Commercial Success
Dunaliella
Israel and Japan Collaboration

Namibia - Sanumarc
June 4, 2010

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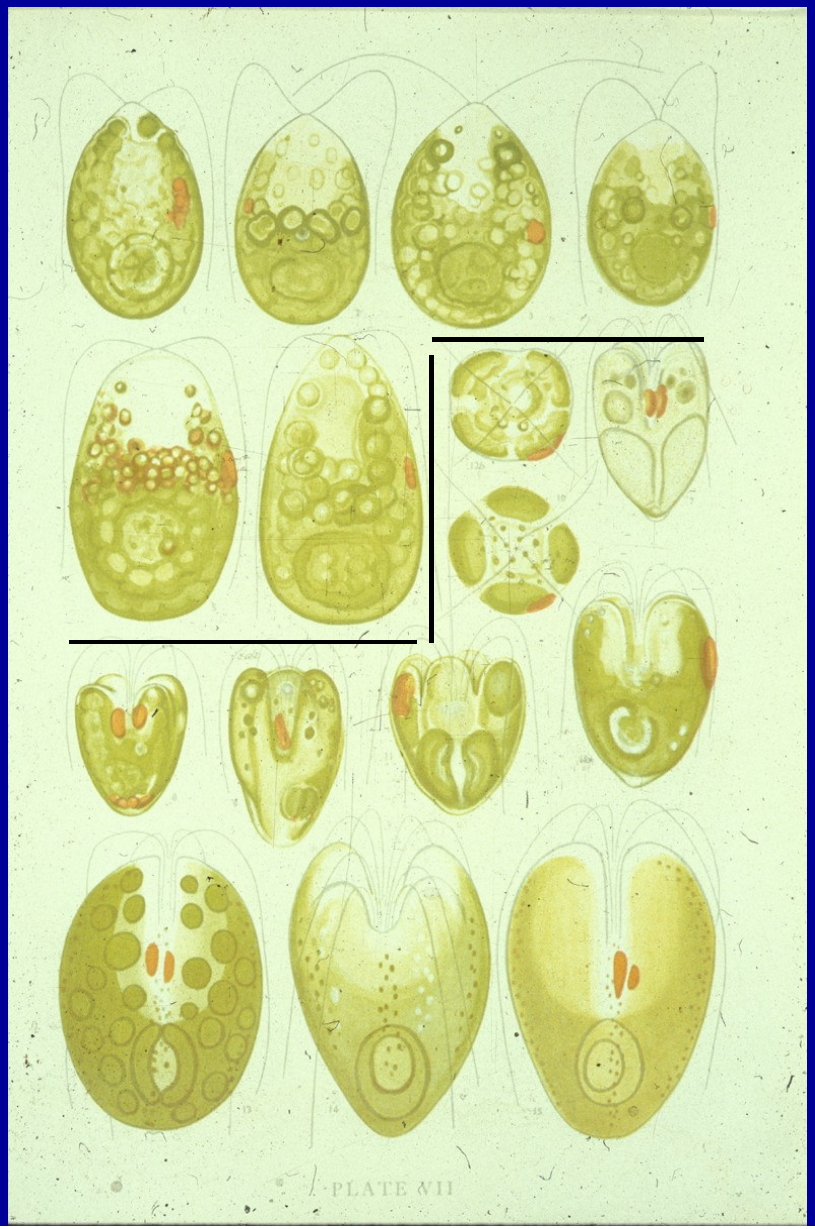
Dunaliella

Introduction

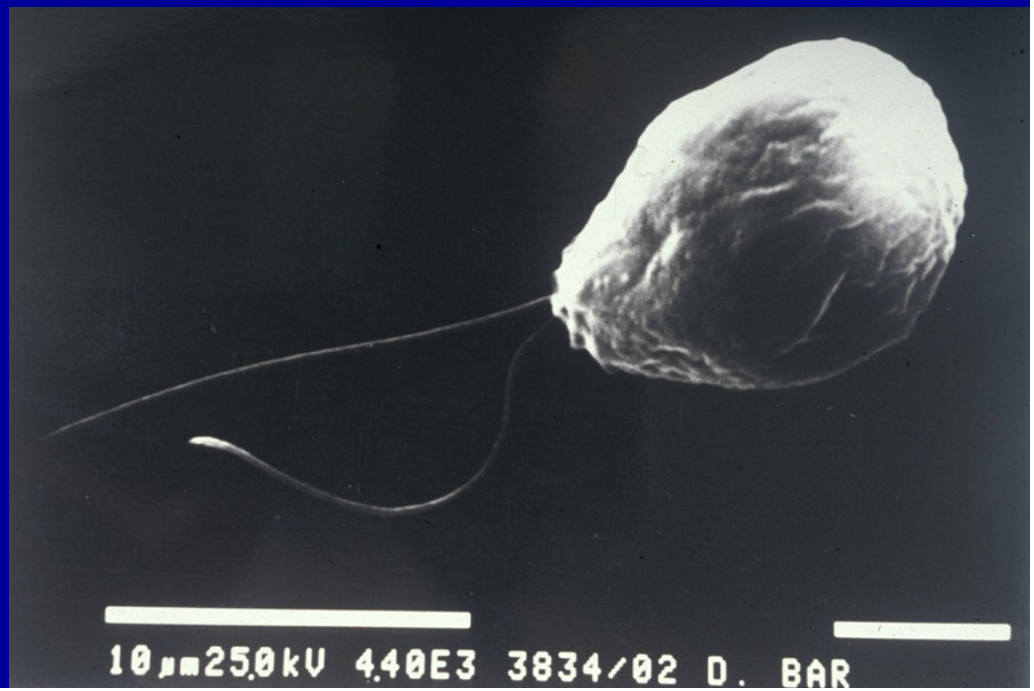
&

Background

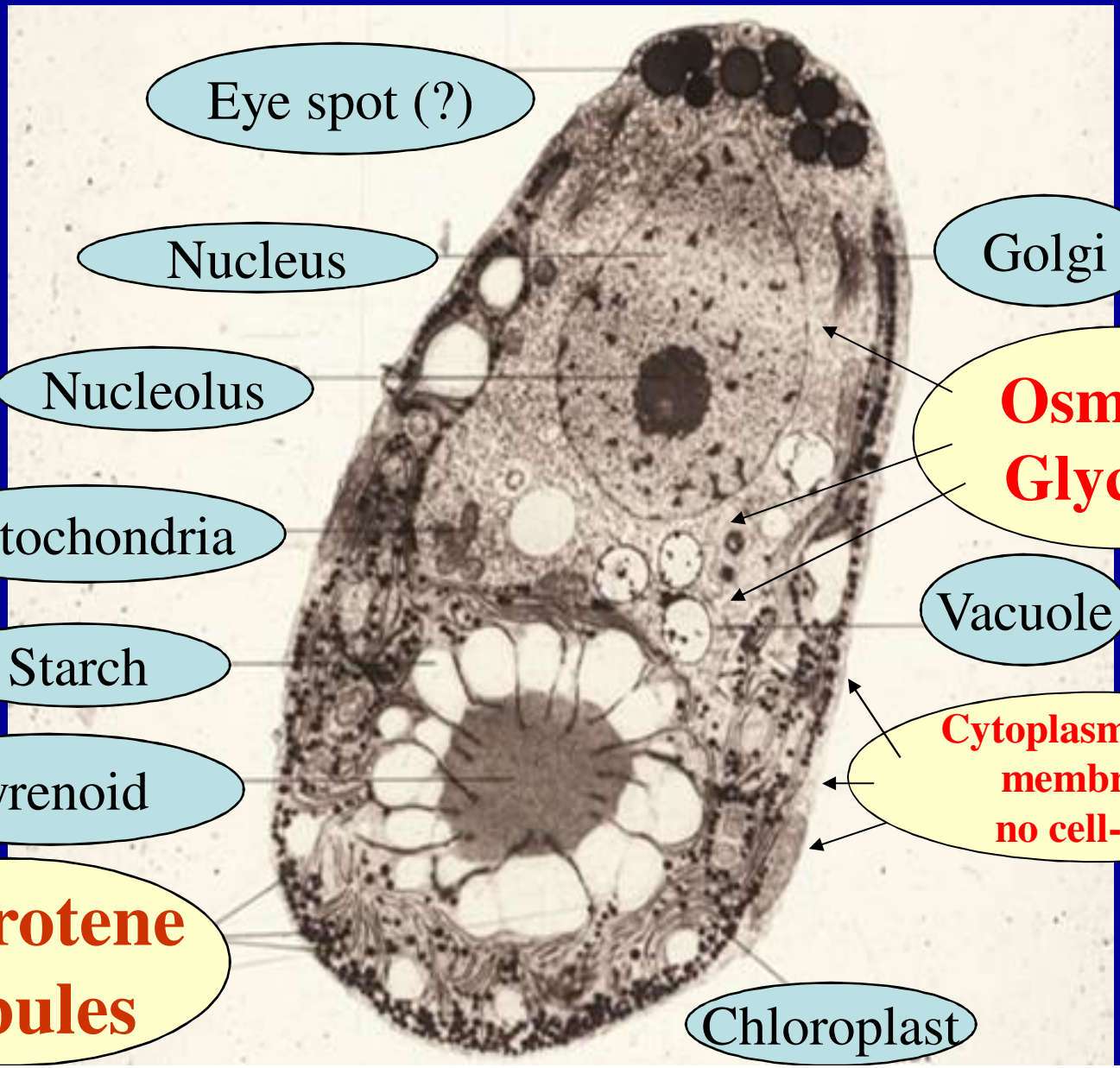
Dunaliella, Chlorophtya, Volvocales



Marine unicellular alga

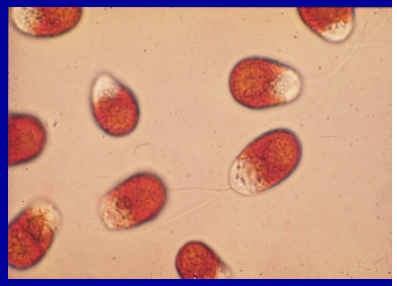


Dunaliella


Eye spot (?)
Nucleus
Nucleolus
Mitochondria
Starch
Pyrenoid
β-Carotene globules
Chloroplast
Golgi
Osmotic Glycerol
Vacuole
Cytoplasm elastic membrane, no cell-wall

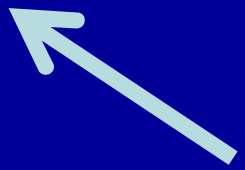
Natural Bloom of *Dunaliella* Extreme Halotolerant Alga

Surviving in saturated salt (>32%)



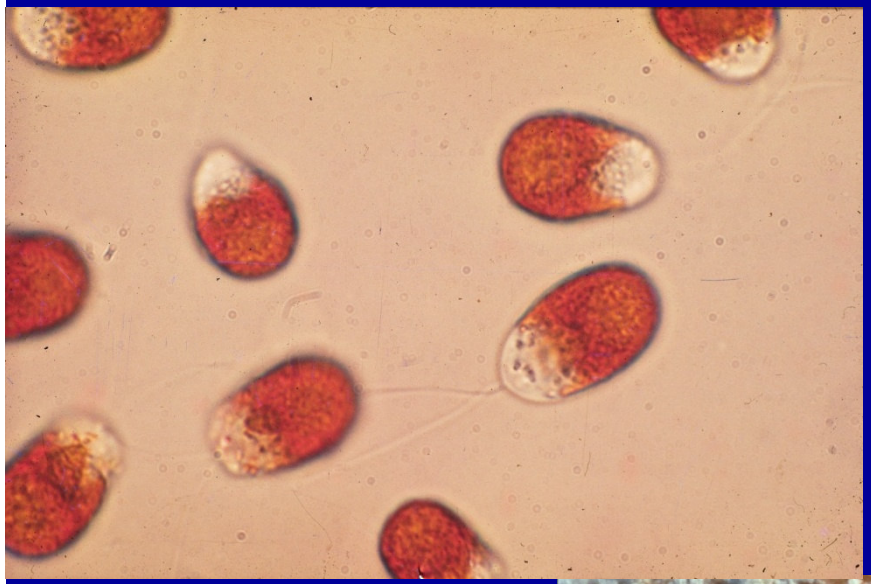
Accumulating osmotic cellular glycerol (>60%)

