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Research Article

# Squid as a Model Organism - Part 2: Biomedical and Tissue Engineering Technologies of Skin

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## Abstract

Nature has always been the greatest teacher for humans, and many of our inventions have been inspired by a careful examination of natural phenomena. Even today, despite significant advancements in technology, scientists continue to turn to nature to find solutions to problems and enhance various systems. Discovering the complex mechanisms within the bodies of living organisms has consistently provided a foundation for numerous ideas and innovations. The colossal squid, with its unique characteristics, has recently garnered the attention of many scientists, serving as a source of inspiration for diverse medical and engineering designs. In this study, we will focus on the squid's skin and its camouflage mechanisms, highlighting how humanity has leveraged this remarkable creature for scientific, and engineering progress.

**Keywords:** squid, nature-inspired, biotechnology, biological and engineering technologies

## 1. Introduction

Our planet is home to a remarkable variety of animals, and among them, countless specimens exhibit astonishing colors and patterns, spanning both vertebrates and invertebrates. While in some animals, color changes serve as a means of concealment and survival, in others, these diverse hues play specific roles in signaling, and some might even serve both functions concurrently. The colors displayed on the bodies of animals are produced by specialized structures that transform colorless cells into a spectrum of different hues by accumulating various pigments. In the world of cephalopods, particularly the squid family, the ability to change their skin color and pattern is a valuable trait. This feature aids in their camouflage, a significant advantage for many cephalopods, even some of which are color-blind.

The colossal squid, with its immense size and unique color-changing capabilities, has been the focus of scientific interest due to its remarkable mechanisms for rapid skin color transformation and concealment. Using direct neural control systems, the colossal squid boasts the fastest concealment abilities among all living creatures [1]. By manipulating the absorption, transmission, and reflection of light on its skin's surface, the colossal squid can swiftly change its color in a matter of seconds to match its surrounding environment. Each pigment within the skin reflects a particular wavelength of light, allowing the colossal squid to reflect the precise wavelengths of white light and exhibit the corresponding colors. Investigating these mechanisms may have various applications in engineering and improving human life.

The color change and skin patterns of the colossal squid result from the presence of numerous specialized skin cells. The skin of the colossal squid's body contains three types of cells, each with unique features. These cells are known as chromatophores, iridophores, and leucophores[2], and they are distributed in three separate layers within the skin, as schematically illustrated in Figure **1**.

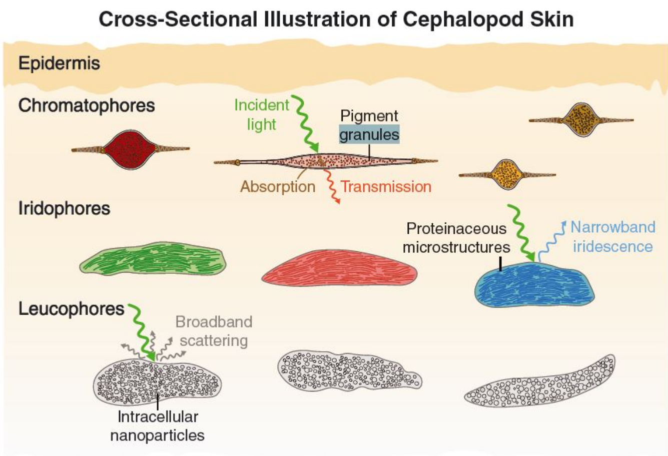


**Figure 1**: Three-layered cellular structure in the colossal squid's skin. The skin of the colossal squid's body consists of three types of cells, namely chromatophores, iridophores, and leucophores, distributed within three distinct layers [2].

