

Bulgarian experience in aquaculture (microalgae) - cultivation, production and development

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The search for natural raw materials rich in biologically active and harmless substances for the pharmaceutical, cosmetics and food industries is extremely significant nowadays, when modern life is characterized by numerous ecological problems.

Microalgae are an untraditional but promising means to obtain the above purpose owing to possibilities to control, optimize and run large-scale production of useful and healthy biomass. The quality and economic efficiency of this production is site specific and highly dependent on climate, environmental conditions, developed know-how technology, as well as the existing local tradition of its use. The application of thermal waters, geothermal energy and carbon dioxide for microalgae cultivation provides high process optimization and considerably reduces the production costs.

The article deals with the existing experience in Bulgaria for more than 35 years in terms of geothermal resource, cultivation, production and application of microalgae products. Special attention is paid to the economic efficiency of its cultivation.

Keywords: microalgae, cultivation, thermal water, geothermal energy

Introduction

Microalgae development has a long tradition of cultivation and various applications all over the world. Only two bases operating with geothermal water and CO₂ have been established in Europe so far - in Roupi field, SW Bulgaria (set up in 1967) and Therma-Nigrita field, N.Greece (set up in 1997) by a cooperation with specialists from the Bulgarian Academy of Sciences [8].

Microalgae development in Roupi geothermal field (23°E.Long.,42° N.Lat.) was established by the Bulgarian Academy of Sciences both for cultivation and processing of green (*Chlorella* and *Scenedesmus*) and blue-green (*Spirulina*) algae biomass. Technologies for production of various types of products for medical and cosmetic application currently exist in Bulgaria.

Microalgae are used for stimulation of the immune system, support for the cardio-vascular system, raising the content of the “healthy “ cholesterol, improvement of the gastrointestinal and digestive tracts, improvement of the natural purgation and detoxification, reducing the risk of cancer through antioxidant protection, etc.

The quality and economic efficiency of microalgal production is site specific and highly dependent on climate, CO₂ supply, mineral nutrition and environmental conditions. The investigations of the Bulgarian scientists proved that the use of thermal water, geothermal carbon dioxide and geothermal energy provide a high optimization of the cultivation process and considerable reduction in production costs [7,8,9].

1. Roupi base

The Roupi geothermal field is located in SW Bulgaria close to the Struma river, Figure 1.