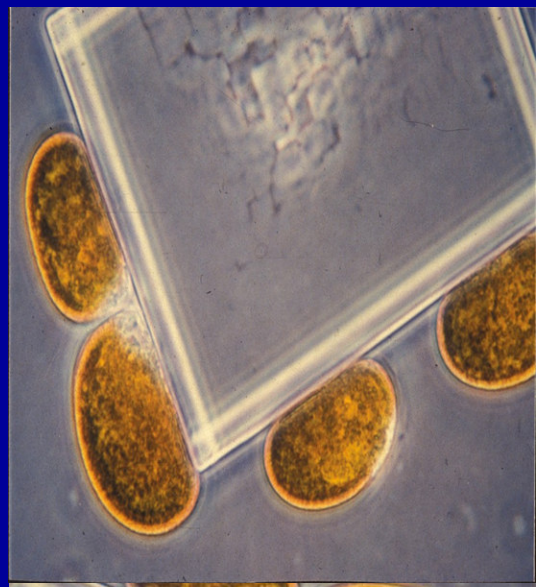
to the Great Salt Lake



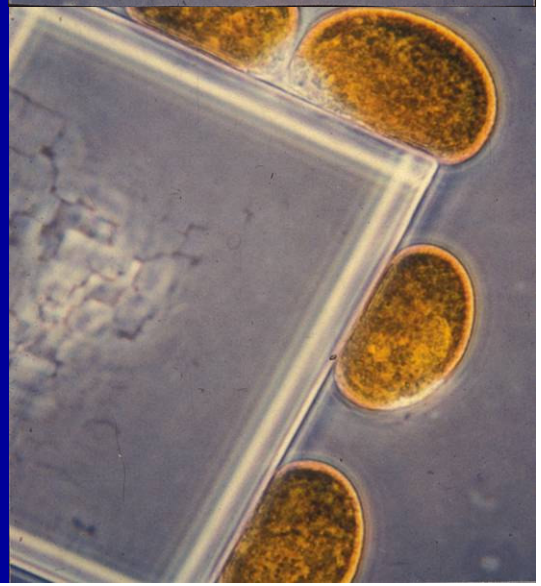
From Sea Water

Halotolerant Algae





Surviving in saturated salt (>32%)



Osmotic Adaptation

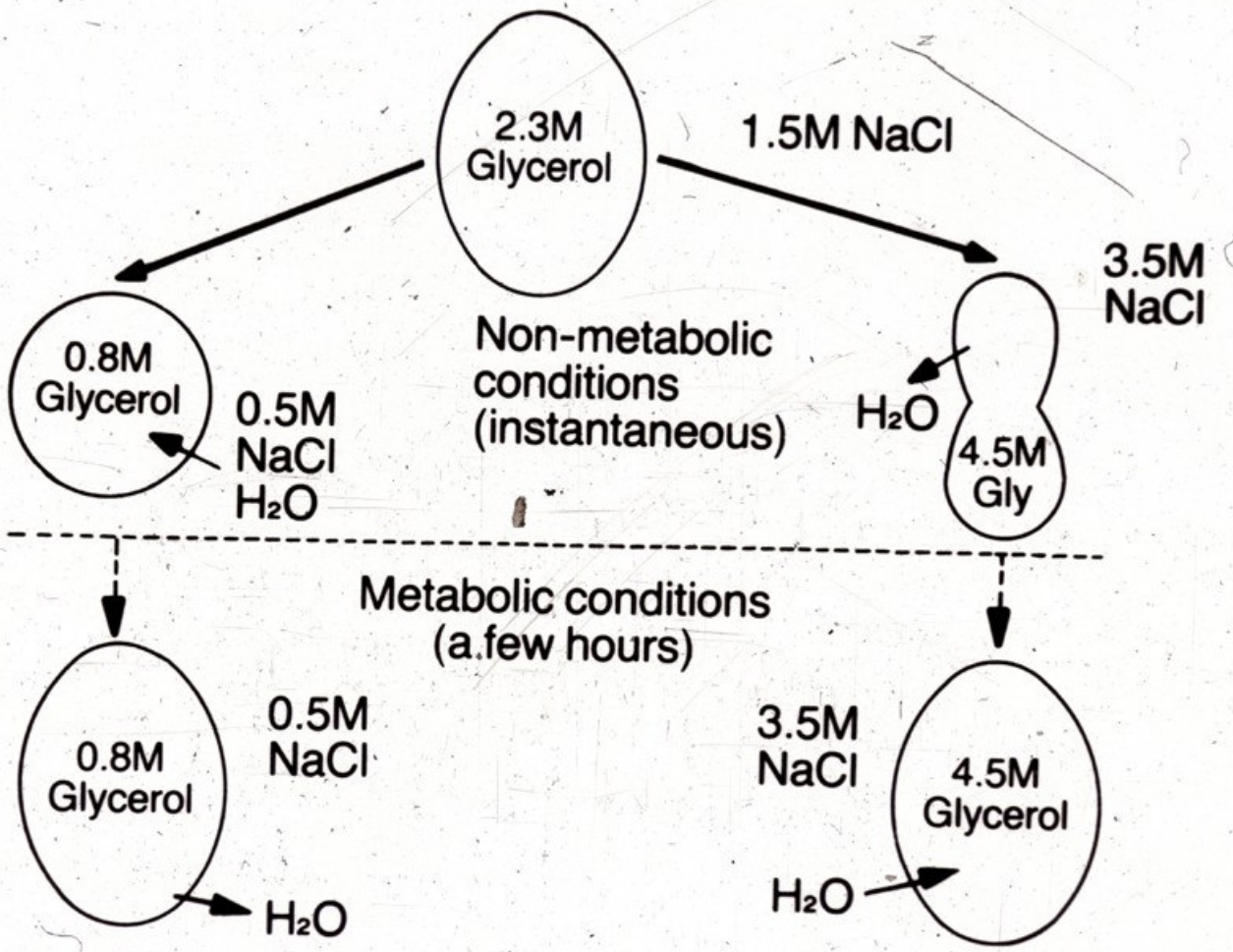
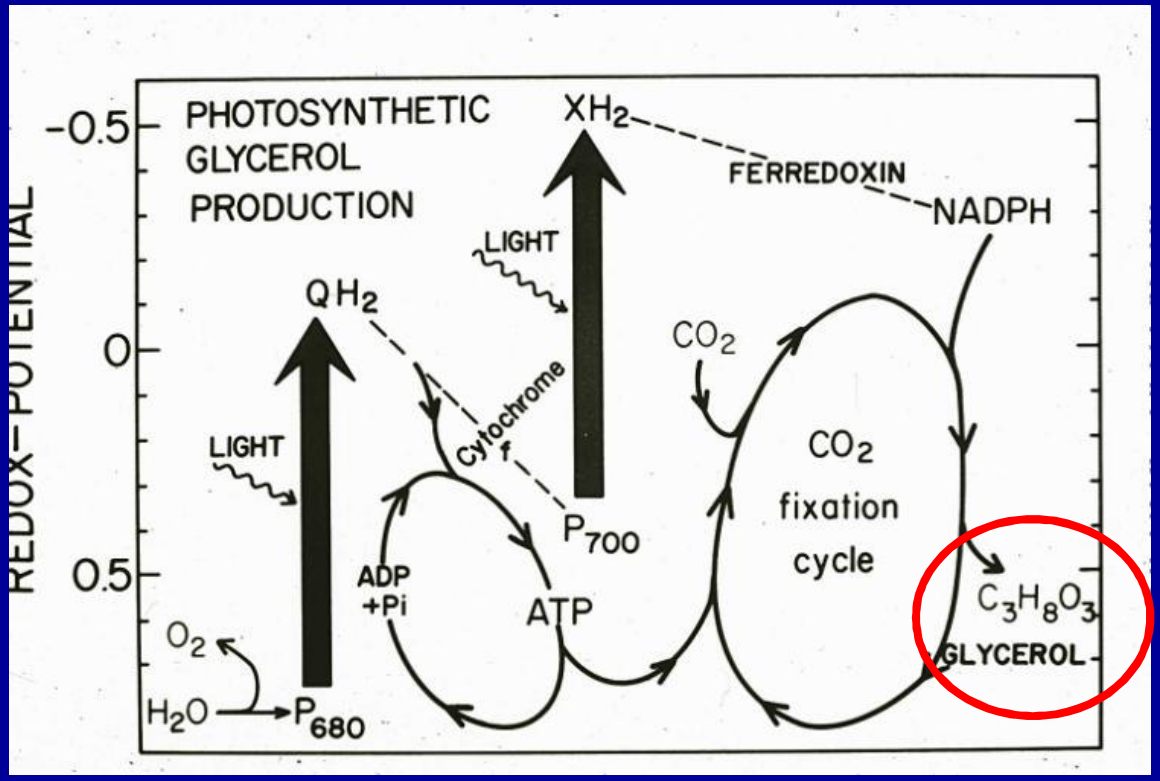
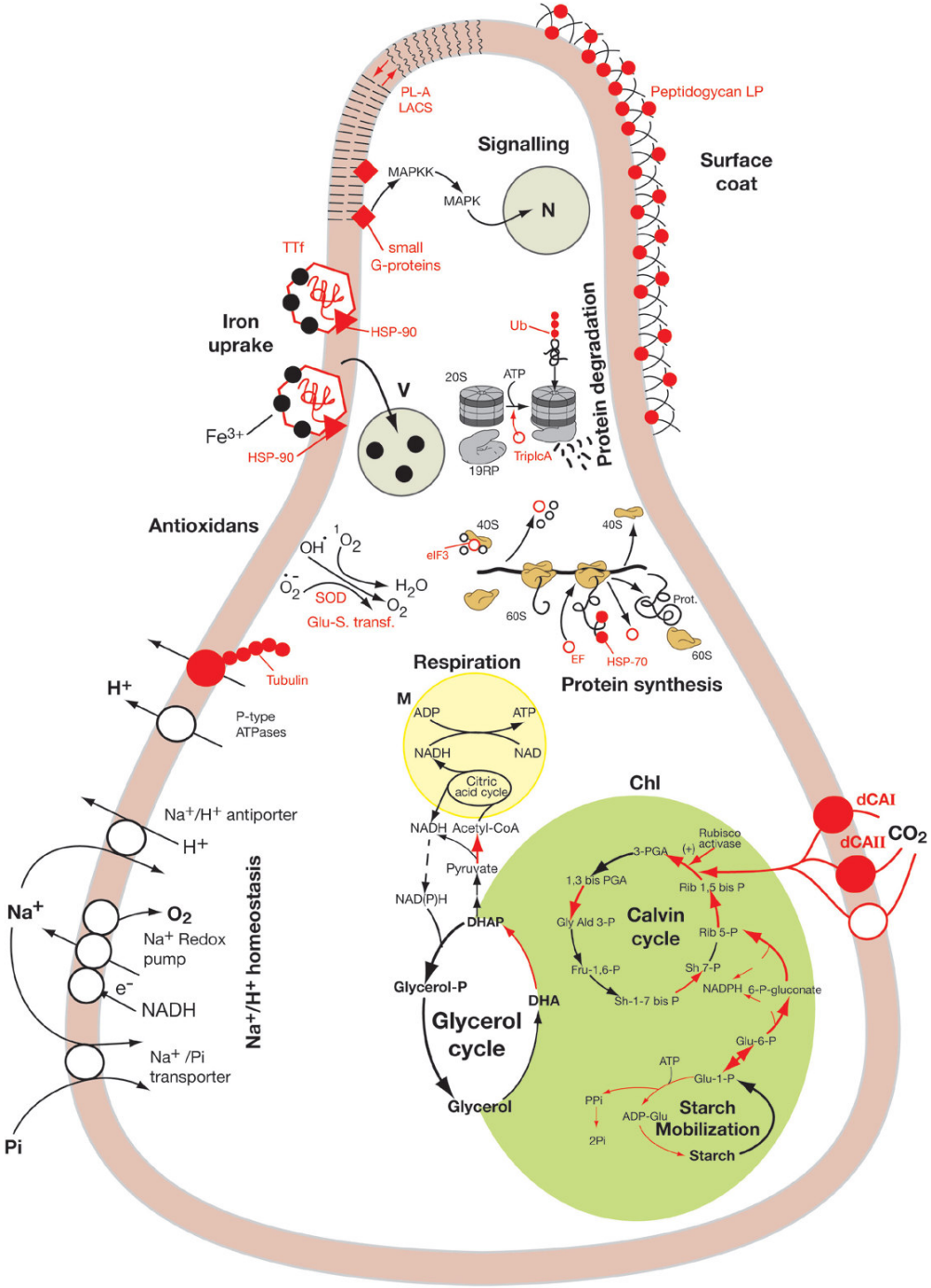


Fig. 4. Schematic representation of the volume changes of Dunaliella *in response to hypertonic or hypotonic conditions.*

