

Vasil Georgiev^{1,2} Anton Slavov²Ivelina Vasileva²Atanas Pavlov^{1,3}

Review

Plant cell culture as emerging technology for production of active cosmetic ingredients

¹Laboratory of Applied Biotechnology - Plovdiv, The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, Plovdiv, Bulgaria

²Department of Organic Chemistry and Inorganic Chemistry, University of Food Technologies, Plovdiv, Bulgaria

³Department of Analytical Chemistry and Physicochemistry, University of Food Technologies, Plovdiv, Bulgaria

Plants have always been the main source for active cosmetic ingredients, having proven health beneficial effects on human, such as anti-aging, antioxidant, anti-inflammatory, UV-protective, anti-cancer, anti-wrinkle, skin soothing, whitening, moisturizing, etc. Extracts from herbal, aromatic and/or medicinal plants have been widely used as effective active ingredients in cosmeceuticals or nutricosmetics, especially in products for topical application and skin-care formulations. However, over the past decade, there has been an increasing interest to plant cell culture – derived active cosmetic ingredients. These are “new generation” of high quality natural products, produced by the modern plant biotechnology methods, which usually showed stronger activities than the plant extracts obtained by the classical methods. In this review, the advantages and the current progress in plant cell culture technology for the production of active cosmetic ingredients have been summarized, and discussed in details within a presented case study for calendula stem cell product development.

Keywords: *Calendula officinalis* L. / Cosmeceuticals / Nutricosmetics / Plant cells / Plant stem cells / Polysaccharides / Tissues and organ cultures

Received: April 4, 2018; accepted: June 20, 2018

DOI: 10.1002/elsc.201800066

1 Introduction

Plants are the oldest source of natural compounds with medicinal and cosmetic properties explored by the mankind. Even nowadays, between 70 and 80% of the people worldwide rely on traditional herbal medicine to cover their basic healthcare needs [1]. In fact, 11% of the existed essential drugs used in modern human medicine are still with plants origin [2]. The percentage of plant-derived natural products, applied as active ingredients in cosmetics, is much higher. In accordance with the continuous increase of world population, the global demand for natural plant ingredients is expected to continue its exponential growth [1]. As result, we are already witnesses of mass overexploitation of natural habitats of some herbal and aromatic plants, placing them under threat or even leading to their extinction [3]. This negative tendency is especially dangerous for medicinal plants, which in most cases are rare and endemic species. These species often grew under extreme climate, unique soil composition or specific latitude. Their populations usually have been characterized with low

levels of genetic diversity, which makes these species extremely vulnerable to genetic erosion and decrease their chance to survive in case of environmental pollution [4]. Moreover, plant derived active ingredients are usually very complex mix of bioactive molecules, which cannot be easily replaced by chemically synthesized analogs. To solve the above problems, plant biotechnology has provided the tool, which can secure more eco-friendly and sustainable supply of valuable phytochemicals with health-beneficial properties, to cover the growing demands of cosmetic industry. Over the last decade, the advantages of plant cells and tissues culture technologies have been widely explored in development of highly-efficient platforms for more rapid production of pharmaceutically important molecules of plant origin or for heterologous expression of therapeutic proteins [5]. Nowadays, there are many cosmetic products, including both cosmeceuticals and nutricosmetics, which have active ingredients, derived by plant cell culture technology. The aim of this paper is to review the recent progress in plant cell technology for cosmetic application and to provide short overview on commercialized plant cell culture – derived active cosmetic ingredients available on the market. The information in this review was retrieved by an exploratory electronic survey of plant tissue culture - derived cosmetic ingredients and their technical specification datasheets, conducted by search in the technical websites of the major suppliers such as Prospecor[®] search engine, SpecialChem, Cosmetic Design Europe, the academic search engines Scopus,

Correspondence: Dr. Vasil Georgiev (vasgeorgiev@gmail.com), Laboratory of Applied Biotechnology - Plovdiv, The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, 139 “Ruski” Blvd., 4000 Plovdiv, Bulgaria

Abbreviations: CMC, cambial meristematic cells; EGF, Epidermal Growth Factors

ScienceDirect, Google Scholar, and PubMed, and the popular search engine Google. In addition, the main steps in development and characterization of plant cell culture – derived active ingredients are closely discussed in the presented case study for development of InnoVStemCell Calendula product.

2 Plant extracts as active ingredients in cosmetics

Plants are rich source of countless metabolites with potential application in cosmetic products. Since time immemorial, the mankind has used different plants and their extracts to create cosmetics with the aim to establishing a state of eternal youth [6]. Nowadays, the plant extracts are becoming the most popular active ingredients of cosmetics due to the ever-increasing demand for natural compounds which in addition to esthetic looks can provide additional health benefits. Development of such products, known as “cosmeceuticals”, reflects a most recent trend in the modern cosmetics and personal care industries. Indeed, plants are rich in endogenous bioactive metabolites with potential cosmetic and pharmaceutical applications [7–9]. Most of these phytochemicals, such as polyphenols, phenolic acids, triterpenes, flavonoids, stilbenes, steroids, carotenoids, steroidal saponins, sterols, fatty acids, polysaccharides, sugars, peptides, etc., could be extracted with appropriate solvents and used as active ingredients present in cosmetic formulations [10]. Because of this, huge numbers of plant sources have been explored by the cosmetics industry in search of innovative active ingredients which combine some specific pharmacological properties, such as antioxidant, antimicrobial, antiviral, anti-cancer, antifungal, anti-inflammatory, anti-allergy, etc., and also showing strong moisturizing, anti-ageing, anti-wrinkle and UV protective effects [11]. However, the quality and phytochemical profiles of plant extracts varied in wide range, depending on climate, soil, latitude, seasonal factors, time of harvest, and the field management practice, which could be a challenge to standardize their activities [12]. The search for novel natural phytochemicals has led to the gathering bioactive extracts not only by plants, but also from mushrooms, algae, and also, by utilization of by-products of plant origins [13–17]. Nowadays, with the growth in consumers’ interest to effective and safe natural products, a “new generation” of high quality bioactive phytochemicals, produced by the plant cell culture technology, have been introduced in the past decade, and now, their presence in the cosmeceuticals market have been steadily on the rise.

3 Plant cell culture technology: principles for production of active cosmetic ingredients

Plant cell culture technology is a technique for growing of plant cells under strictly controlled environmental conditions. Because plant cells are considered totipotent, they have the potential to express the full genetic machinery coded in the nucleus, and thus, they are able to produce the full spectrum of characteristic

secondary metabolites, found in mother plants. Plant cells are amenable to good manufacturing practice procedures and can be easily propagated by using large volume bioreactors independently on climate or soil or field management practices [5, 18]. Moreover, in vitro cultured plant cells are characterized with fast growth, and the ability to accumulate large amount of uniform biomass for a short period of time [19, 20]. This is very important advantage especially for the production of rare bioactive compounds, as resveratrol, paclitaxel or terpenoids, which are usually found in low concentrations in plants and their isolation and purification requires the processing of large amounts plant biomass [21–25]. Additionally, plant cell culture technology offers a reliable and powerful production platform for continuous supply of contamination-free, phytochemically uniform biomass from herbal, aromatic, medicinal, and even from rare and threatened plant species [26]. The perspective to obtain natural phytochemicals by using an environmentally sustainable biosynthetic platform made plant cell culture technique exceptionally attractive for the production of active ingredients for high added values “green” cosmetic formulations [27–30]. It should be noted, that active cosmetic ingredients, obtained by the plant cell culture technology are popularized among the customers under the name “plant stem cells”. Here it is important to understand that the term “stem cells” used in this phrase is not always referred to real plant stem cells. The most of the existed plant cell culture production platforms are in fact developed on the basis of the use of dedifferentiated plant cells rather than on the culturing of the true plant stem cells. Dedifferentiated plant cells are obtained by dedifferentiation of already differentiated mature plant cells from different specialized tissues, whereas the true plant stem cells should be never differentiated in their life cycle. Because raised from the dedifferentiation of differentiated cells, the dedifferentiated plant cell cultures could inherit some epigenetic modifications, characteristic for the type of the tissues they have obtained from, and thus, they could be very heterogeneous in their biosynthetic and growth properties. This fact made it possible to generate almost unlimited numbers of plant cell lines with unique phytochemical profiles and growth characteristics even from the same plant, used for their initiation. For this reason, the terms as “leaf stem cells”, “meristems stem cells”, “root stem cells” “rhizome stem cells”, “flower stem cells”, “fruit stem cells”, etc. could be often seen in INCI names of active cosmetic ingredients, but in fact all of these refers to dedifferentiated plant cell cultures. On the other hand, there are plant cell technologies existed, which are developed on the basis of the cultivation of true plant stem cells – the cambial meristematic cells (CMC). These cells are isolated by cambial layer and consist only of true meristem stem cells [31, 32]. Cambial meristematic cells are characterized with fast and uniform growth, lack of epigenetic modifications and ability to produce predictable yields of secondary metabolites when treated with stimulating factors such as elicitors [33]. This technology have been used by the Korean company “Unhwa Corp.” to produce several active cosmetic ingredients, by using the patent protected expression platform Ddobyul[®], developed on the basis of propagation of cambial meristematic cells (Table 1). Another important fact, concerning the plant cells used for cosmetics, is that the term “plant stem cells” is often equally used for active ingredients, produced by either callus cultures, cell suspensions or hairy roots. It is