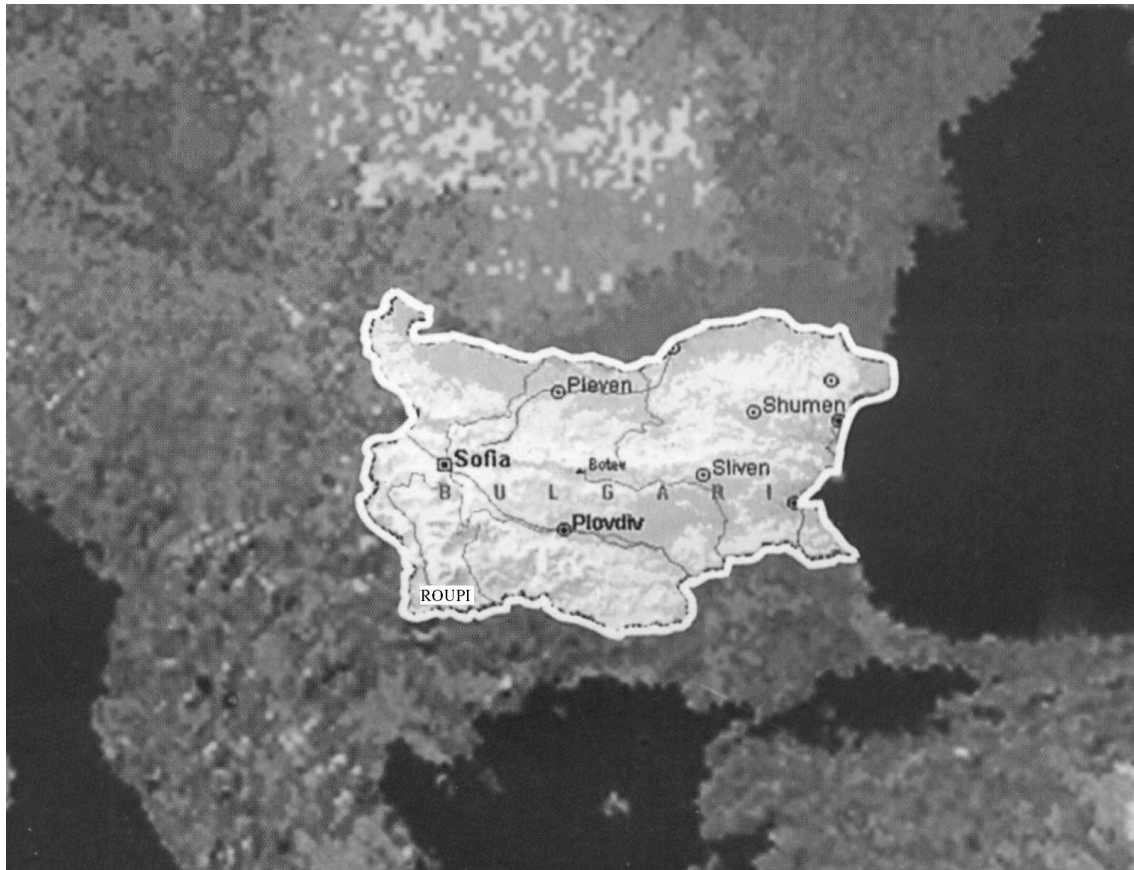


Figure 1. Location map of Roupi region

1.1. Climatic and environmental conditions

The climatic data characterize Roupi as a dry region of mean annual precipitation of 400-600 mm. The mean annual temperature is about 13-14°C as the mean monthly temperature in January is 1 -2°C, in April – 12-14°C, in July – 23-25°C and in October – 13-18°C. The mean daily solar radiation is 1680 J/cm² and there are about 240 sunny days per year. No air pollution exists in the region due to the lack of any industrial activities.

The microalgal cultivation ponds are located on the southern slope of the Kozuh extinct volcano at a distance of 2km from the drilled wells. The total used flow rate is about 15 l/s, water temperature - 74°C and pH - 7.6. The total dissolved solids (TDS) are 2.4 g/l and of them 2.11 g/l are for the hydro-carbonates. Chemical composition of the thermal water in Roupi is as follows (in mg/l): anions: F - 5.80, Cl - 35.78, J - 0.04, SO₄ - 123.04, HCO₃ - 1444.95, HPO₄ - 0.05, HS - 1.09, S₂O - 7.36; cations: Na - 594.28, K - 41.40, Ca - 31.43, Mg - 12.91, Al - 0.74; H₂SiO₃ - 65.91 and HBO₂ - 9.86 [3]. No harmful elements like As and Pb are found in the geothermal water. The freely released carbon dioxide is about 500 mg/l and the dissolved one is 266 mg/l.

The abovementioned data confirm that Roupi region satisfies all requirements for open microalgal cultivation.

1.2. Information on cultivation and processing cycle

Microalgal cultivation is based upon the logic of the photosynthetic process - solar energy is used for synthesis of organic compounds out of inorganic substances. Open mass cultivation still is a basic method for production of industrial amounts of microalgal biomass because it utilizes solar energy as a source of light and heat for photosynthesis.

Microalgal technology includes the following stages (Fig. 2):

I - inoculation (intensification of the cultivation process)