Photosynthesis in *Dunaliella* Sugar & Glycerol Biosynthesis

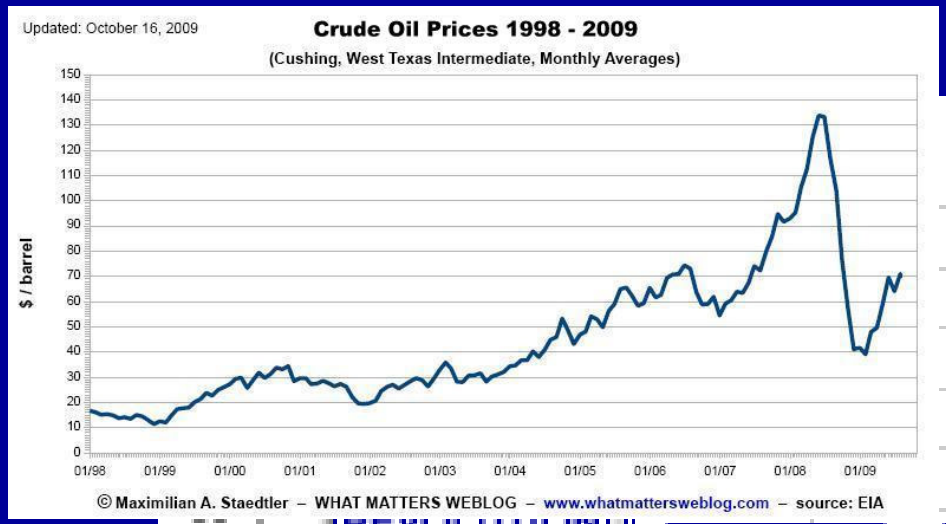


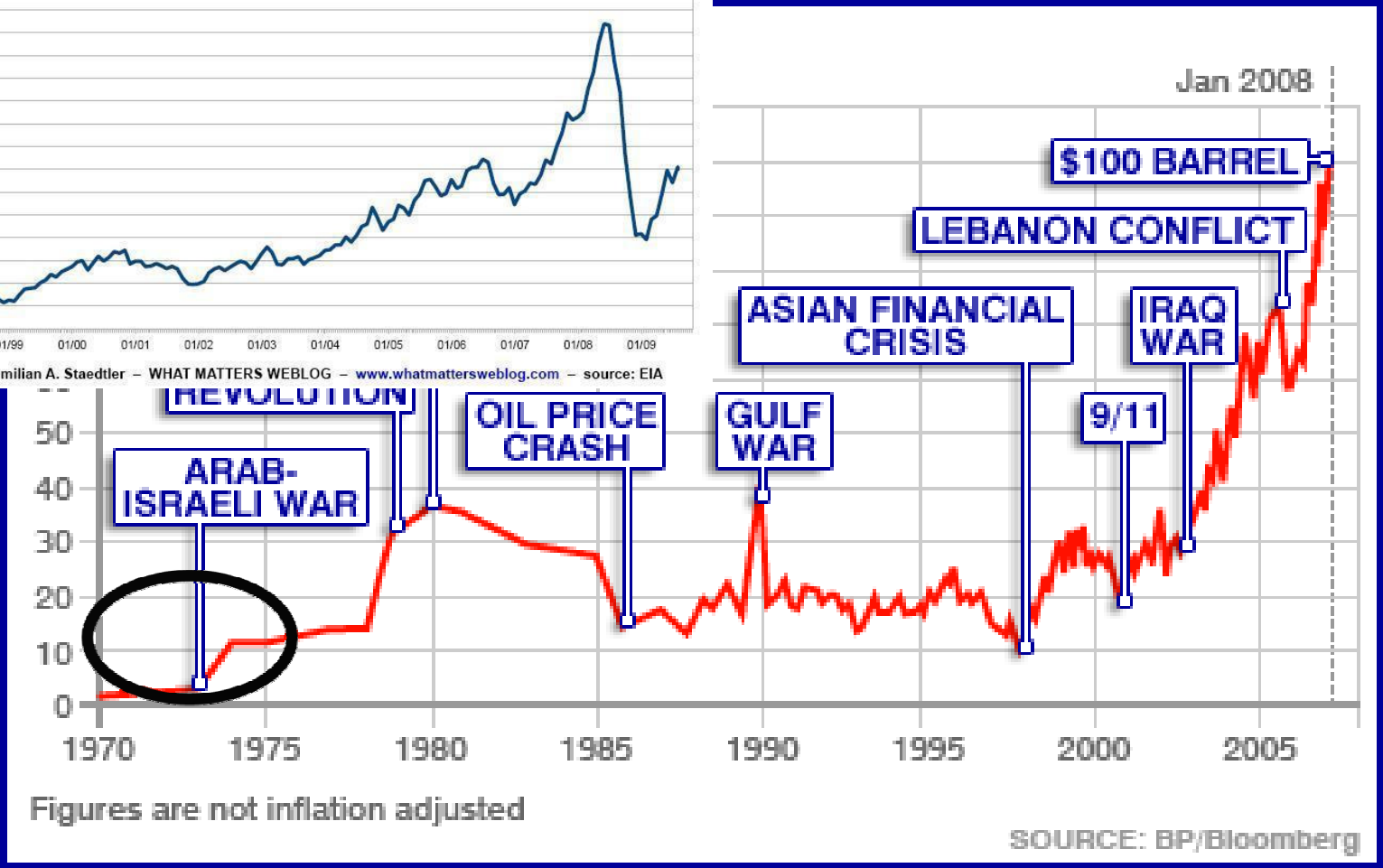
Halotolorance in *Dunaliella*
Katz et al., 2009



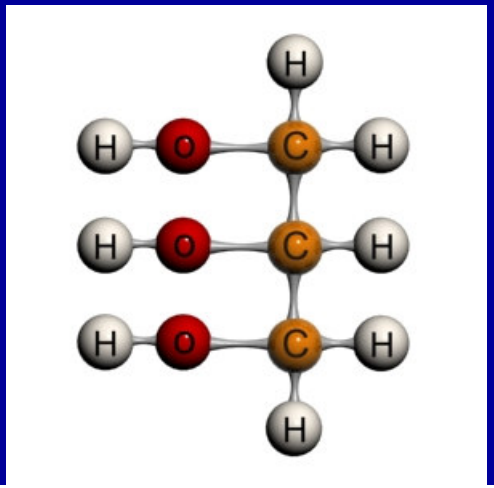
Oil Price and Wars

Feasible algae for Energy at above $100/barrel (1975)

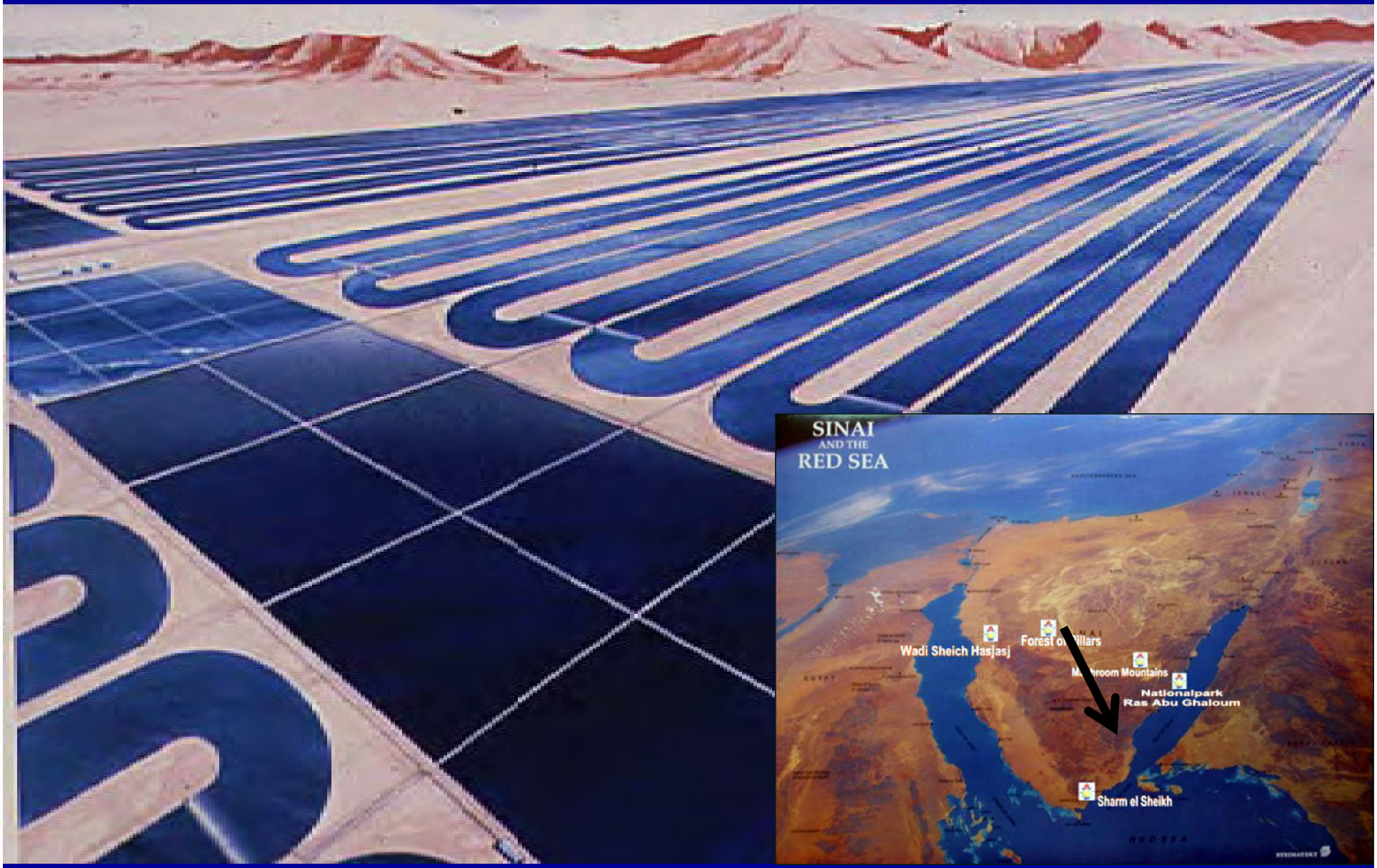




The 1st Energy Crisis
1973-5
Algae for Energy
Large scale production of
Glycerol
by
Dunaliella