Table 1. Some of the most popular plant cell culture technology – derived active cosmetic ingredients, currently available on the market

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Phyto-Biotics Perilla®	<i>Perilla frutescens</i>	cell suspension extract	anti-aging, antimicrobial, soothing effect	Active Concepts LLC; USA
Phyto-Biotics Quercus®	<i>Quercus alba</i>	meristem stem cells extract	antioxidant, soothing effect, antimicrobial, anti-aging	Active Concepts LLC; USA
Phyto-Biotics Acai®	<i>Euterpe oleracea</i>	cell suspension extract	antioxidant, anti-aging, anti-wrinkle, soothing effect, moisturizer	Active Concepts LLC; USA
AKOSKY® APIUM	<i>Apium graveolens</i>	callus culture extract	skin regeneration	Akott Evolution S.R.L.; Italy
VITADENIA®	<i>Gardenia taitensis</i>	callus culture extract	anti-aging, anti-wrinkle, regenerative, repairing	Biocosmethic; France
UrbanEthic®	<i>Gossypium herbaceum</i>	stem cell extract	anti-photo-aging, protection against atmospheric pollutants and heavy metals, protection from oxidative stress	Biocosmethic; France
NatureCells Hydragenesis	<i>Vitis vinifera</i> cv. Verdejo	liposomal complex of cell suspension extract	antioxidant, moisturizer, hydrating effect	Infinitec; Spain
NatureCells Antiaging	<i>Vitis vinifera</i> cv. Mencía	liposomal complex of cell suspension extract	anti-aging, antioxidant, anti-wrinkle	Infinitec; Spain
NatureCells Anti Stretch Marks	<i>Centella asiatica</i>	liposomal complex of cell suspension extract	skin-firming, anti-stretch mark effect, anti-inflammatory, anti-cellulite	Infinitec; Spain
InnovaStemCell Calendula W	<i>Calendula officinalis</i>	cell suspension extract	regeneration, moisturizer	Innova BM; Bulgaria
InnovaStemCell Calendula EM	<i>Calendula officinalis</i>	emulsified cell suspension	anti-wrinkle, skin regeneration, deep hydration	Innova BM; Bulgaria
InnovaStemCell Rosa damascena W	<i>Rosa damascena</i>	cell suspension extract	skin regeneration, anti-aging, antioxidant	Innova BM; Bulgaria
InnovaStemCell Rosa rugosa W	<i>Rosa rugosa</i>	cell suspension extract	anti-inflammatory antioxidant, antimicrobial, UV protective	Innova BM; Bulgaria
Plant C-Stem™ Vigna Radiata	<i>Phaseolus radiatus</i>	meristem cell culture vacuole extract	anti-aging, anti-wrinkle, protection/repair of environmental damage	Innovacos Corp.; USA
Roseroot (Rhodiola rosea L.) Plant Stem Cell Extract	<i>Rhodiola rosea</i>	callus culture extract	antioxidant	In vitro Plant-tech AB; Sweden

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Milk thistle (<i>Silybum marianum</i> L.) plant stem cell extract	<i>Silybum marianum</i>	callus culture extract	antioxidant	In vitro Plant-tech AB; Sweden
Greater plantain (<i>Plantago major</i> L.) plant stem cell extract	<i>Plantago major</i>	callus culture extract	antioxidant	In vitro Plant-tech AB; Sweden
PhytoCellTec™ Malus Domestica	<i>Malus domestica</i>	liposomal complex of cell suspension extract	anti-wrinkle	Mibelle AG Biochemistry; Switzerland
PhytoCellTec™ Solar Vitis	<i>Vitis vinifera</i> cv. Gamay Teinturier Fréaux	cell suspension extract	anti-photo-aging, extending skin vitality	Mibelle AG Biochemistry; Switzerland
PhytoCellTec™ Alp Rose	<i>Rhododendron ferrugineum</i>	cell suspension extract	protecting longevity, increasing skin vitality, protection/repair of environmental damage	Mibelle AG Biochemistry; Switzerland
PhytoCellTec™ Argan	<i>Argania spinosa</i>	cell suspension extract	improve activity of human dermal stem cells, anti-wrinkle	Mibelle AG Biochemistry; Switzerland
PhytoCellTec™ Symphytum	<i>Symphytum officinale</i>	cell suspension extract	increase skin renewal, rejuvenates skin epidermis	Mibelle AG Biochemistry; Switzerland
PhytoCellTec™ nunatak®	<i>Saponaria pumila</i>	cell suspension extract	increase skin elasticity, firmness and density, protection/repair of environmental damage, anti-aging	Mibelle AG Biochemistry; Switzerland
RootBioTec HO	<i>Ocimum basilicum</i>	hairy root culture extract	reduce hair loss, inhibit 5 α reductase activity, stimulate dermal papilla cells	Mibelle AG Biochemistry; Switzerland
EGF (Epidermal Growth Factor);	<i>Oryza sativa</i>	recombinant proteins expressed by rice cell culture	restore reduced activity with age, stimulate epidermal skin cells, anti-aging, anti-wrinkle, wound healing, stimulation on collagen synthesis in aged skin	Natural Bio-Materials (NBM) Inc.; Korea

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
FGFb (basic Fibroblast Growth Factor)	<i>Oryza sativa</i>	recombinant proteins expressed by rice cell culture	anti-aging, anti-wrinkle, anti-hair loss, hair growth	Natural Bio-Materials (NBM) Inc.; Korea
IGF-1 (Insulin-like Growth Factor-1)	<i>Oryza sativa</i>	recombinant proteins expressed by rice cell culture	growth support for skin cells with EGF, support young and healthy skin	Natural Bio-Materials (NBM) Inc.; Korea
KGF (Keratinocyte Growth Factor; Fibroblast Growth Factor-7)	<i>Oryza sativa</i>	recombinant proteins expressed by rice cell culture	specific growth factor for keratinocyte	Natural Bio-Materials (NBM) Inc.; Korea
VEGF165 (Vascular Endothelial Growth Factor)	<i>Oryza sativa</i>	recombinant proteins expressed by rice cell culture	skin cell growth and support, wrinkle and skin aging improvement, wound healing after laser resurfacing or injury, hair cell growth and support, hair cycle regulation and anti-hair loss	Natural Bio-Materials (NBM) Inc.; Korea
Foresseen Shield Nopal	<i>Opuntia ficus indica</i>	powdered leaf cells	antioxidant, anti-wrinkle, anti-aging, UV protective, decrease melanine spots	Naolys, France
Initial E [PT+TMG]	<i>Polygonia tuberosa</i>	callus culture extract	anti-aging, anti-wrinkle, anti-circle,	
anti-puffines, brightening, firming, soothing, moisturizing	Naolys, France			
Power Extension [HSB+R]	<i>Hibiscus syriacus</i>	leaf cell extract with rutin	anti-aging, anti-wrinkle, antioxidant	Naolys, France
All Even Sweet Iris	<i>Iris pallida</i>	glycerin based leaf cell extract	anti-aging, anti-wrinkle, firming	Naolys, France
All Fiber Booster Olive tree	<i>Olea europea</i>	glycerin based leaf cell extract	anti-aging, restructuring	Naolys, France
All Fiber Booster Chinese hibiscus	<i>Hibiscus rosa sinensis</i>	glycerin based leaf cell extract	anti-aging, restructuring	Naolys, France
All Fiber Booster Green tea	<i>Camellia sinensis</i>	glycerin based leaf cell extract	anti-aging, firming, regenerating, softening, restructuring	Naolys, France
Fiber Booster Plus Sequoia and Vitis flower	<i>Sequoia sempervirens</i> and <i>Vitis vinifera</i>	glycerin based leaf and flower cells extract	anti-aging, antioxidant	Naolys, France