## 2. Dynamic Control of Chromatophores in Colossal Squid Skin

The chromatophores are found in the outermost layer of the skin and contain sacs filled with red, yellow/orange, and brown/black pigments, depending on the species of the colossal squid. These pigment-containing sacs enable cephalopods to transform white light into various colors, such as red, blue, green, and more, depending on the type

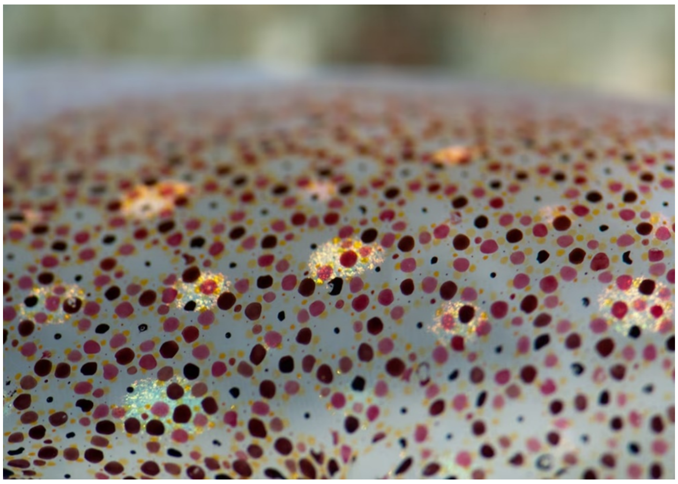

**Figure 2**: A view of the colossal squid's skin. The image shows a regular color pattern created by dark chromatophores and bright iridophores [5].

of pigment. These pigment-containing cells allow light at specific wavelengths to pass through while absorbing other wavelengths, effectively creating the color changes in the squid's skin [2].

One fascinating characteristic of chromatophore cells is that each one is surrounded by dozens of radial muscle cells directly controlled by the central nervous system. Consequently, these muscle cells, which connect to individual chromatophore cells, allow them to expand or contract in a coordinated manner. The mechanical action of radial muscle cells on chromatophore cells results in the expansion and contraction of chromatophore cells, leading to the reflection and transmission of specific visible light wavelengths [3]. Notably, the skin cells of cephalopods, including chromatophores, are regulated by nerve signals, environmental cues, and other factors.

Furthermore, the chromatophores can vary in the number of radial muscle cells surrounding them, thus influencing the extent to which their pigment sacs expand or contract. This variation results in a wide range of color changes and patterns that colossal squids can exhibit, enhancing their camouflage abilities [4]. Each chromatophore can have between 10 and 30 radial muscle cells, all of which are controlled by the central nervous system. The expansion and contraction of these muscle cells allow the chromatophore cells to change from a compact, nearly spherical state to an expanded, thin, and colorful disc shape in less than a second. The change in the diameter of chromatophore cells depends on the type and species of the cephalopod [5], as shown in Figure **3**.

