 hours. The application of AC 1% and exposing cycle were repeated 12 times, totaling 12 applications. During the experiment, the total exposure was equivalent to 67 days of sunlight (6h.day^{-1}) divided in 12 exposures of 5.6 days and between each a shampoo followed by a new 2h-application of AC at 1%. The EMIC instrument, model DL500, equipped with a dynamometer with a 20N load cell was used. Each strand was held by a lower claw and an upper claw connected to the load cell of a dynamometer on the upper part. The following parameter was assessed 1) elongation at break, 2) force at the specific elongation of 20%.

Finally, hair gloss is one of the most important and desired cosmetic attributes[14] that can be altered by too higher sun exposure. The objective of this study was to evaluate the effect of AC 1% on hair gloss after solar exposure (UV-A, UV-B, IR and visible light). Fifteen tresses were prepared of natural light brown Asian hair, weighing 2.5g each and 25 cm long. All tresses underwent a standard pre-cleaning process with 10% Sodium lauryl ether sulfate (SLES) solution for 1 minute then rinsed with running water. The tresses were dried in a standardised environment at 55 ± 5 % relative humidity and 22 ± 2°C, during 24 hours before tests. Tresses were submersed in AC 1% for 24 hours (10 g of product.g^{-1} of hair), rinsed 30 seconds and dried in a controlled environment for 10 hours. This process was repeated two times more, completing 3 cycles of application corresponding thus to a chronic application during 3 days. At the third application, tresses were not rinsed. Tresses were exposed to the full-spectrum of solar radiation (UV-A, UV-B, IR and visible light) using the Q-Sun Xenon Test Chambers, model Xe-1-BC equipped with a 1800W xenon lamp. Exposure was equivalent to 30 days of sunlight exposure (6h.day^{-1}) in summer day. Gloss was evaluated using the glossmeter (BYK Gardner®).

Results

A natural protection down to the heart of hair

As shown by the 3D Raman maps, which illustrates the permeation in a half hair fibre, AC is present from the hair surface (0 µm) to the hair center (45 µm) (Fig 1). The active complex could therefore be effective against environmental stresses by acting in depth, from the cuticle to the medulla of hair fibre.

Protection against protein carbonylation induced by UV and pollution

Application of the active complex before stress exposure significantly decreased the level of protein carbonylation induced by PM10+UV-A in the cuticle and the cortex region (-48 and -54%, respectively). Thus, the active complex provided to Asian hair a protection of 81% on cuticle region and 84% on cortex region against daily hair protein damage induced by urban pollution and UV rays (Fig 2).

Protection against hair component photodamages

Exposure to solar spectrum induced a degradation of tryptophan (-46%) (Fig 3) and higher production of cysteic acid (+21%) (Fig 4). Application of the active complex limited up to 95% tryptophan degradation induced by solar irradiation (Fig 3). In addition to protect from amino acid degradation, the 13% reduction in cysteic acid formation (Fig 4) indicated that the active complex also limits the breakage of disulfide bonds and therefore protects the structure of keratin.

Increase in hair gloss

Tresses of Asian hair exposed to solar spectrum (UV-A, UV-B, IR and visible light, equivalent to 6h.day^{-1} during 30 days) showed significant reduction of hair gloss (-11%, p<0.0001) compared to the non-exposed tresses. Chronic application of AC 1% during 3 days before solar exposure significantly improved hair gloss (+208%, p<0.0001) compared the UV-exposed group.

Increase in mechanical resistance and hair strengthening

Repeated application of the active complex totally restored the level of elongation at break which was reduced by solar exposure in absence of application (Fig 6A). Moreover, despite exposure to solar radiation, the active ingredient strengthened the hair and increased the basal hair force (+29%) (Fig 6B). Thus, by limiting hair protein photodamage, the active complex makes the hair stronger.