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Inside Heart Egyptian blue lily Revive Commiphora and Rose from Damas	<i>Nymphaea caerulea</i> <i>Commiphora myrrha</i> and <i>Rosa damascena</i>	glycerin based leaf and flower cells extract glycerin based leaf cells extract	anti-aging, antioxidant anti-aging, energizing	Naolys, France Naolys, France
StandStill Rose from Damas New ReGeneration Cocoa	<i>Rosa damascena</i> <i>Theobroma cacao</i>	glycerin based leaf cells extract glycerin based leaf cells extract	anti-aging, firming anti-aging, firming, regenerating, protective, restructuring anti-aging, regenerating	Naolys, France Naolys, France
Total Generation Sequoia and Egyptian blue lily	<i>Sequoia sempervirens</i> and <i>Nymphaea caerulea</i>	glycerin based leaf cells extract	anti-aging, regenerating	Naolys, France
Total Generation Curry plant Inside Light Poet's narcissus Bright Light Madonna lily LightWaves Defense [JS+M]	<i>Helicrysum italicum</i> <i>Narcissus poeticus</i> <i>Lilium candidum</i> <i>Jasminum sambac</i>	glycerin based leaf cells extract callus culture extract powdered leaf cells sunflower oil based leaf cells extract	anti-aging, regenerating brightening, lightening and anti-blemish, anti-aging brightening, regenerating protective, anti-aging, radiance, firming, anti-redness	Naolys, France Naolys, France Naolys, France Naolys, France
Global Protect Common juniper OxyRelax California poppy OxyRelax Cherry tree	<i>Juniper communis</i> <i>Eschscholzia californica</i> <i>Prunus cerasus</i>	glycerin based leaf cells extract glycerin based leaf cells extract glycerin based leaf cells extract	protective, anti-pollution protective, antioxidant, anti-aging protective, antioxidant, cell relaxing	Naolys, France Naolys, France Naolys, France
Smooth Lightening White rose	<i>Rosa alba</i>	glycerin based leaf cells extract	protective, antioxidant, radiant, regenerating, detoxifying	Naolys, France
Sun Protect Date palm	<i>Bombax costatum</i>	glycerin based leaf cells extract	protective, soothing, sunscreen, anti-aging	Naolys, France
Whole Protection Edelweiss Whole Protection Red-flowered silk cotton tree Refine Ginger	<i>Leontopodium alpinum</i> <i>Bombax costatum</i> <i>Zingiber officinale</i>	sunflower oil based leaf cells extract glycerin based leaf cells extract glycerin based leaf cells extract	protective, soothing, repairing, anti-aging protective, soothing astringent, firming, mattifying, moisturizing, antioxidant	Naolys, France Naolys, France Naolys, France

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Pure Light Chinese peony	<i>Paeonia lactiflora</i>	powdered leaf cells	matifying, radiance, moisturizing, soothing, relaxing	Naolys, France
Unwind Sacred Lotus	<i>Nelumbo nucifera</i>	sunflower oil based leaf cells extract	soothing, relaxing, radiance	Naolys, France
Essential Being Indian jasmine	<i>Jasminum sambac</i>	glycerin based leaf cells extract	regenerating, detoxifying	Naolys, France
Full Detox Ylang Ylang	<i>Cananga odorata</i>	sunflower oil based leaf cells extract	radiance, matifying, regenerating, antioxidant, anti-pollution	Naolys, France
Purify Apothecary's rose	<i>Rosa gallica</i>	sunflower oil based callus extract	soothing, regenerating, radiance	Naolys, France
Purify Aloe vera	<i>Aloe barbadensis</i>	sunflower oil based callus extract	soothing, regenerating, radiance	Naolys, France
First Light Snow lotus	<i>Saussurea involucrata</i>	sunflower oil based callus extract	radiance, brightening, regenerating, antioxidant	Naolys, France
HydraSourcing [AM+PS]	<i>Argemone mexicana</i>	glycerin based callus extract	moisturizing, protective, regenerative, anti-aging	Naolys, France
HydraGeneration Papyrus	<i>Cyperus papyrus</i>	powdered leaf cells	hydrating, protective, barrier function	Naolys, France
HydraGeneration Pale rose	<i>Rosa centifolia</i>	glycerin based leaf cells extract	moisturizing, regenerative	Naolys, France
HydraSoothing Indian olibanum	<i>Boswellia serrata</i>	sunflower oil based leaf cells extract	moisturizing, regenerative, antioxidant, soothing	Naolys, France
Soothing Light Apple tree	<i>Malus domestica</i>	powdered leaf cells	soothing, lightening, radiance	Naolys, France
Fragile Vitis flower	<i>Vitis vinifera</i>	glycerin based flower cells extract	soothing, antioxidant	Naolys, France
Fragile Cotton	<i>Gossypium arboreum</i>	glycerin based leaf cells extract	soothing, protective	Naolys, France
Fragile Japanese Cherry tree	<i>Prunus serrulata</i>	sunflower oil based leaf cells extract	soothing, antioxidant	Naolys, France
Soothing Light Apricot	<i>Prunus armeniaca</i>	glycerin based leaf cells extract	soothing, lightening	Naolys, France
OvernightEnhance [M]+C]	<i>Mirabilis jalapa</i>	glycerin based callus extract	radiance, energizing, repairing, detoxifying, antioxidant	Naolys, France
Balancing Energy Asian ginseng	<i>Panax ginseng</i>	sunflower oil based leaf cells extract	energizing, antioxidant	Naolys, France
Light&Energy Coffee and Saffron	<i>Crocus sativus</i> and <i>Coffea arabica</i>	sunflower oil based callus and leaf cells extract	energizing, antioxidant, radiance	Naolys, France
Full Energy Vanilla	<i>Vanilla planifolia</i>	sunflower oil based leaf cells extract	energizing, antioxidant, regenerating	Naolys, France

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Splint&Slim Great bougainvillea	<i>Bougainvillea spectabilis</i>	powdered leaf cells	firming, slimming, lipolitical, anti-aging	Naolys, France
EtHAIRReal Peppermint	<i>Mentha piperita</i>	sunflower oil based leaf cells extract	regulating, soothing, antioxidant, lightening	Naolys, France
Healthy Shine Lilac	<i>Syringa vulgaris</i>	glycerin based leaf cells extract	restoring shine and damaged hair, soothing, energizing, protective	Naolys, France
3HC Hair Stimulation Complex	<i>Vitis vinifera</i>	mix of meristem plant cell culture derived active ingredients and hydrolyzed plant extracts	increase the lifespan of hair and reduce hair loss	Phenbiox SRL; Italy
Soy Cell	<i>Glycine max</i>	callus culture extract		
G-Cell	<i>Vitis vinifera</i>	meristem cell culture from green unripe grapes extract	anti-photo-aging, antiradical, anti-wrinkles	Phenbiox SRL; Italy
P-Cell	<i>Capsicum annuum</i>	fruit meristem cell culture extract	antioxidant, skin protective	Phenbiox SRL; Italy
Citrustem™	<i>Citrus aurantium dulcis</i>	callus culture extract	promote cell protein synthesis, increase skin elasticity	Phenbiox SRL; Italy
Lingostem™	<i>Vaccinium vitis-idaea</i>	callus culture extract	anti-aging, skin conditioner	Provital Group; Spain
RASTEM	<i>Lponoea purpurea</i>	callus culture extract	anti-aging, anti-wrinkle, make up treatments	Provital Group; Spain
Carrot Stem Cell	<i>Daucus carota</i>	callus culture extract	skin-renewal, anti-inflammatory,	Radiant Inc.; Korea
Cucumber Stem Cell	<i>Cucumis sativus</i>	callus culture extract	anti-wrinkle, antioxidant	Radiant Inc.; Korea
Ginseng Stem Cell	<i>Panax ginseng</i>	callus culture extract	antioxidant, skin-nourishing effect	Radiant Inc.; Korea
Lotus Stem Cell Extract	<i>Nelumbo nucifera</i>	cell culture extract	skin cooling effect, moisturizer	Radiant Inc.; Korea
Tomato Callus Stem Cell Extract	<i>Solanum lycopersicum</i>	callus culture extract	skin regeneration	Radiant Inc.; Korea
			controls excessive sebum secretion, absorbs sebum, maintains clean skin condition	Sandream Impact LLC; USA
			moisturizer	Sandream Impact LLC; USA