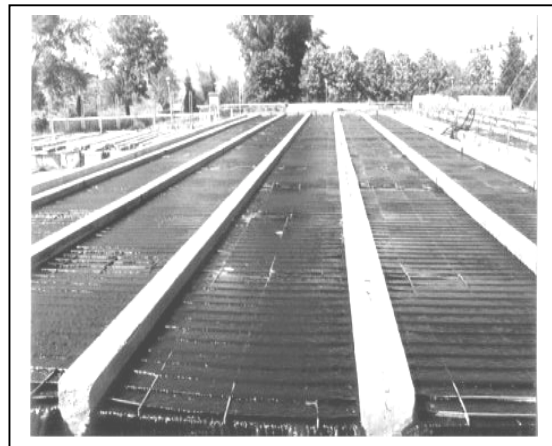
II - cultivation in production ponds

III - separation by centrifugation

IV - drying (by spray dryer) and packaging



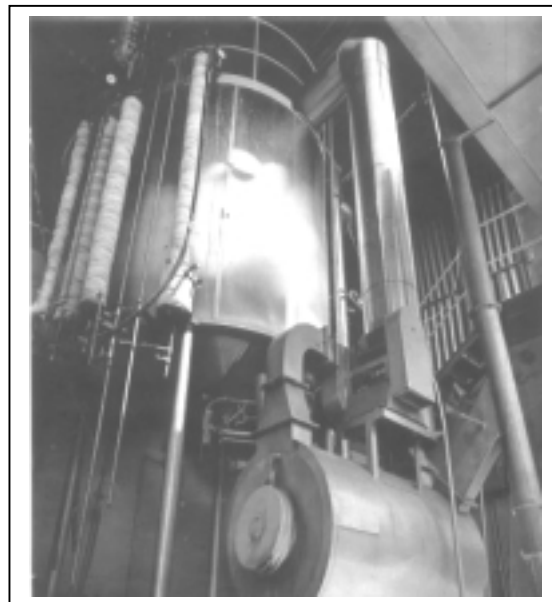
a) Inoculum production ponds



b) Production pond



c) Centrifuge



d) Dryer

Fig. 2. Views from Roupi base

The microalgal ponds cover a total area of 2600 m² and are of different size, varying: 50m², 220m², 900m² and 1300 m² respectively. They are oriented to north-south direction and are of

3° slope. The installation lines are 30 m long each of them with baffles at every 15 cm. The dark volume of suspension is 10-15 % and suspension's layer thickness is 5 cm.

Specialized technological equipment is in operation for the different stages of cultivation. The downstream biomass processing is developed both for dry product (in a powder form) and fresh biomass (centrifuged algal suspension of up to 10% of dry weight).

Complex climatic, biological and technological factors influence on the biomass productivity and its biochemical composition [1,2,3,5]. An optimization of the whole technological process of cultivation and processing has been achieved as a result of the comprehensive investigations of the Bulgarian scientists. The optimization concerns pond design, temperature and radiation regime of the suspension, nutrition media preparation, CO₂ supply, selection of high productive species and strains and as well as reduction of the production cost [3,6]. The relationship between the major climatic factors (temperature and radiation) influencing on the microalgae cultivation is presented on Figure.3.

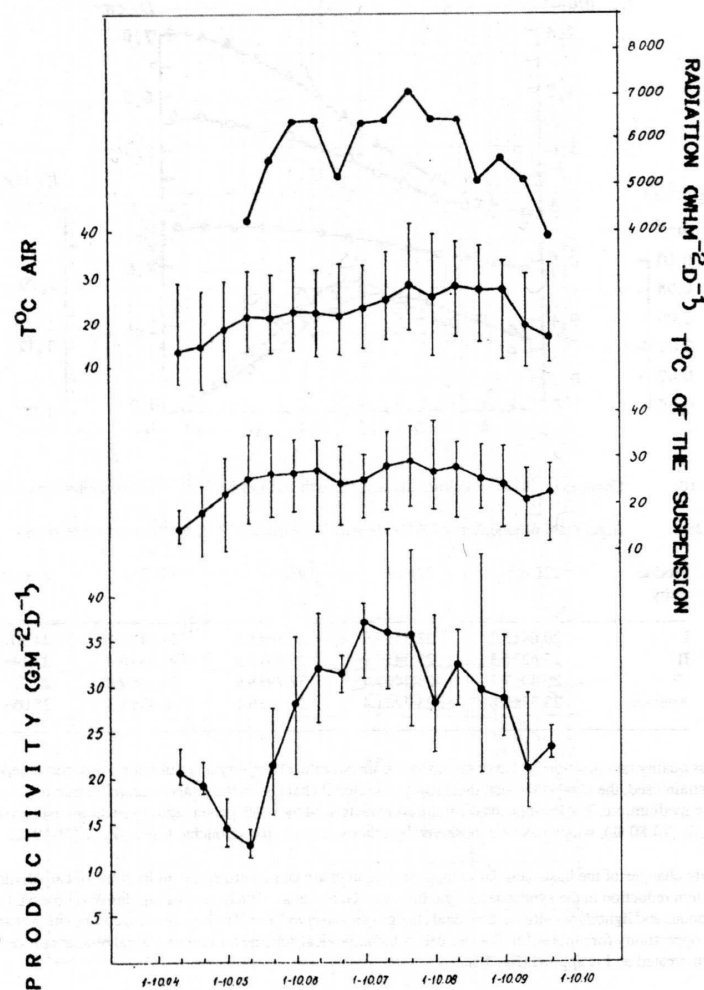


Figure 3. Seasonal changes of radiation, air temperature and suspension temperature (maximum, minimum and mean) and algae productivity (diversions are for the individual installations)

At optimum combination of external cultivation parameters the efficiency of light utilization may increase up to 15 % which means a potential daily yields up to 42g/m²/day [4].

The Roupi base is in operation from March to October and produces about 5-6 tons/year of dry product and the average annual productivity amounts to 20 – 23 g/m²/day.