1st Energy Crisis 1975, Glycerol Algal Farm,
500 hectares in Sinai

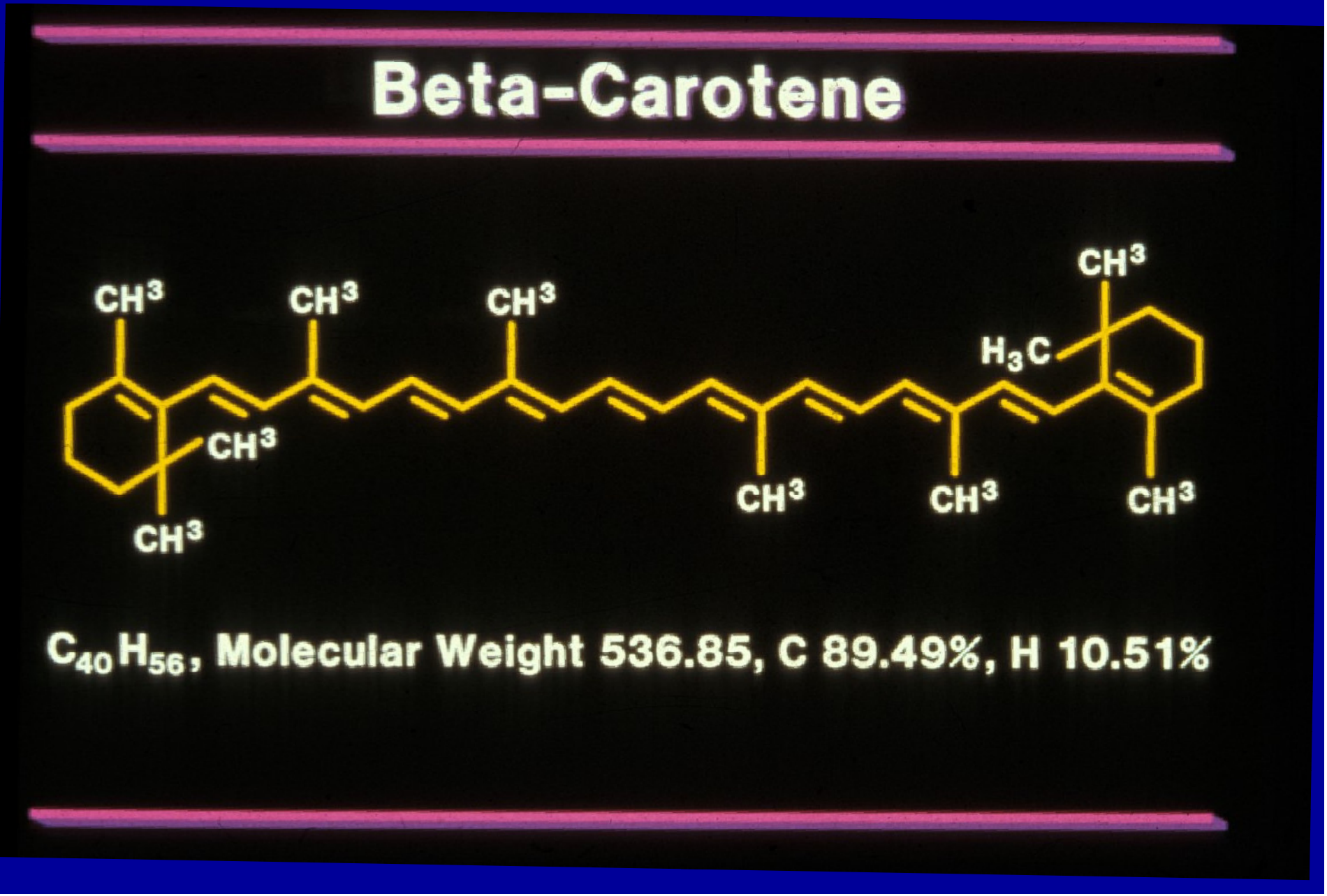


Algae & Oil Price
1975
oil price dropped down
Dunaliella Project was stopped

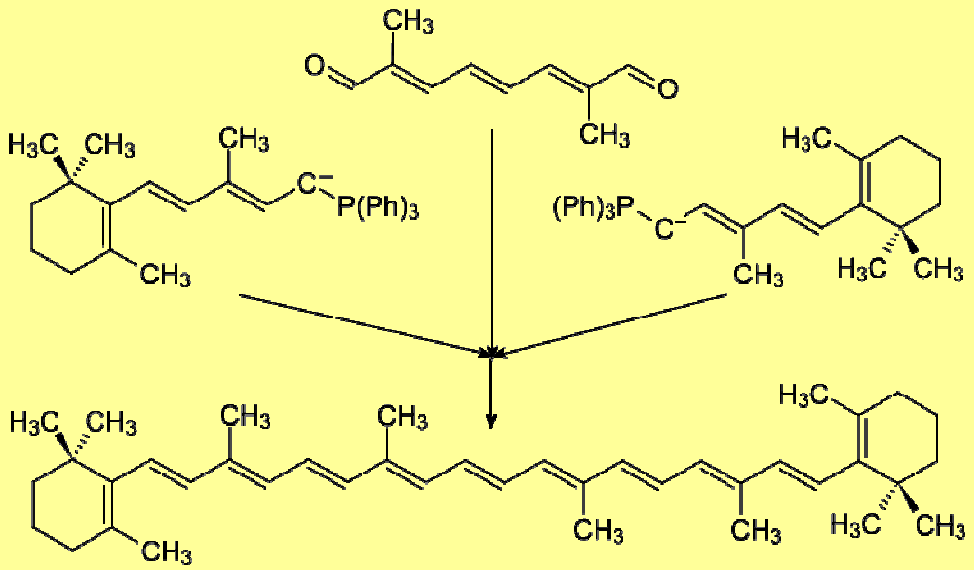
Until then
Search for High Value Algal Products

Dunaliella High Value Products

all-trans β-Carotene



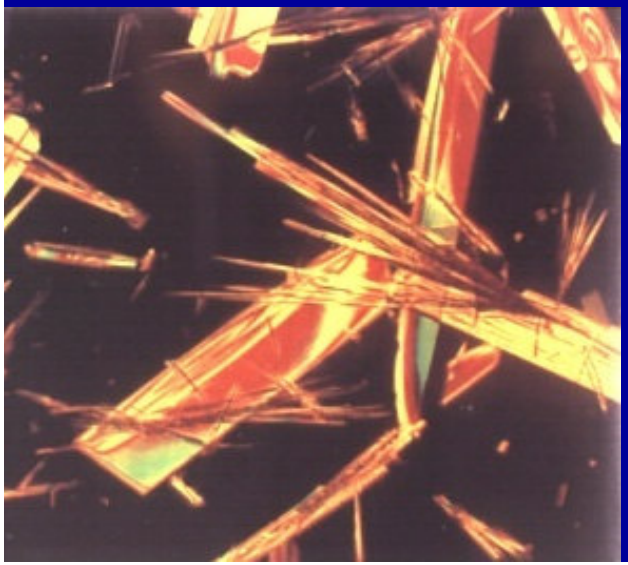
Chemical Scheme of Carotenoids Synthesis all-trans β-Carotene, BASF & Roche ~1950