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Rice Callus Stem Cell Extract	<i>Oryza sativa</i>	callus culture extract	antioxidant, anti-inflammatory, whitening moisturizer	Sandream Impact LLC; USA
Carrot Callus Stem Cell Extract	<i>Daucus carota</i>	callus culture extract	protecting longevity, delaying senescence, increasing skin vitality, boosting epidermal skin cell regeneration	Sandream Impact LLC; USA
Rose Callus Stem Cell Extract	<i>Rosa damascena</i>	callus culture extract	antioxidant, anti-inflammatory	Sandream Impact LLC; USA
Grape Callus Stem Cell Extract	<i>Vitis vinifera</i>	callus culture extract	promote skin growth and proliferation, moisturizer, skin rejuvenation, soothing effect	Sandream Impact LLC; USA
Orchid Callus Stem Cell Extract	<i>Orchis</i> spp.	callus culture extract	restorative effect, tonic	Sandream Impact LLC; USA
Ginseng Callus Stem Cell Extract	<i>Panax pseudoginseng</i>	callus culture extract	antioxidant, anti-inflammatory	Sandream Impact LLC; USA
Green Tea Callus Stem Cell Extract	<i>Camellia sinensis</i>	callus culture extract	protective effect, pollution defense, skin conditioner	Sandream Impact LLC; USA
Marrubium Stems GX™	<i>Marrubium vulgare</i>	cell suspension extract	anti-photo-aging, anti-aging, protects from UV-induced oxidative stress, anti-inflammatory, matrix metalloproteinase activation	Sederna (Croda Personal Care); United Kingdom
Buddleja Stems GX™	<i>Buddleja davidii</i>	cell suspension extract	reduces skin redness, straightening capillary structure, fights against rosacea	Sederna (Croda Personal Care); United Kingdom
Centella Stems GX™	<i>Centella asiatica</i>	cell suspension extract		

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Dermasyr 10™	<i>Syringa vulgaris</i>	verbascoside concentrate extracted from leaf cell culture	reduce skin blemishes and inflammatory hyper-pigmentation, control skin redness and balances of seborrhea, soothing, purifying, skin conditioning	Sederna (Croda Personal Care); United Kingdom
Echinacea Stems GX™	<i>Echinacea angustifolia</i>	cell suspension extract	stimulates collagen synthesis, prevents collagen loss, reduces capillary permeability	Sederna (Croda Personal Care); United Kingdom
Gardenia Stems GX™	<i>Gardenia jasminoides</i>	cell suspension extract	reduces MMP-1 synthesis, inhibits collagenase activity, stimulates collagen synthesis	Sederna (Croda Personal Care); United Kingdom
Leontopod Stems GX™	<i>Leontopodium alpinum</i>	cell suspension extract	prevents collagen loss, antioxidant, anti-wrinkle	Sederna (Croda Personal Care); United Kingdom
Celtosome™ Eryngium Maritimum ST	<i>Eryngium maritimum</i>	dried stem cell	anti-aging, improving skin firmness, skin renewal	SEPPIC (Air Liquide Health-care); France
Celtosome Crithmum Maritimum ST	<i>Crithmum maritimum</i>	dried stem cell	skin lightening, wound healing, lightening effect, pigmentation regulation, anti-wrinkle	SEPPIC (Air Liquide Health-care); France
Ddobyul®	<i>Panax ginseng</i>	cambial meristematic cells suspension extract	anti-aging	Unhwa Corp.; Korea
Ddobyul®	<i>Taxus cuspidata</i>	cambial meristematic cells suspension extract	anti-allergic	Unhwa Corp.; Korea

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
Ddobyul®	<i>Ginkgo biloba</i>	cambial meristematic cells suspension extract	whitening	Unhwa Corp.; Korea
Ddobyul®	<i>Solanum lycopersicum</i>	cambial meristematic cells suspension extract	antioxidant	Unhwa Corp.; Korea
Vita SeneBlock	<i>Citrus limon</i>	mix of cell wall peptides and sugars from somatic embryos	inducing youth and longevity markers, protect DNA	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Vita iLux	<i>Capsicum annuum</i>	mix of peptides and sugars from cell cultures	firming effect, promoting bilirubin degradation, improves skin's regenerative process	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Hibiskin Vita	<i>Hibiscus syriacus</i>	cell culture extract	restore collagen, improve skin barrier function, moisturizer, improves skin's regenerative process	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Mythos Vita	<i>Actinidia arguta</i>	mix of stem cell culture derived extract and hydrosoluble plant fruit extract	anti-photo-aging, protect DNA	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Vita Nova	<i>Lotus japonicus</i>	mix of cell wall peptides and sugars from somatic embryos	extends cellular vitality and longevity, anti-aging, reactivate skin's natural repair process	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Vita Genesis White	<i>Brassica rapa</i>	mix of cell wall fraction and hydro-ethanolic extract derived from hairy root cultures	inhibits pigmentation process, reduce melanin synthesis	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Vita Freeze	<i>Solanum lycopersicum</i>	mix of peptides and sugars from cell cultures	prevent premature skin ageing, improve skin elasticity, extend cellular vitality and longevity	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Daphne VitaSense	<i>Daphne odoracell</i>	cell suspension extract	strengthens skin protecting barrier, accelerate healing of cutaneous micro-lesions, reduce skin inflammation	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy
Vitalight	<i>Cirsium eriophorum</i>	cell suspension extract	reduce sebum production, reduce skin inflammation, increase skin desquamation	Vitalab s.r.l. (Arterra Bioscience s.r.l); Italy

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
VitaShape	<i>Coleus forskohlii</i>	cell suspension extract	anti-fat properties, anti-oedema effect, antioxidant, detoxifying capacity	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
BerryFlux Vita	<i>Rubus idaeus</i>	cell culture extract	moisturizer, hydrating effect, improve production of ceramides	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
FicuCell Vita	<i>Opuntia</i> spp.	cell culture extract	protect extracellular matrix, extend cellular longevity, delay premature skin ageing, improve skin firmness and elasticity	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
DoliCos PhotoProtect	<i>Dolichos</i> spp.	cell culture extract	UV protective, detoxify cells, soothing effect	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
Cell Pulse	<i>Coffea</i> spp.	cell culture extract	antioxidant, cell energiser, anti-wrinkle	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
Lycoskin Defence	<i>Solanum lycopersicum</i>	water-soluble fraction from cell cultures	extend cellular vitality, protect DNA, radiance effect	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
BioNymph Peptide	<i>Nicotiana sylvestris</i>	mix of peptides and sugars from cell cultures	antioxidant, extend cellular vitality and longevity, anti-wrinkle, improve skin smoothness	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
CellIntegrity	<i>Rubus idaeus</i>	concentrated water-soluble fraction from cell cultures	antioxidant, anti-inflammatory, protect DNA, enhance cellular longevity, soothing effect, strengthening action	Vitalab s.r.l. (Arterra Bioscience s.r.l.); Italy
SENSIA CAROTA PRCF	<i>Daucus carota sativa</i>	lysate of root stem cells culture	anti-aging, anti-inflammatory, antioxidant	Vytrus Biotech; Spain
LUMINIA GRANATUM PRCF	<i>Punica granatum</i>	powdered plant stem cells	antioxidant, reduces hyper-pigmentation, enhances skin radiance and glow	Vytrus Biotech; Spain

(Continued)

Table 1. Continued.