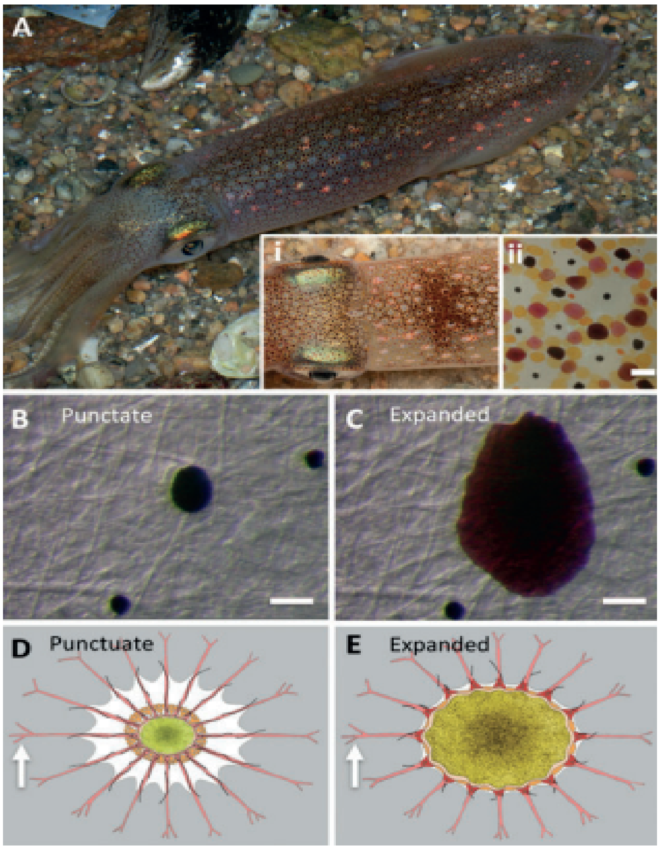


**Figure 3**: Expansion and contraction of chromatophore cells. Chromatophore cells are specialized skin cells found in some fish. These cells are responsible for producing and storing pigments that result in skin color changes. The expansion and contraction of chromatophore cells have a direct relationship with the color change of the fish. In the contracted state, the chromatophore cells are closely packed, and pigments are concentrated inside the cells, resulting in darker or black skin color. In the expanded state, the chromatophore cells move apart, dispersing pigments within the cells, leading to lighter and more colorful skin. Changes in the expansion and contraction of chromatophore cells can occur due to neural signals, variations in stress levels, environmental shifts, and other factors, and these variations serve cephalopods in resistance, body temperature regulation, attracting attention, and social interaction [6].

## 3. Iridophores: The Science of Iridescent Light Reflection in Squid

The iridophores are specialized cells residing in the second layer of the skin. They possess the ability to reflect incident white light into iridescent colors due to their microprotein-based structures. This phenomenon follows the principles of Bragg's law. Iridophores are colorless cells with varying sizes, typically having diameters smaller than 1 millimeter [7,8]. They feature stacks of thin plates that exhibit light-reflecting properties. The reflected light, generally displaying iridescent colors, results from two prerequisites: (i) differences in refractive indices between the plates and the separating spaces and (ii) varying thicknesses of these layers, leading to the phenomenon of color reflection.

The mechanism behind this iridescent light reflection closely resembles what we observe in soap bubbles. Soap bubbles consist of multiple layers, where shorter wavelengths such as blue light are predominantly reflected when illuminated. Conversely, thicker layers tend to reflect longer and thicker wavelengths like yellow and red [9].

Furthermore, multi-layer reflectors exhibit distinct optical characteristics, with the most prominent being their sensitivity to changes in the angle of incidence, affecting brightness and color observed in the reflected light spectrum. In essence, a greater inclination of the light angle results in shorter wavelengths in the reflected light. For instance, the light reflected from multi-layered plates, appearing red when viewed directly, displays various colors like yellow, green, and blue at different viewing angles. Additionally, in the vicinity of the Brewster angle (the angle at which all incident light passes through a surface without reflection), incident light becomes strongly polarized. This intriguing feature may play a significant role in the behavioral performance of iridescent organisms, such as squid, which have the ability to detect polarized light. Squid, depending on their environment, can change their iridescent colors. Behaviorally, especially in aggressive interactions like attack and defense, they demonstrate the ability to alter their iridescent colors [10].

In some cases, iridophores are actively controlled by a unique nonsynaptic cholinergic system. Scientists demonstrated that localized application of acetylcholine, with the effect on muscarinic cholinergic receptors, can alter the iridescent colors of iridophores [10,11]. Unlike chromatophores, which change color within fractions of a second, iridophores exhibit more prolonged color-changing processes. For example, in the case of the *Doryteuthis pealeii* squid, it takes approximately 2 minutes from the application of acetylcholine until the response, causing a shift from red to orange, with a wavelength displacement of about 100 nanometers [12].

Interestingly, in the throat region of the *D. pealeii* squid, iridophores sensitive to near-infrared light (approximately 800 nanometers) have been identified. However, it is still unclear how this wavelength affects the creature's lifestyle. Overall, the phenomenon of iridophore-based iridescent color change is attributed to the existence of specialized protein-based structures in these cells, particularly the proteins known as reflectins. Reflectin proteins are capable of altering refractive indices when they undergo conformational changes, which, coupled with changes in layer thickness, explain the observed variations in light reflection.