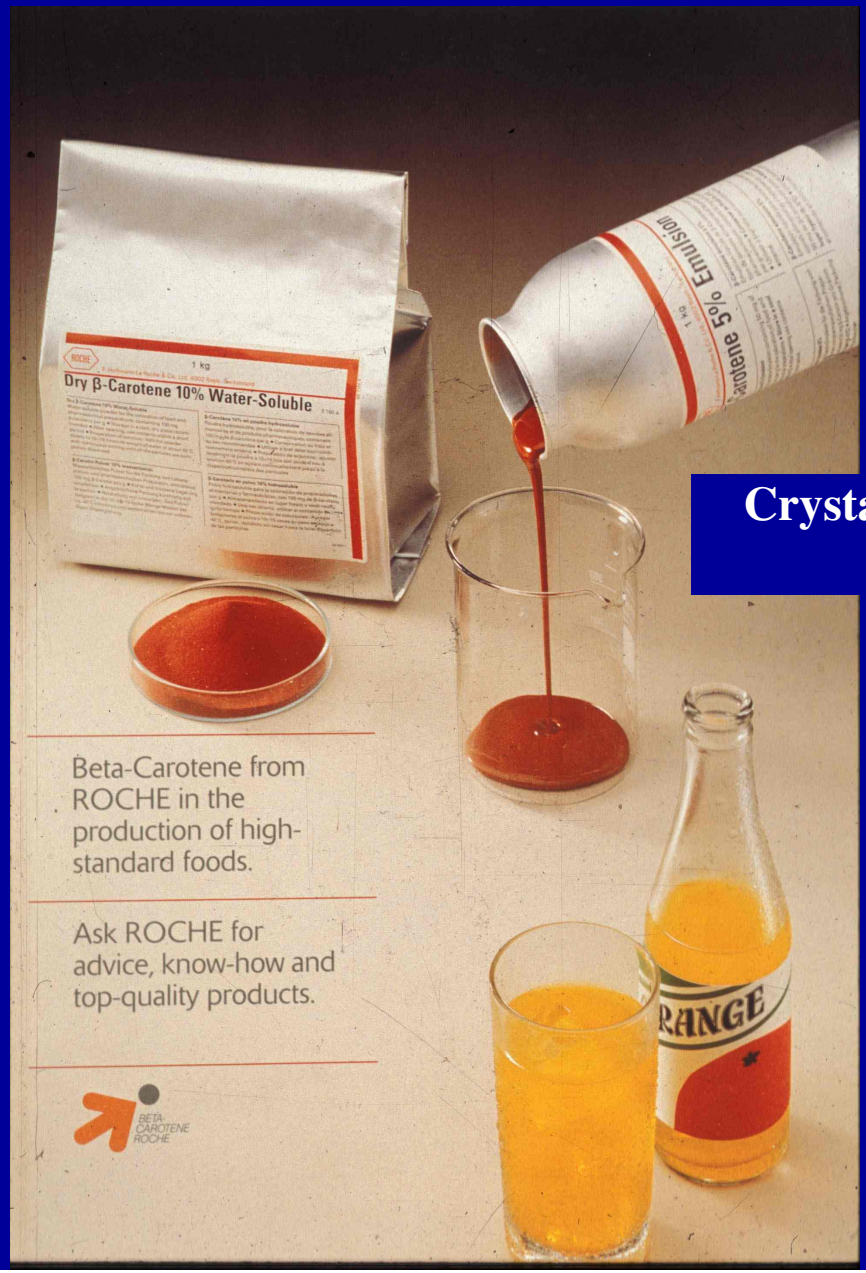


all-trans β-carotene

Synthetic all-trans-β-Carotene

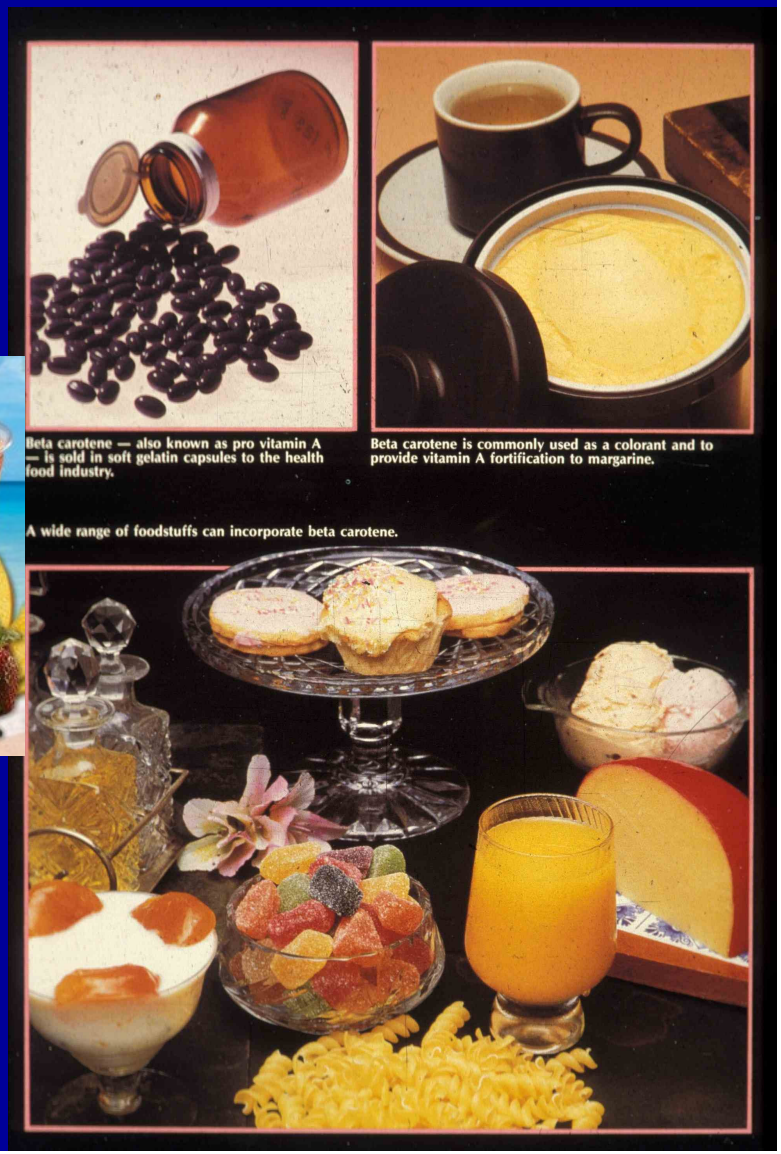
Low fat solubility



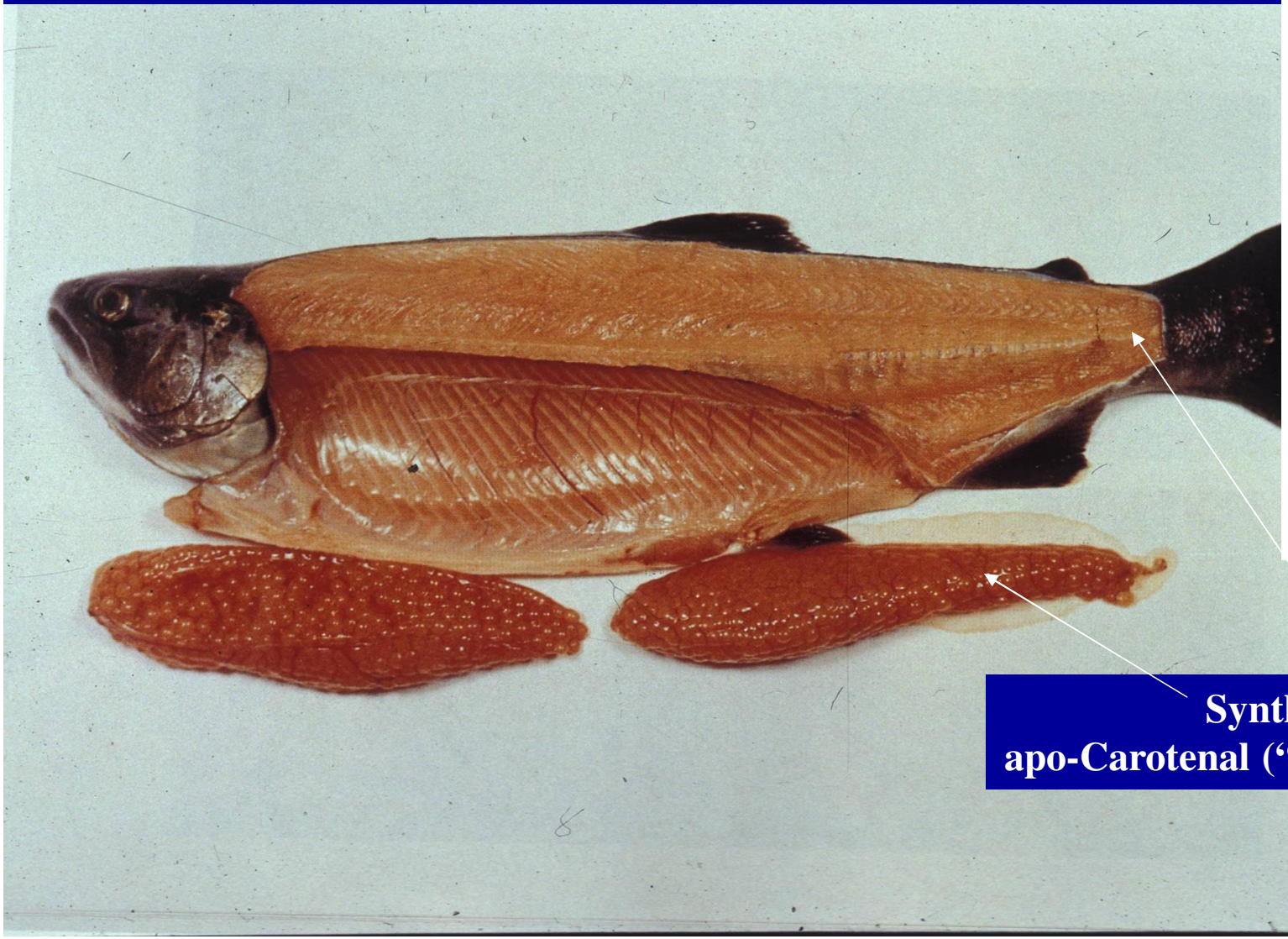


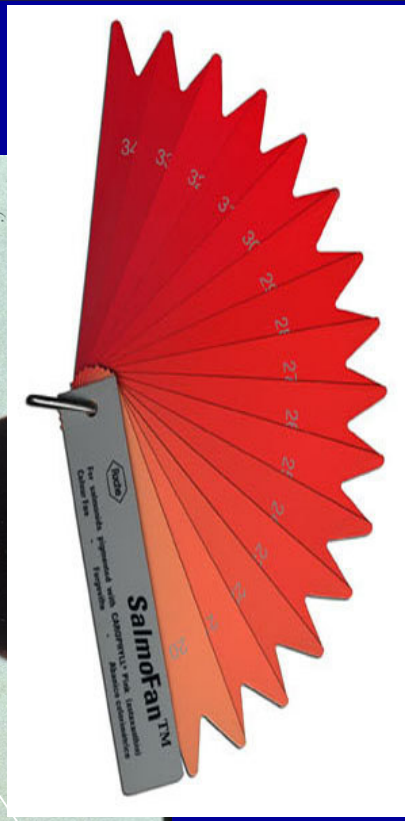
Crystals & Oil emulsion

Colored Food
Synthetic all-trans-β-Carotene



Colored Fish





Synthetic
apo-Carotenal (“Carotene Pink”)

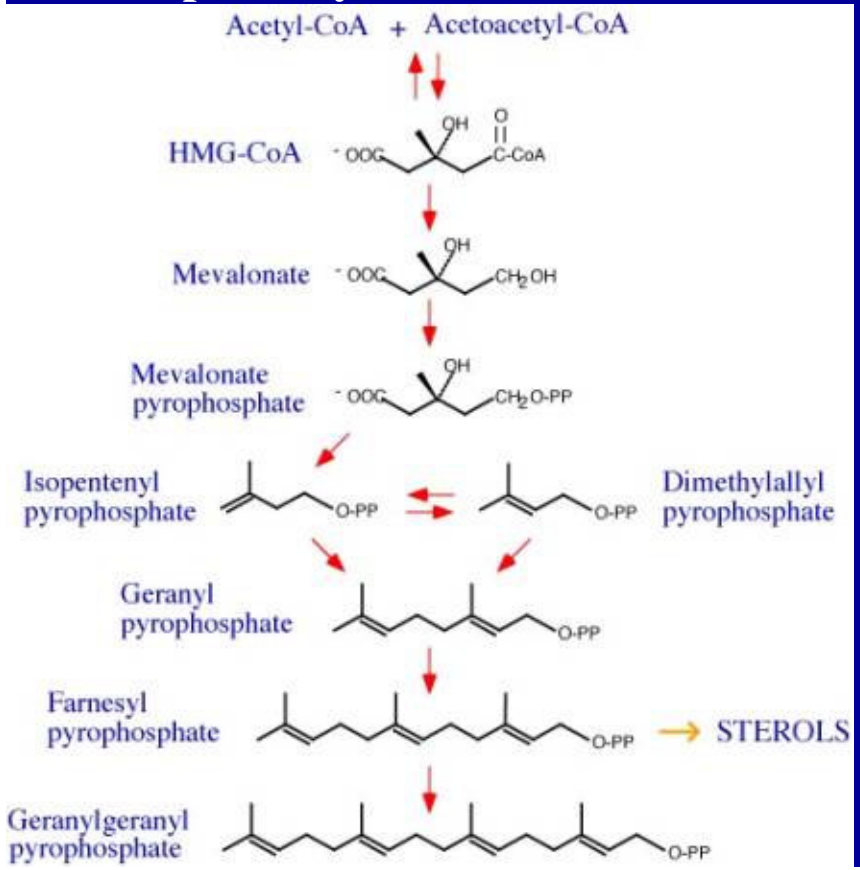
Colored Eggs

Synthetic
apo-Carotenal

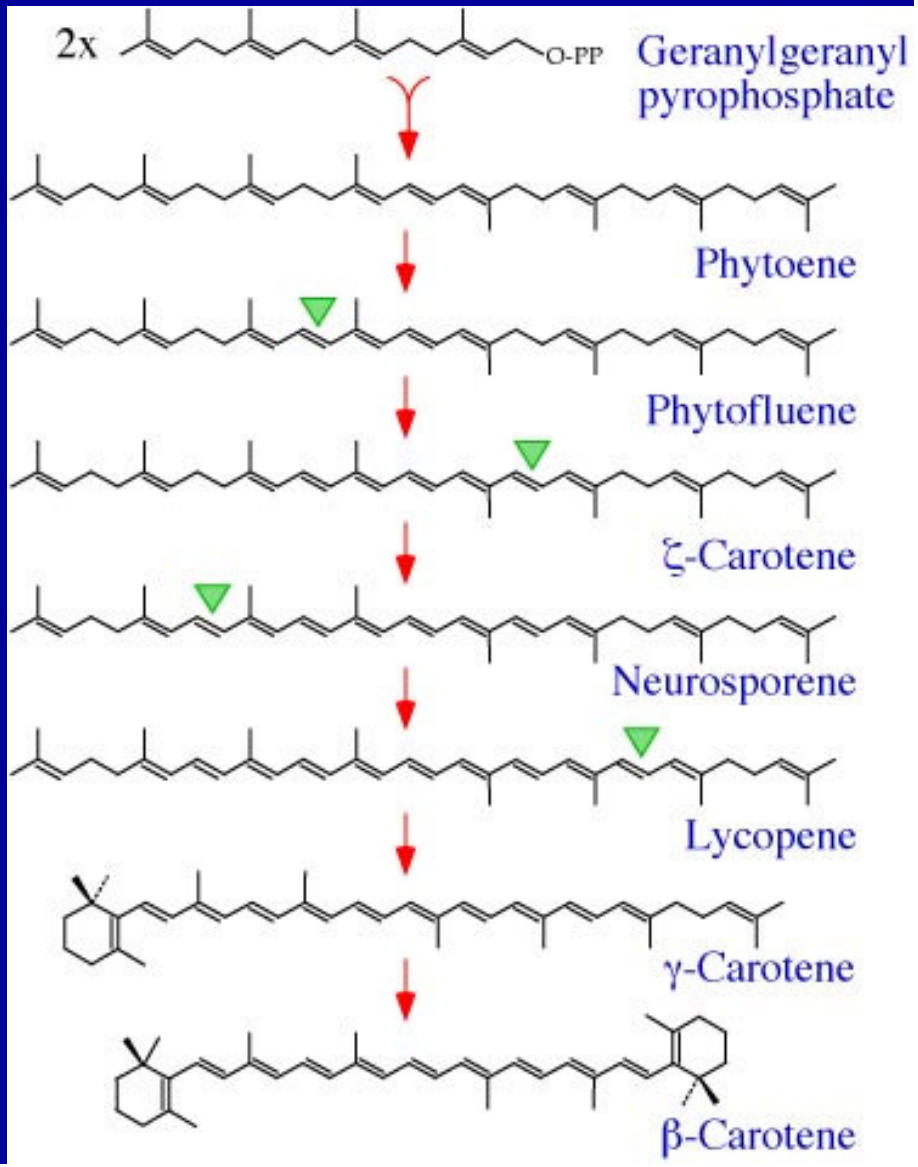
Natural Carotenoids

BIO-Synthesis of Carotenoids

Polar pathway



Lipophilic pathway



Dunaliella Research (nutrient optimization studies of 2x2 & 3x3)
The Weizmann Institute