Market names of active ingredient	Plant species	Type of cell culture and extracts	Benefits	Company; country
ARABIAN COTTON PRCF	<i>Gossypium herbaceum</i>	lysate of plant stem cells culture	soothing, anti-inflammatory, antioxidant, photoprotection, regeneration	Výtrus Biotech; Spain
TURMERIA ZEN PRCF	<i>Curcuma longa</i>	glycerin based lysate of rhizome cells culture	anti-stress wrinkles, emotional hydration manager, modulator of Brain-skin connection	Výtrus Biotech; Spain
SARCOSLIM RE-SHAPE PRCF	<i>Sarcocapnos crassifolia</i>	lysate of callus culture	skin regeneration and repair	Výtrus Biotech; Spain
CAPILLA LONGA PPF	<i>Curcuma longa</i>	plant growth factor peptides - concentrated secretome of totipotent rhizome cells	hair follicle regeneration, hair growth re-activation	Výtrus Biotech; Spain
CENTELLA REVERSA PPF	<i>Centella asiatica</i>	plant signaling peptides - concentrated secretome of totipotent petioles cells	rebuilding the core skin structure	Výtrus Biotech; Spain

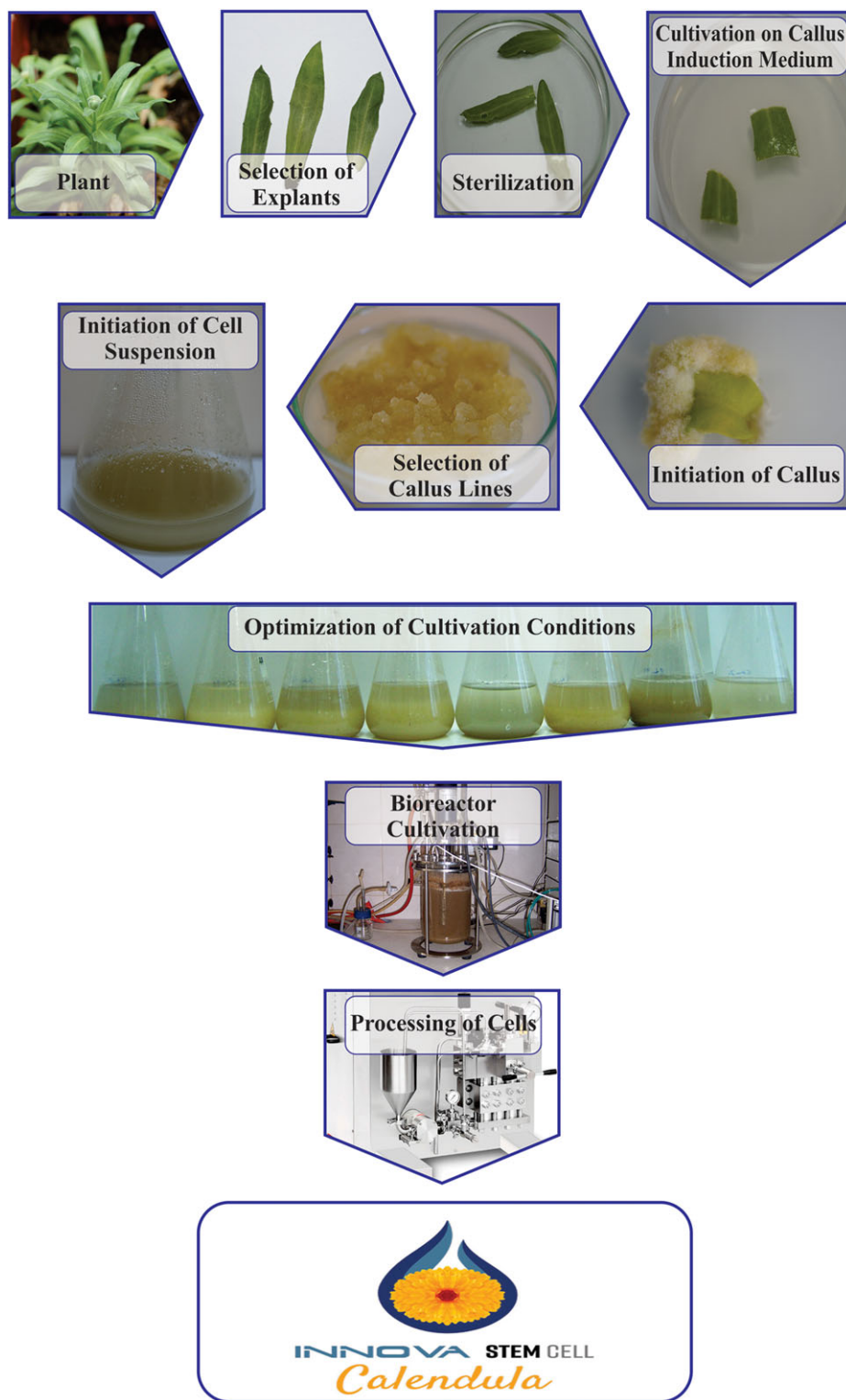


Figure 1. Schematic presentation of technological steps in development of InnovaStem-Cell Calendula active cosmetic ingredient.

important to understand, that the callus cultures are plant cells, cultured on solid medium, whereas the cell suspension cultures are single plant cells or small cell aggregates cultivated under submerged conditions in liquid medium. Both callus cultures

and cell suspensions could consist of dedifferentiated or true stem cells. On the opposite, the hairy roots are organ cultures, obtained by genetic transformation of plant cells [23, 34]. Some of the plant cell culture technology – derived active cosmetic

ingredients, currently available on the market are reviewed in Table 1.

3.1 Plant stem cells

Plant cells can be propagated and used for continuous supply of fresh plant biomass for cosmetic formulations. However, it is of great importance to understand that we cannot introduce the entire plant cells in cosmetic products and keep them alive as active ingredient there [30,35]. Plant cells are extremely sensitive to environmental factors, nutrient medium composition, osmotic and mechanical (shear) stress, gas exchange and oxygen supply, temperature, light, ionic strength and water content, and thus, they can survive neither during cosmetic products preparation, nor during storage or application of cosmetics on skin. Even a delivery system, able to maintain and supply live stem cells could be developed, the size and the specific structure of plant cells will not allow them to attach or penetrate the skin surface. Because of all discussed concerns, the plant stem cells are rather used as a raw material for preparing different types of extracts, which then could be included in cosmetic formulations as active ingredients. However, there are various products, available on the market, which are based on a whole dried plant cells and standardized on the cells count per gram of active ingredient (Table 1). Such examples are the anti-aging active ingredient Celtsome™

Eryngium Maritimum ST (based on sea holly cell culture with 100 million cells/g of active ingredient), and the tyrosinase inhibitory skin lightening active ingredient Celtsome™ Crithmum Maritimum ST (based on rock samphire cell culture with more than 1 billion cells/g of active ingredient) (Table 1).