1.3. Geothermal application in microalgal technology optimization

Different methods of algal technology optimization by geothermal energy, CO₂ and thermal water application have been discussed in [7,8]. They consist of:

- Use of geothermal water for nutrition algal media preparation.
- Use of geothermal CO₂ for optimizing photosynthesis.
- Use of geothermal energy for optimizing photosynthesis and algal biomass drying.

1.3.1. Use of geothermal water for nutrition algal media preparation

Photoautotrophic microalgae cultivation requires mineral nutrition media containing macro- and microelements [7]. The applied nutrition media are modified according to the mineral content of the geothermal water in order to provide optimal productivity of the growing algal species. Our exploration study showed that the mineral water of this field could be successfully used for cultivation of green algae (*Chlorella* and *Scenedesmus*) and blue-green algae *Spirulina* (*Arthrospira*) [7]. The use of thermal water chemical compounds reduces the expenses for purchasing of NaHCO₃, K₂SO₄, CaCl₂, MgSO₄ and some micro elements, provides conditions for optimal pH required for microalgal growth and increases the biomass productivity by 5%.

According to the standard requirements for a production of 1 kg of green microalgae is needed 0.77 kg of NaHCO₃. The cost of 1 kg of NaHCO₃ is about 0.25 EUR/kg. The annual production of the Roupi base amounts to about 6 tones of biomass and the cost savings reach to 1500 EUR. Also, the biomass growth increases by 300 kg and good physiological algae's state is guaranteed.

1.3.2. Use of geothermal CO₂ for optimizing photosynthesis

Like other phototrophically growing plants algae require an inorganic carbon source to perform photosynthesis. The right quantity of CO₂ provided to the algal cells can guarantee an intensive photosynthesis, a good physiological state.

In contrast to the conventional plants the algal species cannot be supplied with carbon by simple diffusion from the air, as CO₂ natural concentration (0.03%) is too low to sustain optimal growth and high productivity. Chemical analysis have shown that algal biomass consists of about 50% carbon, which means that about 2 kg of CO₂ are required to produce 1 kg of algae.

Desorption losses should not be excluded for the open mass cultivation because they reach up to 50 % of the carbon dioxide supply [1,2,3,5]. Therefore the minimum quantity of CO₂ necessary to produce 1 kg of biomass amounts to at least 3 kg.

According to the international prices 1 kg of CO₂ costs about 0.8-1.0 EUR that is about 2.4 - 3.0 EUR per 1kg of biomass. The utilization of CO₂ of geothermal origin in Roupi base reduces significantly the expenses for cultivation. Due to the insufficient amount of spontaneously released CO₂ in Roupi (about 27 – 30 kg/h) compression and storage in the reservoirs is required. Thus the only expenses associated with the geothermal CO₂ application are for the electricity use during compression and amount to 0.1 EUR per 1kg of biomass. In

other cases when the freely released CO₂ quantity is high enough, like in Nigrita field (N.Greece) - 600kg/h, there are no expenses for its supply to the cultivation ponds.

1.3.3. Use of geothermal energy for optimizing photosynthesis and algal biomass drying

The intensity of photosynthesis, respectively microalgal growth and productivity depends strongly on temperature [1,2,3,5]. Regardless of the individual differences between the various algal species, temperature requirements for algal growth have shown that a raise of the cultivation temperature up to 26°C intensifies considerably the biomass synthesis [9].

Open mass algal cultivation is associated with high day-and-night and seasonal alterations of temperature and solar radiation. It is quite clear that during the early morning hours (7:0 - 10:0 a.m.), solar radiation suffices an intensive photosynthesis but temperature is a restrictive factor. The span of these suboptimal temperature conditions differs during the cultivation season. The temperature increase in the morning hours with only 4-5°C (to reach 24-26°C) and sustaining an optimal night temperature (at least 15°C) will lead to: early spring inoculation of the algal ponds (March-April), prolongation of the cultivation season to the end of October, synchronized cells development and yield increase by 20%.

A geothermal heating device was built for the small cultivation ponds of area 6m² and 225m². As soon as it was introduced the suspension temperature reached its optimum for algal growth even in March and April. Another possible application of geothermal energy is for biomass drying, especially when the water temperatures are high (more than 70°C) and algal suspension density is more than 20%. Such drying devices are not still applied in the practice.