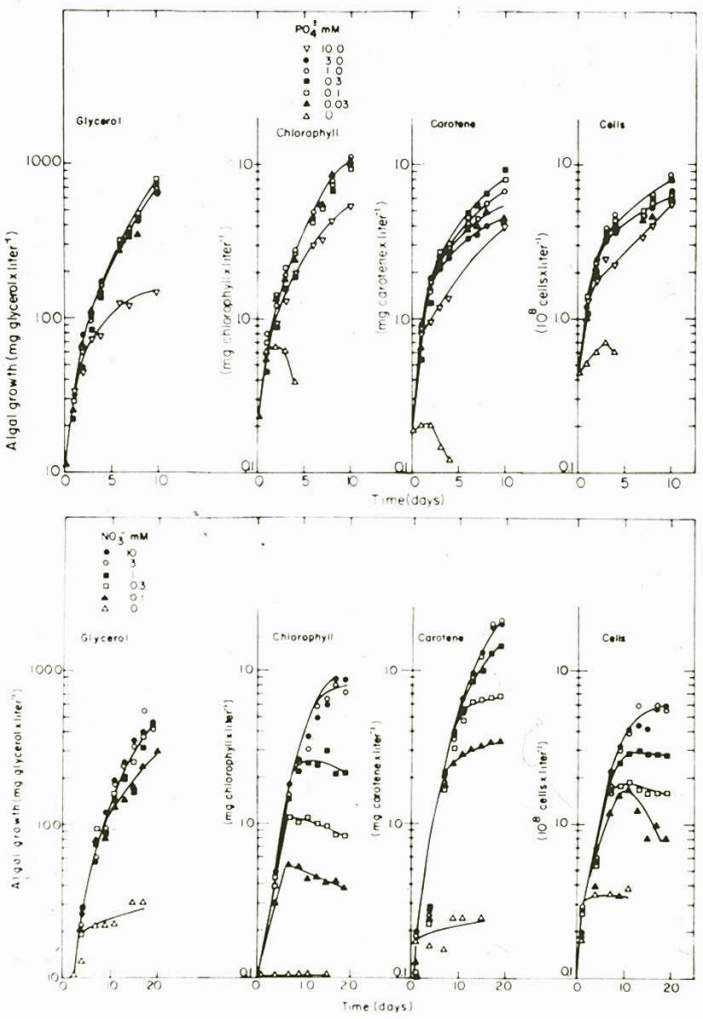
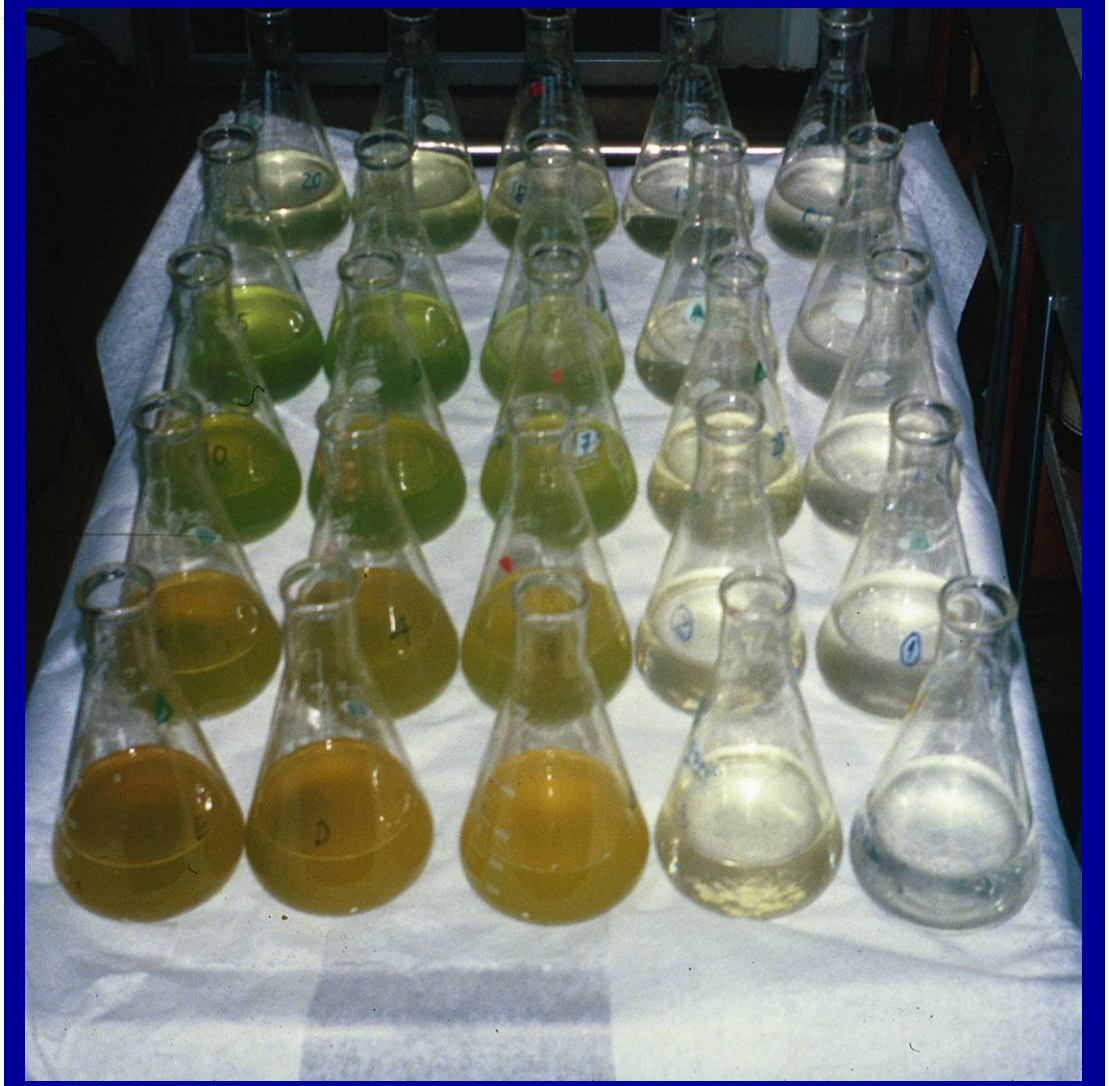
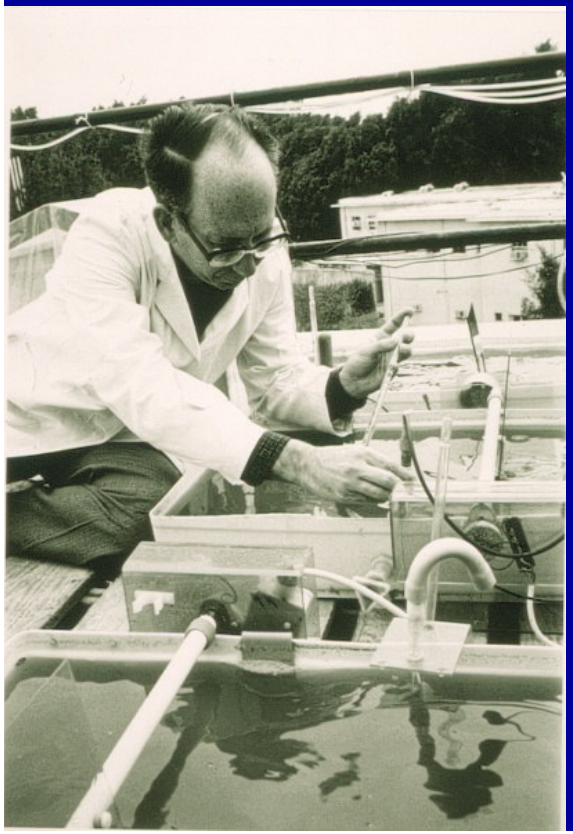


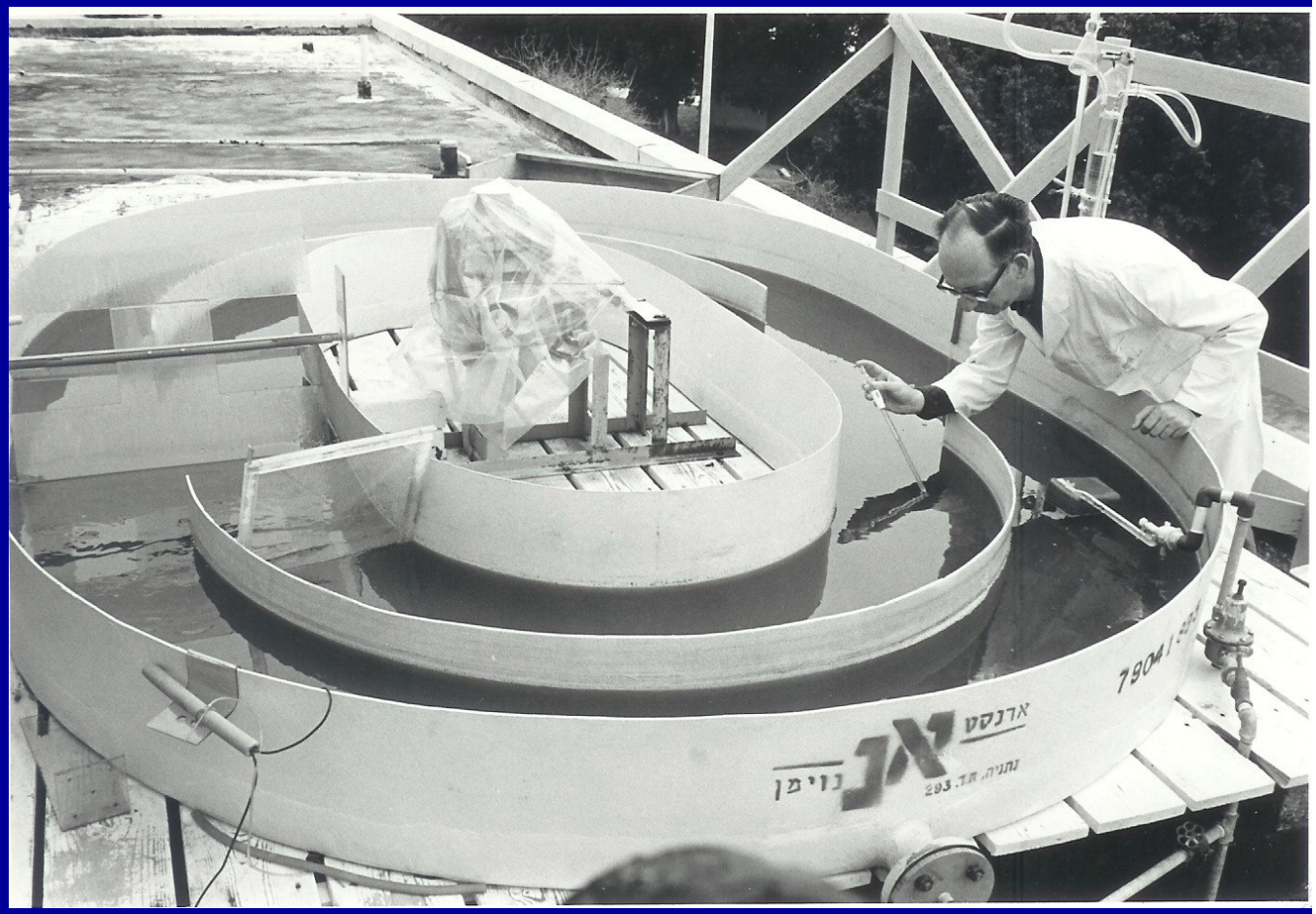
FIG. 3. Effect of phosphate concentration (upper) and nitrate concentration (lower) on the growth of *D. bardawil* and its content of glycerol, chlorophyll and carotene. Algae were preconditioned indoors on a nitrate or phosphate free medium containing 3 M NaCl until growth was retarded, then transferred to media with the indicated nitrate or phosphate concentrations.



The Weizmann Institute
Indoors & outdoor studies

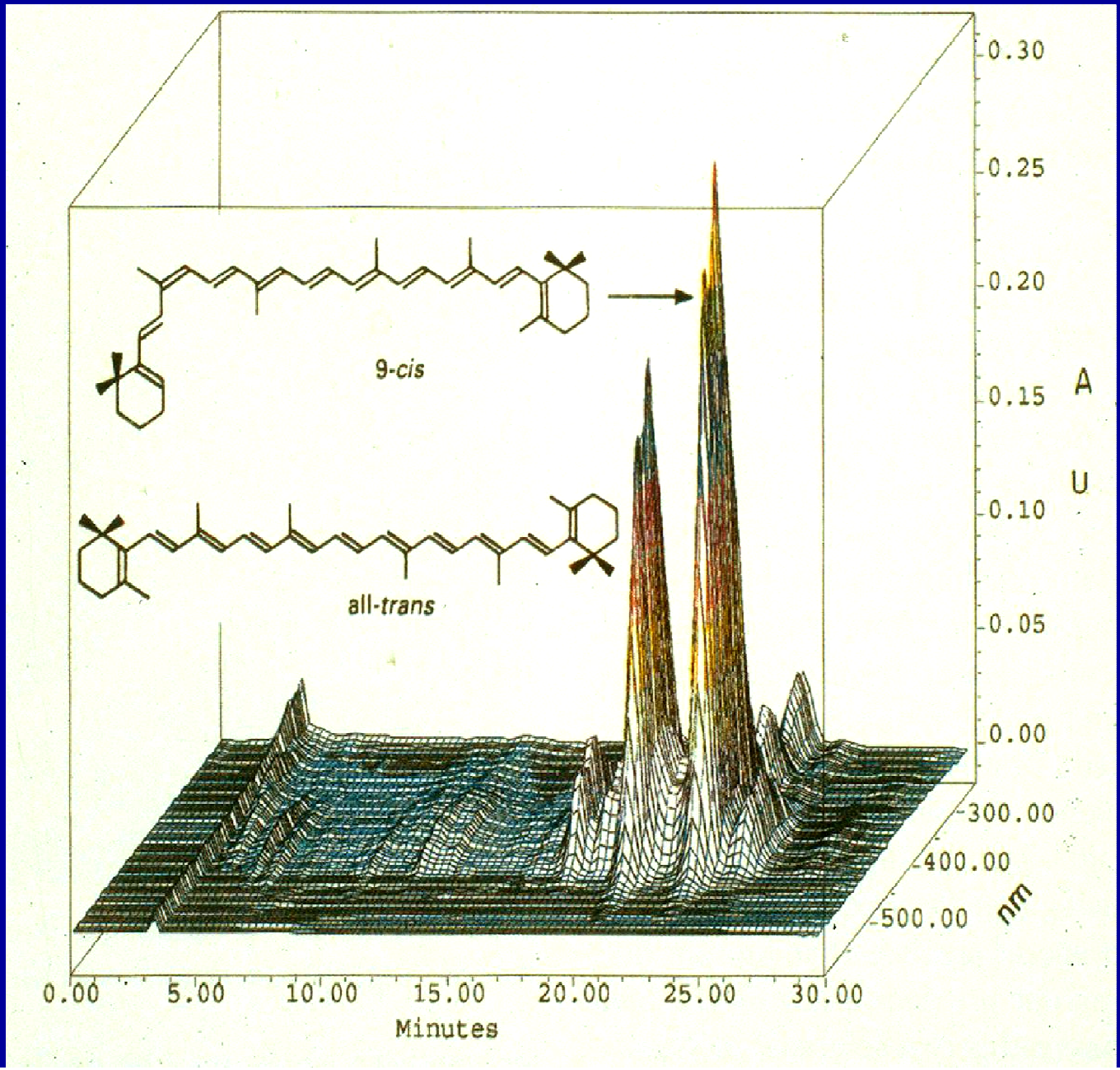
The Weizmann Institute *Dunaliella* Research, (1975-1991, the Late Prof. M. Avron)