3.2 Plant stem cells extracts

Most of the available active cosmetic ingredients, obtained by plant cell culture technology are marketed in the form of different extracts (Table 1). In contrast to the plant – derived extracts, the extracts obtained from plant cell cultures can be easily standardized, and perfectly complying with the strict safety requirements that the high end cosmetic market constantly demands [35]. Plant cell extracts are free of pathogens, agrochemical, toxic substances, allergens and pollutants, because they are produced under controlled conditions, complying with the procedures of good manufacturing practice. Depending of the type of used solvent, the plant cell extracts used in cosmetics could be contingently divided to liposoluble (extracted with oils) and hydrosoluble (extracted with glycerin) extracts, dried extracts (conditioned with maltodextrin), plant cell wall extracts (rich in peptides and sugars), nanoemulsions or suspension extracts [35]. However, most of the existed extracts have been developed on the basis on extraction of target compound or group of closely related

Table 2. GC-MS profiles of *Calendula officinalis* stem cell suspension before and after optimization of nutrient medium composition [47]

Metabolites	Calendula cell suspension before optimization μg/g dry biomass	Calendula cell suspension after optimization μg/g dry biomass
Amino acids		
Glycine	62.77	106.74
Serine	21.09	175.87
L-Proline	73.08	162.56
L-Aspartic acid	10.83	138.41
L-Glutamic acid	161.68	274.91
L-Asparagine	19.19	146.38
L-Glutamine	88.03	224.10
Organic acids		
Succinic acid	6.40	130.88
Fumaric acid	7.23	172.30
Malic acid	5.41	149.20
GABA	50.02	184.38
Pyroglutamic acid	20.41	115.51
Saturated and Non-saturated Fatty Acids		
n-Tetracanoic acid (Myristic acid) (C14:0)	26.68	83.07
n-Hexadecanoic acid (Palmitic acid) (C16:0)	183.91	178.69
9-(Z)-Hexadecenoic acid (Oleic acid) (C 18:1)	39.33	95.36
Octadecanoic acid (Stearic acid) (C 18:0)	197.61	192.01
Sterols		
Campesterol	59.27	59.07
Stigmasterol	65.34	89.14
β-Sitosterol	94.82	112.77
β-Amyrin	12.13	77.46
Phenolic acids		
Quinic acid	13.49	81.92
Caffeic acid	-	62.05
trans-Cinnamic acid	-	170.39
p-Coumaric acid	-	67.63

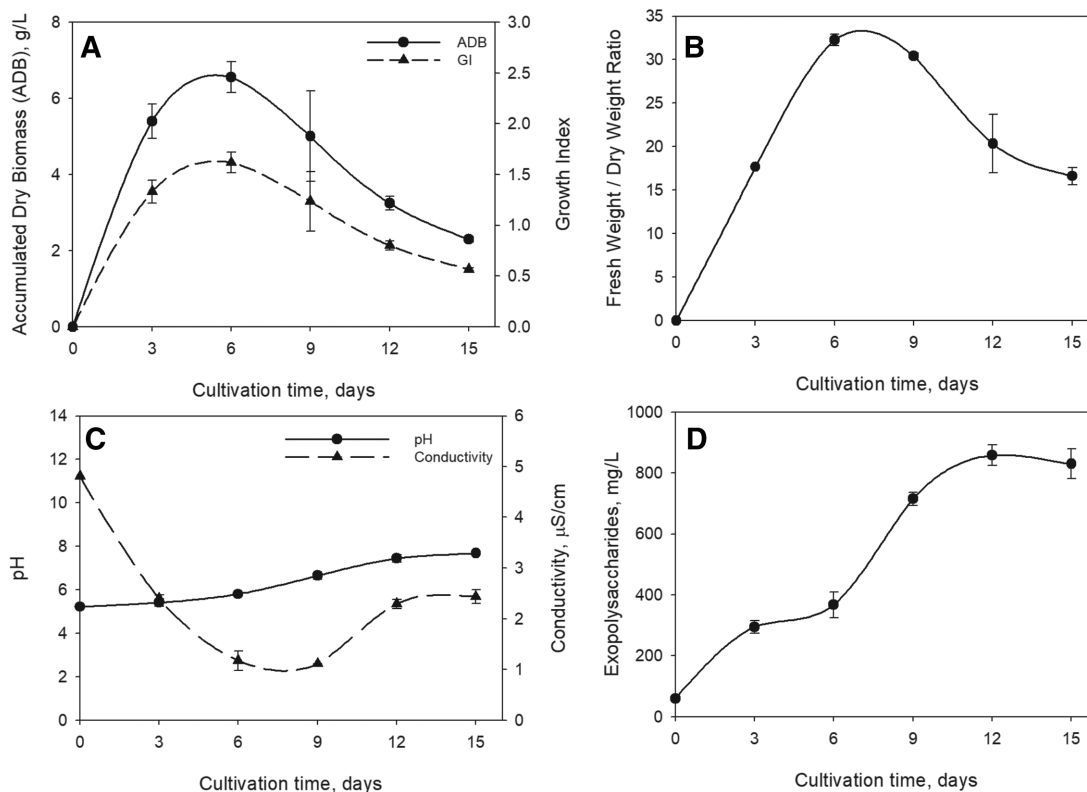


Figure 2. Dynamics of changes in accumulated dry biomass and growth index (A), fresh weight to dry weight ratio (B), pH and specific electric conductivity of culture medium (C), and exopolysaccharides release (D) during 15 days of submerged cultivation in shaking flasks of *Calendula officinalis* L. cell suspension culture (115 rpm, 26°C, on darkness).



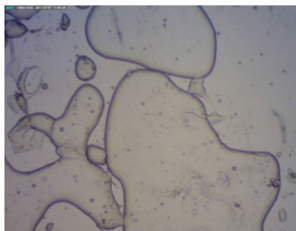
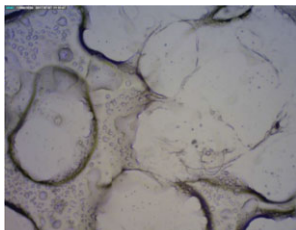
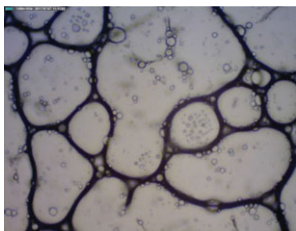
bioactive compounds, and thus, not the entire health beneficial potential of the extracted plant cells could be utilized. Some exceptions could be found, where the entire plant cells were freeze dried and powdered for direct application in cosmetic formulations (see some products offered by “Vytrus Biotech” and “Naolys”, Table 1), or the entire cell suspensions have been, emulsified or encapsulated in liposomal complex (see some products offered by “Innova BM” and “Mibelle AG Biochemistry”, Table 1) by using high-pressure homogenizers.

3.3 Molecular farming for production of recombinant proteins

Plants are excellent production matrixes for expression of recombinant proteins with important pharmaceutical properties [36]. This powerful recombinant protein expression technique, known as “molecular farming” have been widely used for the production of vaccines [37–39], cytokines [40], and even for production of therapeutic protein for human use [5, 41, 42]. The classical molecular farming technology is based on genetic modification of plants for recombinant protein expression. This could be realized by permanent integration of foreign genes into the host DNA to generate stable transgenic lines, or by transient transformation of intact plant leaves [43]. However, both methods requires growing of whole plants under strictly

controlled environmental conditions into contained greenhouses, complying with the strict regulatory standards [36]. By using such technology, The Iceland Company “BIOEFFECT” has applied for the first time a plant-based transgenic platform for large-scale production of Epidermal Growth Factors (EGF) for cosmetic use, expressed in genetically engineered barley seeds. The company offers a wide range of skin-care products containing this cellular activator, which contribute for healthier and younger-looking skin. However, the recent interests in molecular farming have been focused on adaptation of plant cell culture technology for the production of recombinant proteins. This technique offers sustainable and continuous heterologous proteins production by plant cells growing under precisely controlled micro-environmental *in vitro* conditions in bioreactors. Cultivation of genetically engineered plant cell cultures have been recognized as much powerful expression platform, when compared to the classical molecular farming techniques, relying on agricultural-scale production by growing of genetically transformed plants [18, 41]. Recently, the Korean company “Natural Bio-Materials (NBM)”, has launched series of active cosmetic ingredients containing growth factors (Epidermal Growth Factor, Basic Fibroblast Growth Factor, Insulin-like Growth Factor-1, Keratinocyte Growth Factor, Fibroblast Growth Factor-7, and Vascular Endothelial Growth Factor), expressed by recombinant rice (*Oryza sativa* L.) cell cultures (Table 1). However, even that

Table 3. Synergistic effect of exopolysaccharides, released by *Calendula officinalis* stem cell suspension in relation to emulsion stabilizing properties in mixtures with cellulose gum, xanthan gum, guar gum, and sodium-alginate

Model emulsion systems Oil/Water (1:1 v/v) with:	Light microscopy	Turbidimetric method	
		Emulsifying activity index (EAI), m ² /g	Emulsion stability index (ESI), min
0.6 % Calendula Cell Suspension Exopolysaccharides		175.0	16.8
0.6 % Calendula Cell Suspension Exopolysaccharides And 0.3 % Cellulose Gum		38.4	13.9
0.6 % Calendula Cell Suspension Exopolysaccharides and 0.3 % Xanthan Gum		47.6	22.1
0.6 % Calendula Cell Suspension Exopolysaccharides and 0.3 % Guar Gum		322.4	15.0
0.6 % Calendula Cell Suspension Exopolysaccharides and 0.3 % Sodium Alginate		1307.5	60.7

The emulsifying activity index (EAI) and emulsion stability index (ESI) were determined according to [48]. Model emulsion systems were prepared by mixing polysaccharides, water and sunflower oil for 5 min at 10 000 rpm by homogenizer (Ultra Turrax IKA T18 Basic, Germany).

the human recombinant proteins, produced by this technology have undeniable advantages and are characterized with high level of purity, animal-free, virus-free, bacterial-free and exotoxin-free production, there are still some consumers which have concerns to use such products because they are expressed by genetically modified organisms.