Discussion and conclusion

Hair has always been regarded as a symbol of beauty. It has a great importance in the representation of self-esteem of human beings, bringing in innumerable information about the personality of each one . Moreover, maintaining the hair fibre architecture is essential to always having good strength as well as shine and softness. However, everyday day, hair is exposed to exposome, impacting therefore its beauty. It is important to note that exposome has been a consumer concern in Asia but is increasingly becoming a global phenomenon, with a growing number of pollution alerts in big cities around the world making media headlines. In this study, the results have shown that the active ingredient named AC (trade name Zoryalys®) penetrates to the heart of the hair. Thanks to this, it prevents protein oxidation induced by pollution and UV exposure in Asian hair. As a result, the hair structure integrity is protected, inducing strength and resistance of the hair, and also gloss. PC

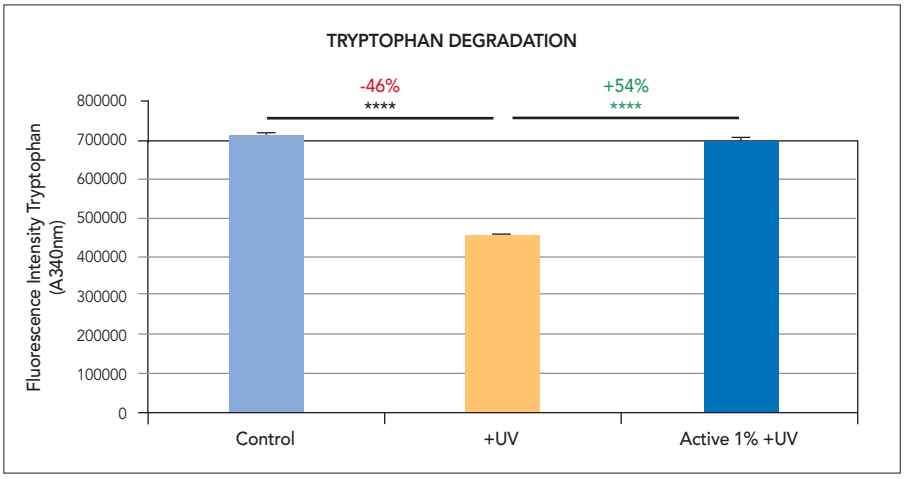

Figure 5: AC increases Asian hair shine. Application of the active complexe on Asian hair during 3x24h before full-spectrum sunlight exposure (UV). Mean values ± SEM are presented. ****p<0.0001.

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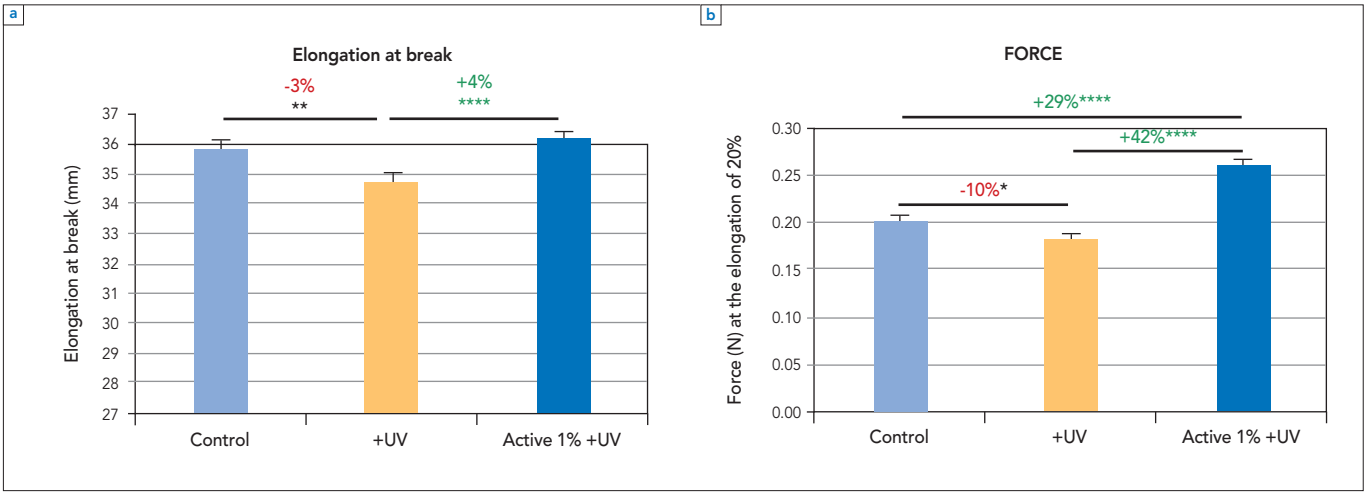

Figure 6: AC increases mechanical resistance and hair strengthening of Asian hair. Elongation of hair at break and hair force at the elongation of 20% were measured after chronic application of the active complex and full-spectrum solar exposure. Mean values ± SEM are presented. *p<0.05, **p<0.01, ****p<0.0001