The Weizmann Institute *Dunaliella* Research (outdoor biotechnology)

Natural Carotenoid Isomers (HPLC Analysis)



Biosynthesis of β-Carotene Isomers

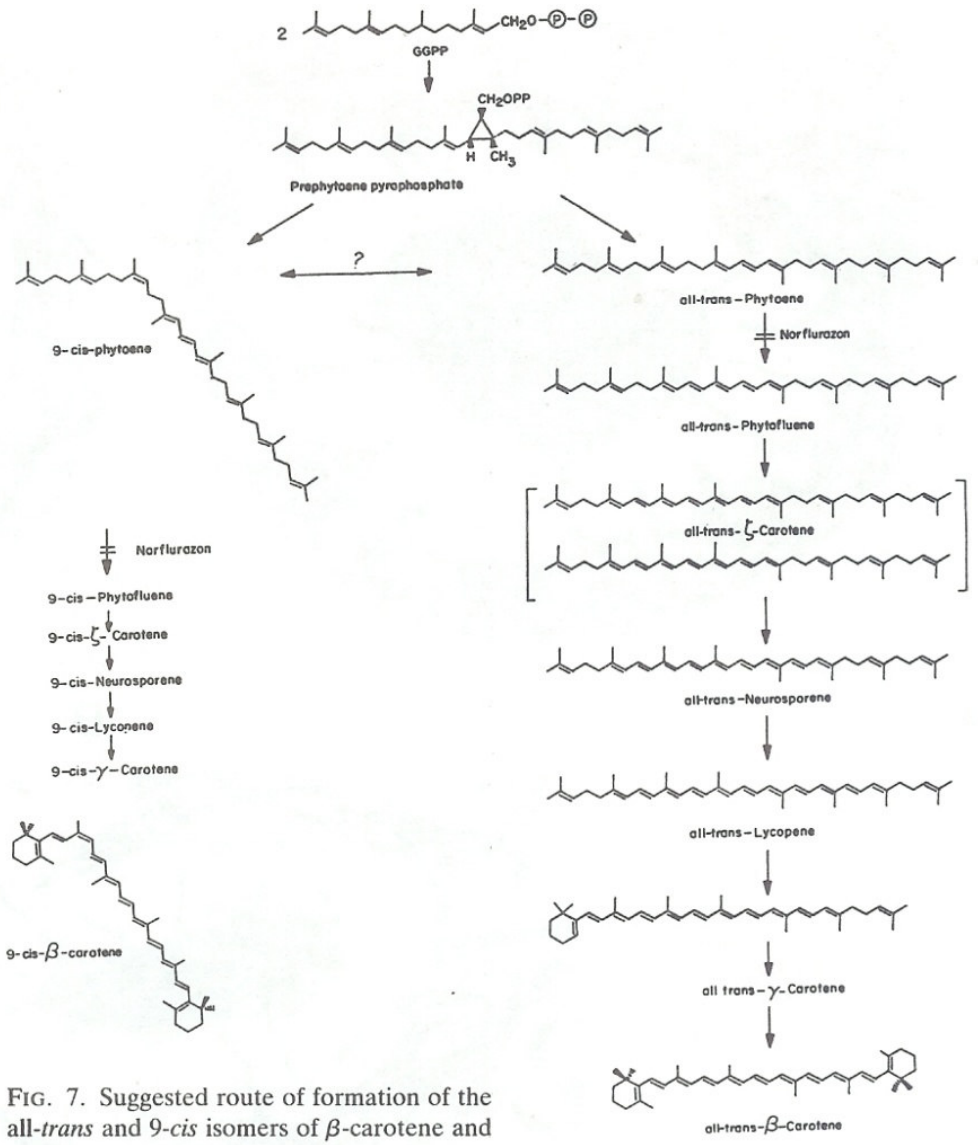
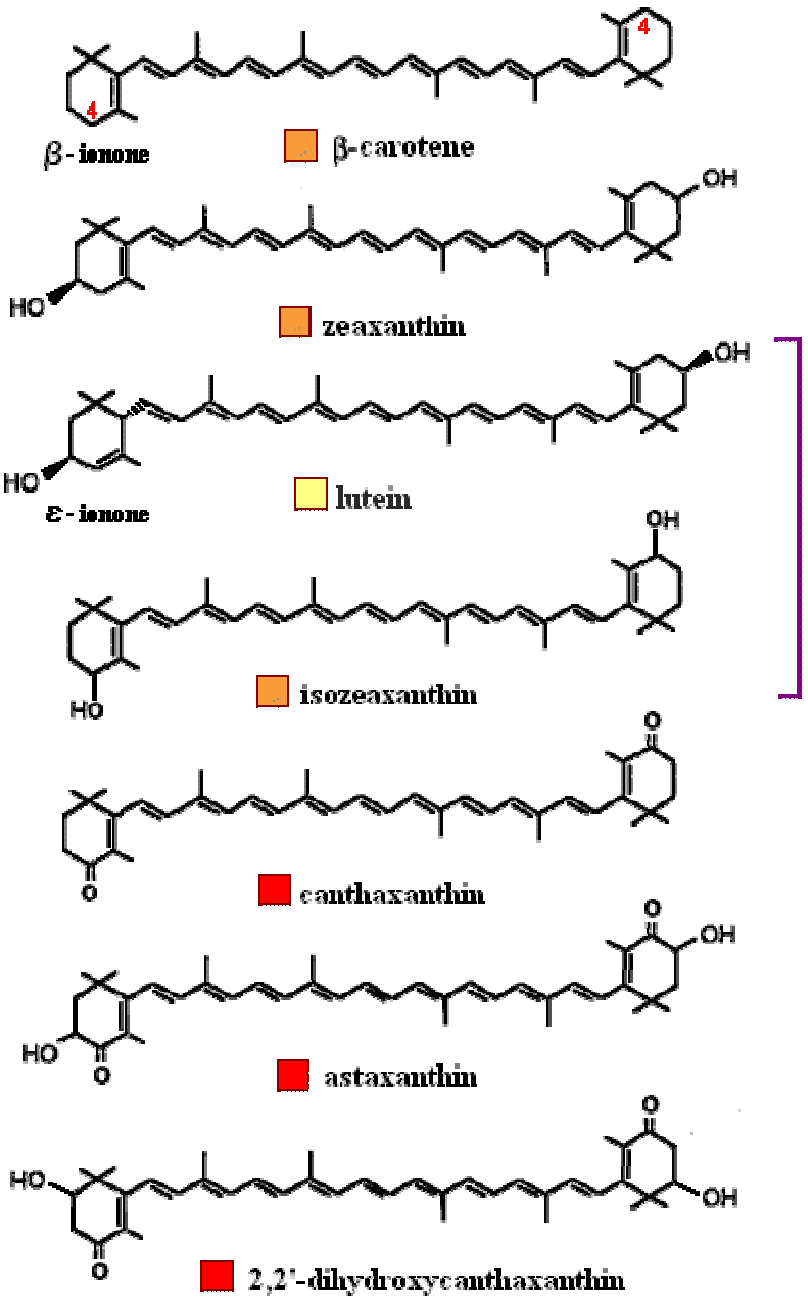


FIG. 7. Suggested route of formation of the all-*trans* and 9-*cis* isomers of β-carotene and phytoene.

Carotenoids and Isomerization

Sacrificial radical scavening

Anti-oxidant (quench singlet oxygen)



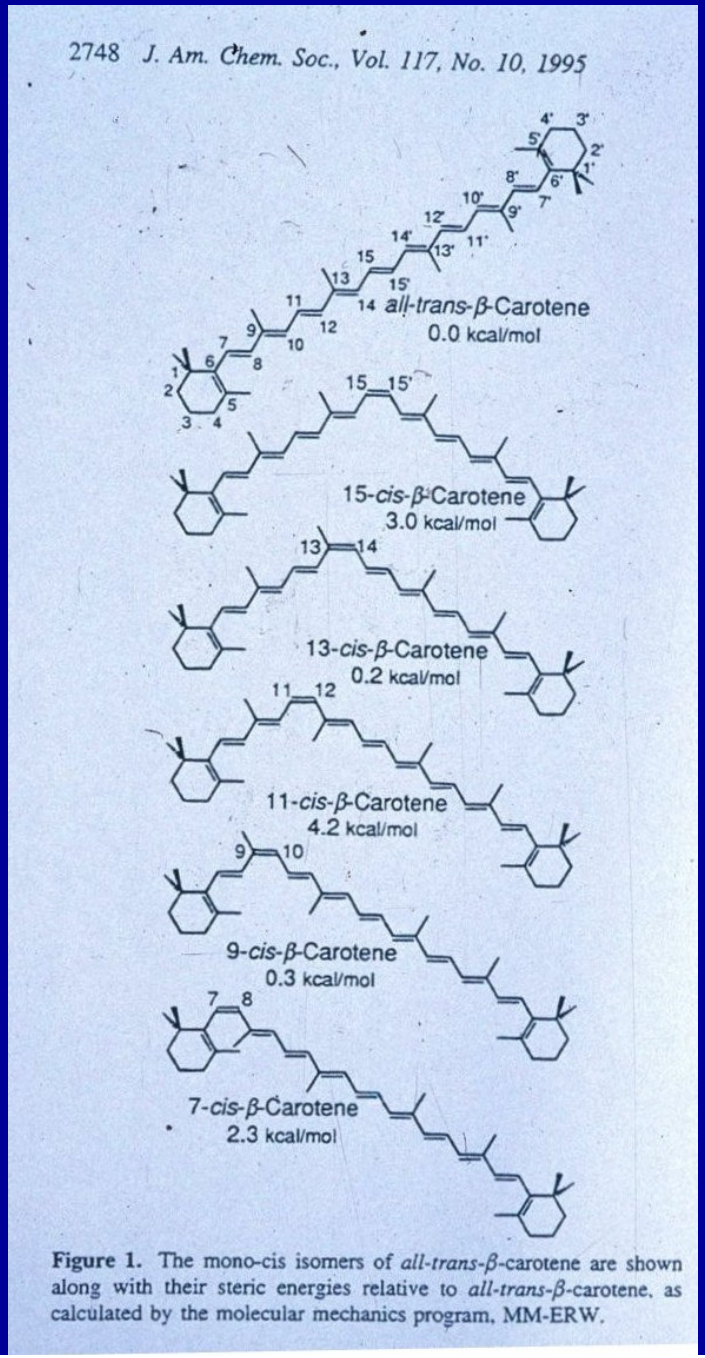
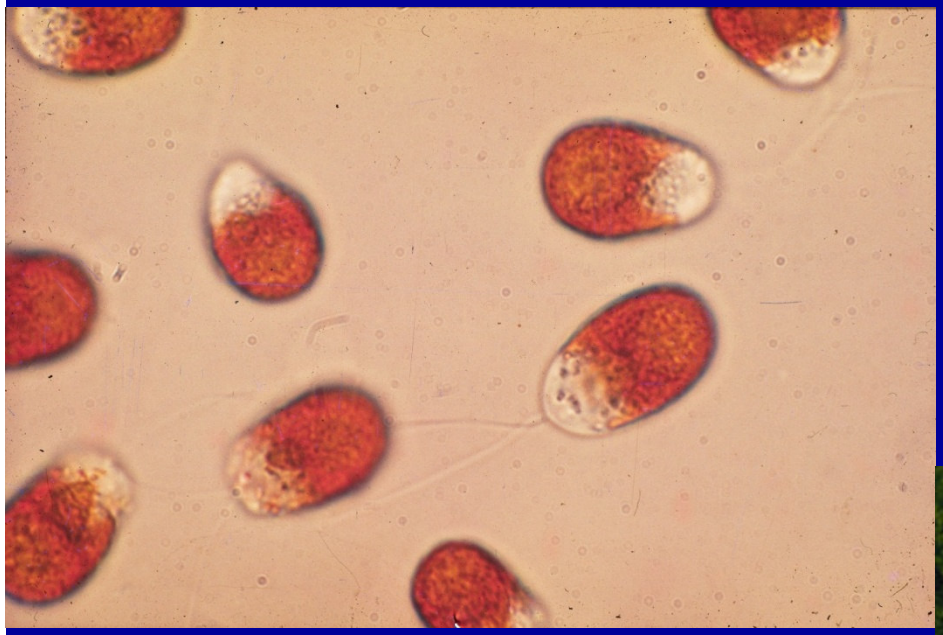


Figure 1. The mono-cis isomers of *all-trans*-β-carotene are shown along with their steric energies relative to *all-trans*-β-carotene, as calculated by the molecular mechanics program, MM-ERW.