4 Case Study: Initiation, growth, phytochemical profile and physicochemical characteristics of exopolysaccharides in INNOVA StemCell Calendula

Calendula (*Calendula officinalis* L.), known as “marigold”, has been widely used in traditional herbal medicine and skin care cosmeceuticals for topical application [11]. The plant was shown to be rich in phenolic acids, flavonoid, triterpenes, carotenoids, aroma compounds and unique mix of polyunsaturated fatty acids [44–46]. Because of its high therapeutic value and the proved cosmetic effects, the Bulgarian company “Innova BM” has developed and released on the market two high quality active cosmetic ingredients, based on Calendula dedifferentiated cell culture (Table 1). The development of these products is schematically presented on Fig. 1. The technological steps include screening of calendula plants with superior phytochemical profiles, selection, sterilization and cultivation of plant explants on callus induction medium, selection of friable cell lines with appropriate phytochemical profiles, initiation of liquid cell suspension culture and optimization of cultivation conditions and nutrient medium composition. The optimization step is critical in our technology, since a significant increase in biosynthetic potential and accumulated biomass of selected cell line can be achieved (Table 2, Fig. 2). After optimization, the selected line was scaled-up to large scale cultivation in stirred tank bioreactor. The produced cell suspension (cells and culture liquid) was then processed by high-pressure homogenizer to produce glycerin extract (50 % wt.) or calendula emulsion (75 % wt. cell suspension) [47]. The produced active ingredients have been found to have superior moisturizing, anti-wrinkle, hydrating and regenerative effects, when applied on skin. These effects are due to the high content of secreted exopolysaccharides, during cultivation of calendula cells (Fig. 2D). The exopolysaccharides have been identified to belong to pectin type. The crude exopolysaccharides fraction contains 879 $\mu\text{g}/\text{mg}$ neutral sugars and 50 $\mu\text{g}/\text{mg}$ proteins. The unique combination of polysaccharides and peptides made this exopolysaccharide fraction almost perfect for application as emulsifier in cosmetic products for topical application. The potential synergistic interactions, when applied with other popular emulsifiers, as well as their emulsion stabilization properties are presented in Table 3. The polysaccharides have molecular weight of 6.7×10^4 Da and contain 413 $\mu\text{g}/\text{mg}$ uronic acids. The full monosaccharide composition of exopolysaccharide fraction was determined as: glucuronic acid (13.6 $\mu\text{g}/\text{mg}$), galacturonic acid (399.7 $\mu\text{g}/\text{mg}$), glucose (185.5 $\mu\text{g}/\text{mg}$), galactose (179.9 $\mu\text{g}/\text{mg}$), rhamnose (178.9 $\mu\text{g}/\text{mg}$), arabinose (166.7 $\mu\text{g}/\text{mg}$), fucose (0.6 $\mu\text{g}/\text{mg}$) and mannose (4.7 $\mu\text{g}/\text{mg}$). The presence of this exopolysaccharides fraction incorporated into Innova StemCell

Calendula products, in combination with bioactive compounds from the released cell content (Table 2) made these products unique and one of the kind in the market of active cosmetic ingredients.

5 Concluding remarks

Following the ever-growing demand for high quality natural products of plant origin for application as active ingredients in cosmeceutical formulations, the plant cell culture technology has developed powerful production platforms which can effectively supply the customers' needs. We are witnesses of exponentially growing number of commercialized plant cell – derived ingredients, offered on the cosmetic industry market and the diversity of utilized plant species, used for their production has continue to increase every year. In fact, the observed interest to production of plant cell – derived ingredients for cosmetic needs could be correlated with the recent advance in development and commercialization of plant cell culture technology in technologically advanced countries. Moreover, the new developed techniques of gene editing, metabolite engineering and synthetic biology could have a significant impact on improvement the yields and development of tailor made cosmetic products with desired activities. The advance in molecular farming has already led to commercial production of rare human activator peptides, cytokines and growth factors, which are the first step in development of cosmetic products with potential to extend skin life by using the body self-repair mechanisms. However, till now there are many open questions, concerning regulatory standards and documentation, unification of health beneficial claims and the methodology for evaluation of pharmaceutical effects, which should be answered in order to help the consumers to make the right choice of their cosmeceutical product.

Practical application

The aim of this paper is to review the recent progress in plant cell technology for cosmetic application and to provide short overview on commercialized plant cell culture – derived active cosmetic ingredients available on the market.

We would like to thanks the Innova BM, Bulgaria, for the financial support, close collaboration and the shared data.

The authors have declared no conflict of interest.

6 References

- [1] Siahshar, B., Rahimi, M., Tavassoli, A., Raissi, A., Application of biotechnology in production of medicinal plants. *Am. Eurasian J. Agric. Environ. Sci.* 2011, 11, 439–444.