The light patterns utilized by squid for camouflage and signaling rely on a complex network of interactions between the contrasting and luminance levels generated by the activities of chromatophores, iridophores, and skin layers. Additionally, in some types of squid, other cells known as leucophores, or white cells, exist in the same skin layer as iridophores. These leucophores play a role in camouflage. They contain spherical nanostructures called leucosomes, which efficiently reflect the surrounding ambient light [10, 13]. These cells effectively match their appearance to the ambient lighting conditions, appearing white in white light, red in red light, and blue in blue light. Leucophores are active within a range of 300 to 900 nanometers, acting as ideal diffusers of light, as they appear equally bright from all viewing angles. Leucophores consist of nano-sized particles, typically ranging in diameter from 250 to 1250 nanometers. They are composed of highly sulfated mucoproteins and weakly acidic mucopolysaccharides [10, 14].

The colossal squid, utilizing these unique features in their chromatophores, iridophores, and leucophores, rapidly adapt their patterns and colors to suit their needs, whether for concealment or signaling. This complex system of light manipulation has inspired scientists in the fields of biological sciences, medicine, and engineering to design various structures and technologies aimed at improving human life [13].

## 4. Bio-inspired Smart Thermal Management Systems: Bridging the Gap Between Passive and Active Heating Technologies

Many modern technologies are focused on precise temperature management and heat transfer in various applications such as electronic circuits, aerospace systems, medical equipment, heating devices, power generation platforms, transportation, packaging materials, textiles, smart environmental control systems, and more. Smart temperature control presents an exciting opportunity for significantly reducing energy consumption worldwide. To date, thermal management systems have been designed in a wide range of applications, adopting either passive (static) or active (dynamic) configurations. Specifically, passive heating technologies utilize materials like insulation, textiles, and reflective coatings with low thermal conductivity that reflect infrared radiation. These systems are cost-effective and contribute to energy savings but lack responsiveness to changing temperature conditions.

On the other hand, active heating technologies, such as electrothermal systems, regulate ambient or device temperature by controlling the flow of heat through electrical and/or mechanical inputs and outputs. These systems offer dynamic temperature control, allowing users to adjust temperature settings easily [1]. However, they tend to be relatively costly and may not be energy-efficient in specific thermal conditions.

Therefore, it is highly desirable to develop an ideal thermal management platform that combines the benefits of passive systems (e.g., cost-effectiveness, simplicity, and energy efficiency) with the dynamic control capabilities of active systems [15]. Within the realm of passive thermal management, space blankets have been introduced by NASA in the 1960s to reduce energy consumption. In their standard configuration, space blankets consist of a plastic sheet (e.g., polyethylene terephthalate) and a continuous thin metallic layer (e.g., aluminum), an architecture that effectively reflects infrared radiation. The concept of space blankets has also found applications in food packaging, emergency shelters, clinical devices, and clothing. Nevertheless, the applicability of space blankets is limited due to their static thermal properties. If responsive stimuli, such as metallic oxides or conductive polymers, were incorporated into these materials (e.g., in the form of thin films within a gap or junction), the development of energy-storing solutions could become achievable [16].

Dynamic color change in the skin of animals, such as squid, offers an intriguing opportunity for achieving optimal energy consumption. The skin of squid comprises multiple layers, with the top layer, in particular, containing specialized chromatophores responsible for red, yellow, and brown coloration. These chromatophores consist of a central pigment cell encircled by muscle nerve cells. This evolved architecture allows pigment cells to be dynamically contracted and expanded by surrounding muscle nerve cells, thereby enabling dynamic switching [8]. Based on this natural mechanism, one can engineer unconventional ways to modulate local light transmission through the skin, thereby controlling color changes and appearance.

In this context, the natural squid skin and its compounds possess all the necessary features, including biomimetic soft active surfaces, optoelectronic displays, electro-mechanochemical materials, stretchable elastomers, electroluminescent materials, and adaptive infrared concealment technologies. These materials can be the foundation for developing, for instance, wearable temperature-regulating devices. Therefore, the ability of squid skin in thermal regulation, which relies on mechanical adjustments, can lead to the creation of an optimized alternative [17].