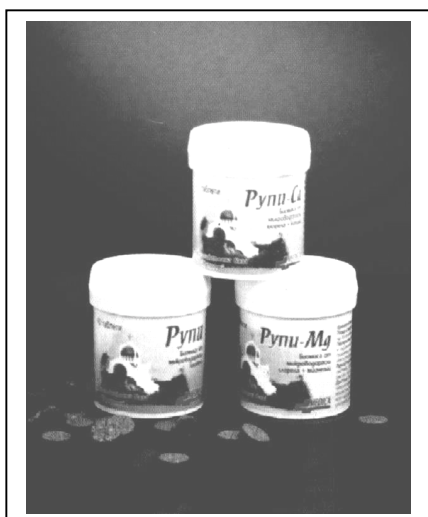
2. Application of microalgal biomass

Microalgal biomass has a very high protein content (more than 50%) with well-balanced amino acid composition, rich mineral content (Fe, Se, Zn, Ca, Mg, etc), vitamins, antioxidants (carotenoids), phycobiliproteins, essential fatty acids and polysaccharides.

Four basic products are obtained from algal biomass: water extract, alcohol extract, hydrolyzed biomass and protein hydrolysate. The alcohol extract is further fractionated to phospholipids, carotenoids, phytol, sterols, fatty acids and chlorophyllines. These products have been standardized and preparations have been developed for cosmetics (Algobad, Roupi Shampoo, Algofem, etc), for human (Roupi tablests, Algomed, Nephromed, Protalmin) and veterinary medicine (Algovit),etc., Figure 4.

Conclusions

- The existing base in Rupi region (SW Bulgaria) for algal biomass cultivation and processing with the use of geothermal water is a successful example for the realization of the idea of industrial photosynthesis.
- The use of geothermal carbon dioxide, thermal water and geothermal energy reduces the total price of the algal biomass up to 40 %.
- Various new and useful products for cosmetics and medicine have been introduced to the market.



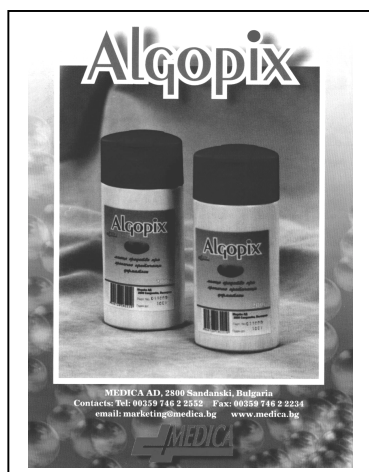
Food supplements (tablets)



Tablets (ulceroprotectivium)



Cosmetic series Medica



*Medicine shampoo (Psoriasis
 Vulgaris, eczemas, seborrea)*



Cosmetic series ALGITA

Figure 4. Bulgarian microalgal products at the market

References

1. A.Richmond,E.W.Becker, *CRC Handbook of Microalgal Mass Culture*, Boca Raton,Fl.(Ed) A.Richmond, 1986, pp.527
2. E.W.Becker, *Microalgae: Biotechnology and Microbiology*, (Eds) J.Baddiley at al.,Cambr.Univ.Press,1994, pp.290
3. Ch.Dilov, *Mass cultivation and use of microalgae*, Bulg.Acad.Sci.Publ.House,S. 1985, pp.194
4. L.Tsoglin,S.Avramova,A.Gabev,V.Semenko, *Sov.Plant Physiol.*27(3),p.644-652 (1981)
5. M.A.Borowitzka,L.J.Borowitzka, *Microalgal Biotechnology and Microbiology* (Eds),Elsevier Appl.Sci. New-Yourk,1988, pp.477
6. S.Fournadzhieva,P.Pilarsky,A.Gabev, *Proceedings of Ist European Workshop on Microalg.Biotech.Algology*,Potsdam-Rehbrucke,10-12.06,p.93-104 (1992)
7. S.Fournadzhieva,D.Georgiev,M.Bozhkova, *Proceedings Geothermal Energy,Technol.,Ecol.*, Bansko, 30.05-6.06.1993,K. Popovski et al.,(Eds),(24) 3-11,(1993)
8. S.Fournadzhieva,G.Petkov,P.Pilarski,R.Andreeva, *Proceedings of Int.Geothermal Days,Direct Utiliz.of Geotherm.Energy*,K. Popovski et al.,(Eds),Oregon,USA,175-179,(1999)
9. S.Fournadzhieva,K.Bojadgieva,M.Fytikas,K.Popovski, *Proceedings Int.Geothermal days"Greece"* Thessaloniki,1-4.09.2002,p.152-159,(2002).