- [2] Rischer, H. T., Hakkinen, S., Ritala, A., Seppanen-Laakso, T., et al., Plant cells as pharmaceutical factories. *Curr. Pharm. Des.* 2013, 19, 5640–5660.
- [3] Bodeker G, Bhat K, Burley J, P., Vantomme, (Eds.) *Medicinal plants for forest conservation and health care*, Food & Agriculture Org. 1997.
- [4] Brown, A. H. D., Hodgkin, T., Indicators of genetic diversity, genetic erosion, and genetic vulnerability for plant genetic resources, in: M. R. Ahuja, S. M. Jain (Eds.) *Genetic Diversity and Erosion in Plants: Indicators and Prevention*, Springer International Publishing, Cham, 2015, pp. 25–53.
- [5] Georgiev, V., Mass propagation of plant cells—an emerging technology platform for sustainable production of biopharmaceuticals. *Biochem. Pharmacol. (Los Angel)*. 2015, 4, e180.
- [6] Charles Dorni, A. I., Amalraj, A., Gopi, S., Varma, K., et al., Novel cosmeceuticals from plants—An industry guided review. *J. Appl. Res. Med. Aromat. Plants*. 2017, 7, 1–26.
- [7] Armendáriz-Barragán, B., Zafar, N., Badri, W., Galindo-Rodríguez, S. A., et al., Plant extracts: from encapsulation to application. *Expert Opinion on Drug Delivery*. 2016, 13, 1165–1175.
- [8] Działo, M., Mierziak, J., Korzun, U., Preisner, M., et al., The potential of plant phenolics in prevention and therapy of skin disorders. *Internat. J. Mol. Sci.* 2016, 17, 160.
- [9] Ferrazzano, G., Amato, I., Ingenito, A., Zarrelli, A., et al., Plant polyphenols and their anti-cariogenic properties: a review. *Molecules* 2011, 16, 1486.
- [10] Ribeiro, A., Estanqueiro, M., Oliveira, M., Sousa Lobo, J., Main benefits and applicability of plant extracts in skin care products. *Cosmetics* 2015, 2, 48.
- [11] Gediya, S. K., Mistry, R. B., Patel, U. K., Blessy, M., et al., Herbal plants: used as a cosmetics. *J. Nat. Prod. Plant Resour.* 2011, 1, 24–32.
- [12] Papaioanou, M., Chronopoulou, E., Ciobotari, G., Efroze, R., et al., Cosmeceutical properties of two cultivars of red raspberry grown under different conditions. *Cosmetics* 2018, 5, 20.
- [13] Taofiq, O., González-Paramás, A. M., Martins, A., Barreiro, M. F., et al., Mushrooms extracts and compounds in cosmetics, cosmeceuticals and nutraceuticals—a review. *Industrial Crops Products* 2016, 90, 38–48.
- [14] Barbulova, A., Colucci, G., Apone, F., New trends in cosmetics: by-products of plant origin and their potential use as cosmetic active ingredients. *Cosmetics* 2015, 2, 82.
- [15] Guillerme, J.-B., Couteau, C., Coiffard, L., Applications for marine resources in cosmetics. *Cosmetics* 2017, 4, 35.
- [16] Juliano, C., Magrini, G., Cosmetic functional ingredients from botanical sources for anti-pollution skincare products. *Cosmetics* 2018, 5, 19.
- [17] Wang, H.-M. D., Chen, C.-C., Huynh, P., Chang, J.-S., Exploring the potential of using algae in cosmetics. *Bioresour. Technol.* 2015, 184, 355–362.
- [18] Hellwig, S., Drossard, J., Twyman, R. M., Fischer, R., Plant cell cultures for the production of recombinant proteins. *Nat. Biotech.* 2004, 22, 1415–1422.
- [19] Yue, W., Ming, Q.-l., Lin, B., Rahman, K., et al., Medicinal plant cell suspension cultures: pharmaceutical applications and high-yielding strategies for the desired secondary metabolites. *Crit. Rev. Biotechnol.* 2016, 36, 215–232.
- [20] Wilson, S. A., Roberts, S. C., Recent advances towards development and commercialization of plant cell culture processes for the synthesis of biomolecules. *Plant Biotechnol. J.* 2012, 10, 249–268.
- [21] Jeandet, P., Clément, C., Courot, E., Resveratrol production at large scale using plant cell suspensions. *Engineer. Life Sci.* 2014, 14, 622–632.
- [22] Rahpeyma, S.-A., Moieni, A., Jalali Javaran, M., Paclitaxel production is enhanced in suspension-cultured hazel (*Corylus avellana* L.) cells by using a combination of sugar, precursor, and elicitor. *Engineer. Life Sci.* 2015, 15, 234
- [23] Kümmitz, S., Haas, C., Winkler, K., Georgiev, V., et al., Hairy Roots of *Salvia* Species for Bioactive Substances Production, in: V. Georgiev, A. Pavlov (Eds.) *Salvia Biotechnology*, Springer International Publishing, Cham, 2017, pp. 271–289–242.
- [24] D’Amelia, V., Ruggiero, A., Tranchida-Lombardo, V., Leone, A., et al., Biosynthesis of salvia specialized metabolites and biotechnological approaches to increase their production, in: V. Georgiev, A. Pavlov (Eds.) *Salvia Biotechnology*, Springer International Publishing, Cham, 2017, pp. 241–270.
- [25] Savona, M., Barberini, S., Bassolino, L., Mozzanini, E., et al., Strategies for Optimization of the Production of Rosmarinic Acid in *Salvia officinalis* L. and *Salvia dolomitica* Codd Biomass with Several Biotechnological Approaches, in: V. Georgiev, A. Pavlov (Eds.) *Salvia Biotechnology*, Springer International Publishing, Cham, 2017, pp. 209–239.
- [26] Imseng, N., Schillberg, S., Schürch, C., Schmid, D., et al., Suspension culture of plant cells under heterotrophic conditions, industrial scale suspension culture of living cells, Wiley-VCH Verlag GmbH & Co. KGaA 2014, pp. 224–258.
- [27] Ananga, A., Phills, B., Ochieng, J., Georgiev, V., et al., Production of anthocyanins in grape cell cultures: A potential source of raw material for pharmaceutical, food, and cosmetic industries, in: D. Poljuha, B. Sladonja (Eds.) *The Mediterranean Genetic Code - Grapevine and Olive*, INTECH Open Access Publisher 2013, pp. 247–287.
- [28] Trehan, S., Michniak-Kohn, B., Beri, K., Plant stem cells in cosmetics: current trends and future directions. *Future Sci. OA* 2017, 3, FSO226.
- [29] Skotnicka-Graca, U., Plant stem cells as innovation in cosmetics. *Acta Poloniae Pharmaceutica* 2014, 71, 701.
- [30] Draelos, Z., Plant stem cells and skin care. *Cosmet. Dermatol.* 2012, 25, 395–396.
- [31] Lee, E.-K., Jin, Y.-W., Park, J. H., Yoo, Y. M., et al., Cultured cambial meristematic cells as a source of plant natural products. *Nat. Biotech.* 2010, 28, 1213–1217.
- [32] Ochoa-Villarreal, M., Howat, S., Jang, M. O., Kim, I. S., et al., Cambial meristematic cells: a platform for the production of plant natural products. *New Biotechnol.* 2015, 32, 581–587.
- [33] Zhou, P., Yang, J., Zhu, J., He, S., et al., Effects of β -cyclodextrin and methyl jasmonate on the production of vindoline, catharanthine, and ajmalicine in *Catharanthus roseus* cambial meristematic cell cultures. *Applied Microbiol. Biotechnol.* 2015, 99, 7035–7045.
- [34] Steingroewer, J., Bley, T., Georgiev, V., Ivanov, I., et al., Bio-processing of differentiated plant in vitro systems. *Engineering Life Sci.* 2013, 13, 26–38.

- [35] Barbulova, A., Apone, F., Colucci, G., Plant cell cultures as source of cosmetic active ingredients. *Cosmetics* 2014, 1, 94.
- [36] Fischer, R., Vasilev, N., Twyman, R. M., Schillberg, S., High-value products from plants: the challenges of process optimization. *Curr. Opin. Biotechnol.* 2015, 32, 156–162.
- [37] Meeusen, E. N. T., Walker, J., Peters, A., Pastoret, P.-P., et al., Current status of veterinary vaccines. *Clin. Microbiol. Rev.* 2007, 20, 489–510.
- [38] Ruiz, V., Mozgovej, M. V., Dus Santos, M. J., Wigdorovitz, A., Plant-produced viral bovine vaccines: what happened during the last 10 years? *Plant Biotechnol. J.* 2015, 13, 1071–1077.
- [39] News In Brief. *Nat. Biotechnol.* 2006, 24, 233.
- [40] Tremblay, R., Wang, D., Jevnikar, A. M., Ma, S., Tobacco, a highly efficient green bioreactor for production of therapeutic proteins. *Biotechnol. Advan.* 2010, 28, 214–221.
- [41] Xu, J., Zhang, N., On the way to commercializing plant cell culture platform for biopharmaceuticals: present status and prospect. *Pharmaceut. Bioprocess.* 2014, 2, 499–518.
- [42] Fischer, R., Schillberg, S., Hellwig, S., Twyman, R. M., et al., GMP issues for recombinant plant-derived pharmaceutical proteins. *Biotechnol. Advan.* 2012, 30, 434–439.
- [43] Sainsbury, F. and Lomonosoff, G. P., Transient expressions of synthetic biology in plants. *Curr. Opin. Plant Biol.* 2014, 19, 1–7.
- [44] Ercetin, T., Senol, F. S., Erdogan Orhan, I., Toker, G., Comparative assessment of antioxidant and cholinesterase inhibitory properties of the marigold extracts from *Calendula arvensis* L. and *Calendula officinalis* L., *Industrial Crops Products* 2012, 36, 203–208.
- [45] Król, B., Paszko, T., Harvest date as a factor affecting crop yield, oil content and fatty acid composition of the seeds of calendula (*Calendula officinalis* L.) cultivars. *Industrial Crops Products* 2017, 97, 242–251.
- [46] Dulf, F. V., Pamfil, D., Baciú, A. D., Pinteá, A., Fatty acid composition of lipids in pot marigold (*Calendula officinalis* L.) seed genotypes. *Chem. Central J.* 2013, 7, 8–8.
- [47] Pavlov, A., Georgiev, V., Phytochemical composition of *Calendula officinalis* L. dedifferentiated cell suspension, BG Patent, Utility Model # 2773/26.09.2017, 2017.
- [48] Diniz, R. S., Coimbra, J. S. d. R., Teixeira, Á. V. N. d. C., da Costa, A. R., et al., Production, characterization and foamability of α -lactalbumin/glycomacropptide supramolecular structures. *Food Res. International* 2014, 64, 157–165.