Metals and metal oxides that reflect or absorb infrared radiation, such as titanium dioxide, thermally transparent elastomers, ethylene derivatives, or other materials, can be considered as potential candidates. These materials should be responsive to stimuli and capable of regulating heat transfer by reflecting or absorbing infrared heat [18].

As illustrated in Figure **4**, a wearable sleeve, inspired by the mechanical temperature regulation of squid skin, has been designed. This sleeve exhibits the capability to reflect or absorb infrared radiation and dynamically control the passage of heat waves. The control mechanism relies on the modification of the spacing between surfaces under the application of mechanical tension. When the wearer's skin temperature is high, heat (infrared radiation) can pass through the gaps, and the temperature is regulated. In cold conditions, the continuous metallic surface prevents heat from escaping, and the infrared radiation is reflected back to the skin. This smart thermal system, inspired by squid skin's dynamic temperature regulation, effectively adapts to the user's needs, maintaining a controlled temperature dynamically.

In conclusion, leveraging the dynamic color-changing abilities of squid skin inspires the development of innovative thermal management solutions. By introducing responsive materials to passive systems, we have the potential to create efficient and dynamically controlled thermal regulation devices that contribute to energy savings while ensuring personalized comfort [3,18].

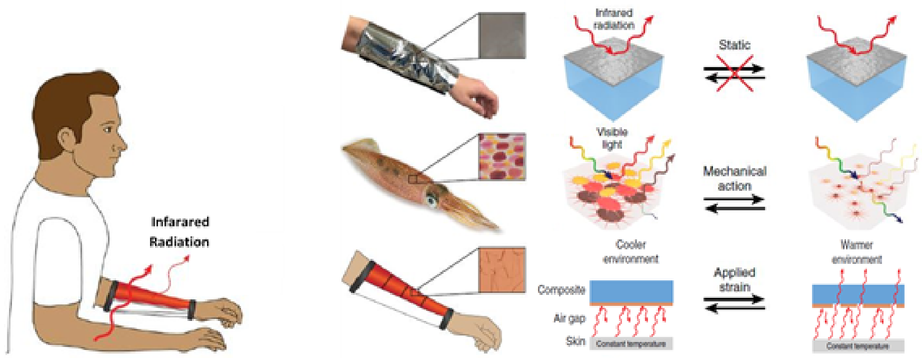


**Figure 4**: Fabrication of a dynamic heat-passing controller inspired by squid skin. Similar to how squid skin allows the passage of specific wavelengths, the heat transfer (infrared radiation) through this design is determined by the application of mechanical forces from the surface [18].

## 5. Bio-inspired Light Control: From Squid Skin to Smart Glass and Human Cells

Some colossal squid species can render parts of their bodies invisible by controlling the transparency or opaqueness, primarily by regulating light absorption or transmission through their skin. The cells in these colossal squid possess a unique protein called "reflectin," which has the capability to reflect light. The intracellular configuration of these proteins dictates the extent to which light is transmitted or reflected, thereby defining the visibility or invisibility of the respective body regions [4].

Drawing inspiration from this phenomenon, engineers have designed glass panels that can be made transparent or opaque using an external driver. These glass panels are constructed with regular arrays of magnetic particles within conical structures. Under normal conditions, the magnetic particles obstruct light transmission, resulting in the glass appearing opaque. Interestingly, when a magnetic field is applied, the particles accumulate at the tips of the conical structures, allowing light to easily pass through the glass, rendering it transparent. In essence, this design enables remote control over the glass's transparency and opaqueness [5].

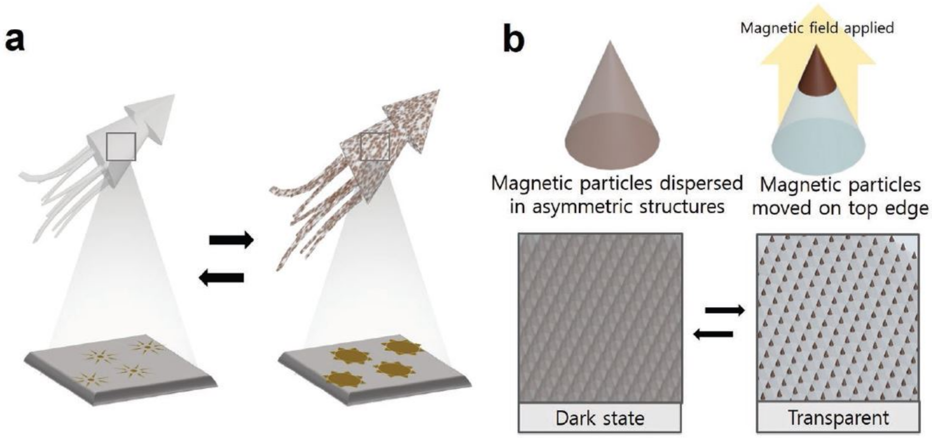


**Figure 5**: Creation of smart glass with controllable opacity and transparency inspired by squid skin. Magnetic particles within the conical structures in the glass respond to a magnetic field, concentrating at the cone tips, facilitating light transmission through the glass [5].

Scientists, inspired by the reflective ability of reflectin protein in squid, envisioned activating this protein within human cells to render humans with less visibility. To achieve this, in a cell culture environment, they introduced the gene responsible for reflectin protein using viral vectors into target human cells to produce a similar protein responsible for light passage and reflection within the squid skin. The results of these experiments indicated that these modified human cells possessed reflectin protein, enabling them to control the passage of light. Subsequently, for demonstrating the intelligence of cells containing reflectin protein in controlling light passage, a chemical stimulus, such as sodium chloride, was employed. Sodium chloride, through its influence on reflectin protein, altered the configuration of these proteins within the cells, thereby allowing control over light passage and transparency [4].

## 6. Conclusion

Squid skin has gained significant attention as an inspiration for various biotechnological and engineering applications. The unique features of squid skin, including its color-changing ability, light reflectance properties, and capability to generate electricity, motivate researchers to develop new and innovative technologies. Moreover, with inspiration from the astounding attributes of squid, it is likely that in the near future, there

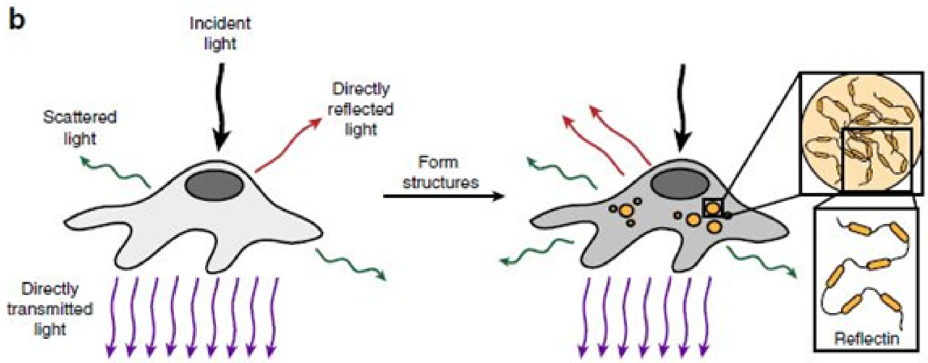


**Figure 6**: Functioning of reflectin proteins in human cells for smart light control. research demonstrates the capacity of these proteins to control light, including color changes and inducing light alterations in response to external stimuli like pressure, temperature, and electricity [4].

will be a substantial transformation in the design and advancement of technologies in display screens, sensors, and other devices. Consequently, squid skin serves as a wellspring of inspiration for the development of biotechnological and engineering innovations, contributing to the creation of intelligent materials, advanced coatings, and novel technologies.

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