The β-carotene in *Dunaliella* (and in fruits & vegetables) is composed of various stereoisomers of possible nutrition/medical interest

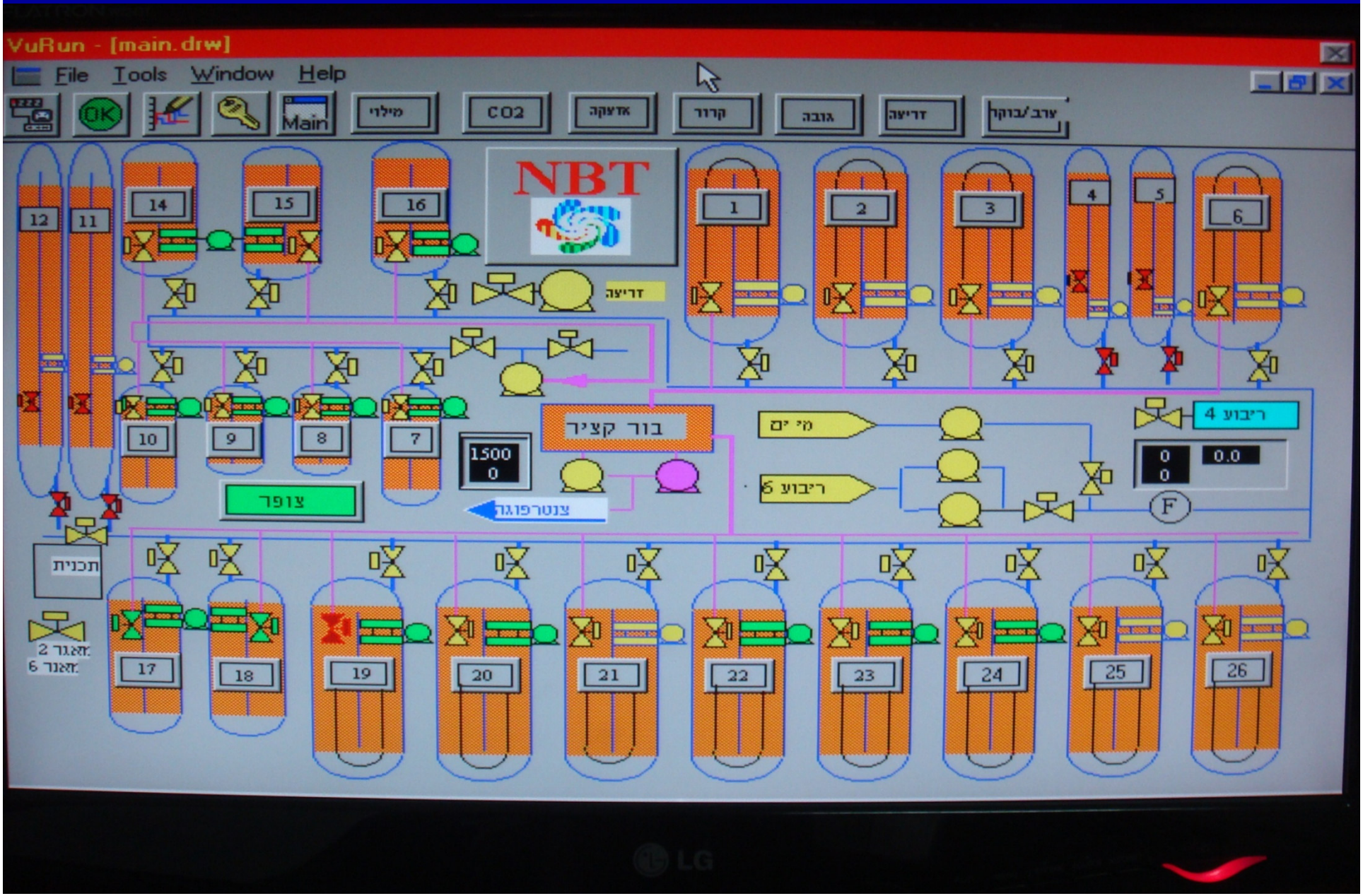


Dunaliella

Large Scale Biotechnology Cultivation & Processing

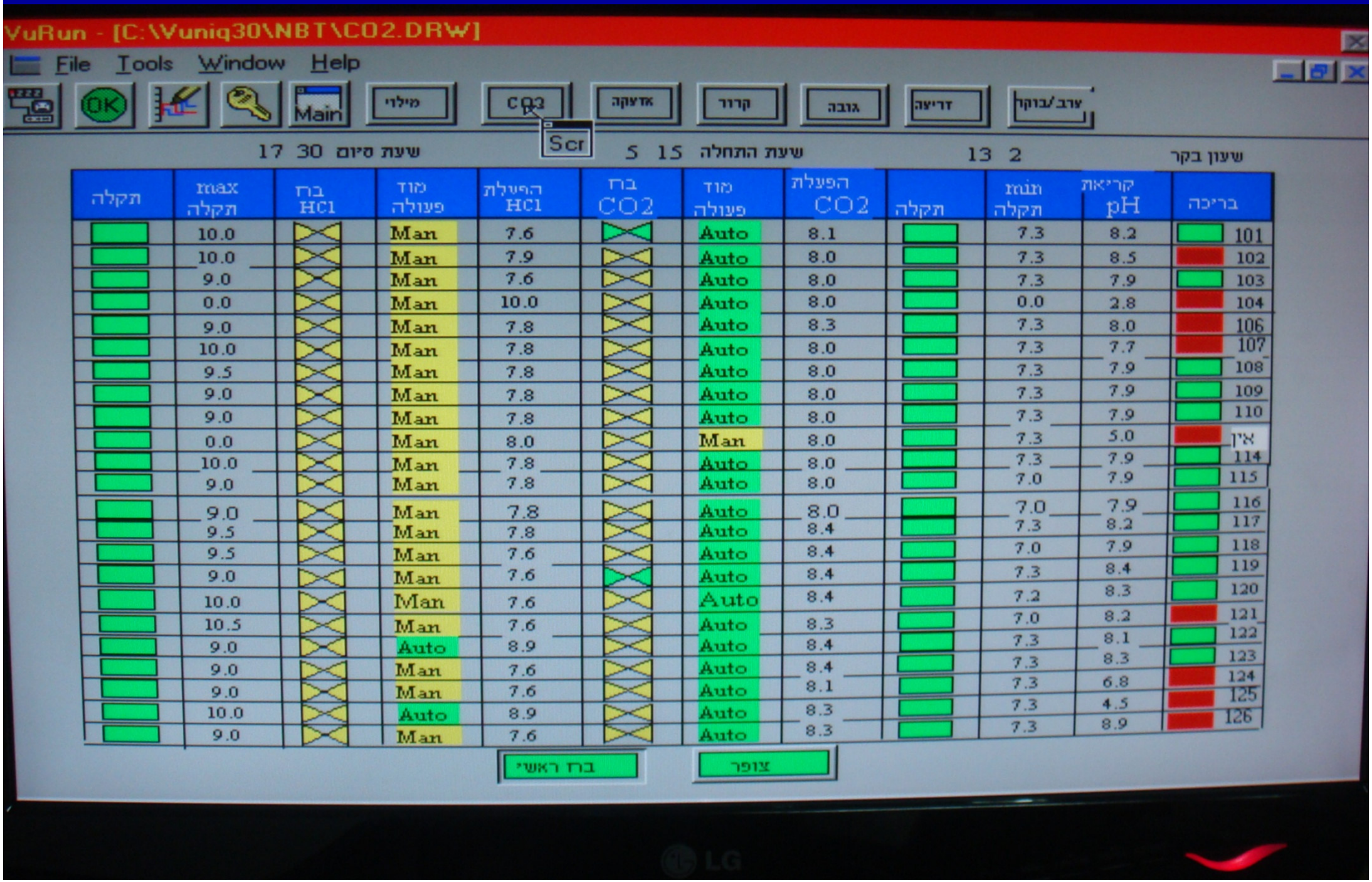
Dunaliella Biotechnology
Intensive Plant, NBT. Ltd., Eilat, Israel

PC Phycology Control

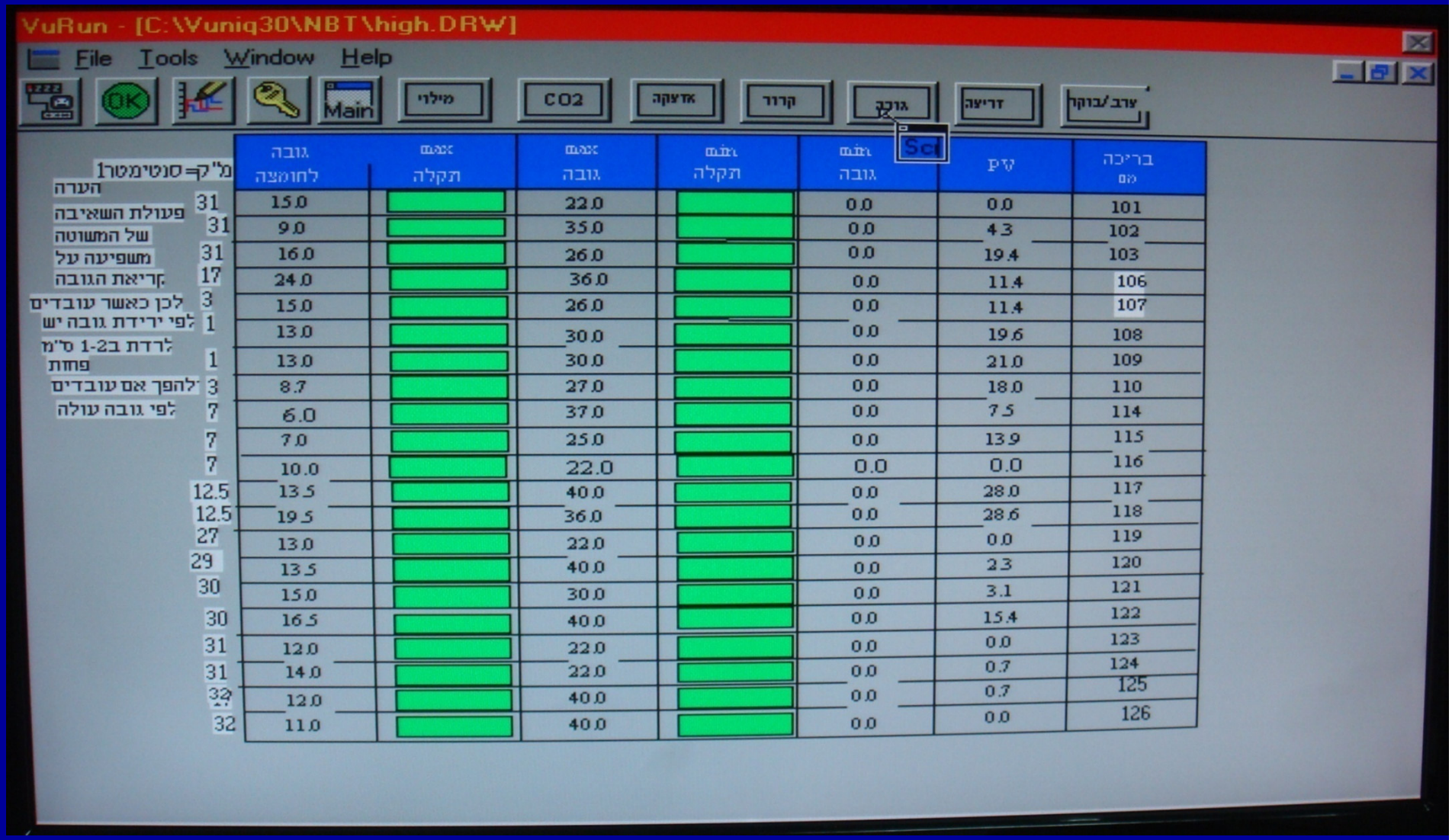


Dunaliella Ponds

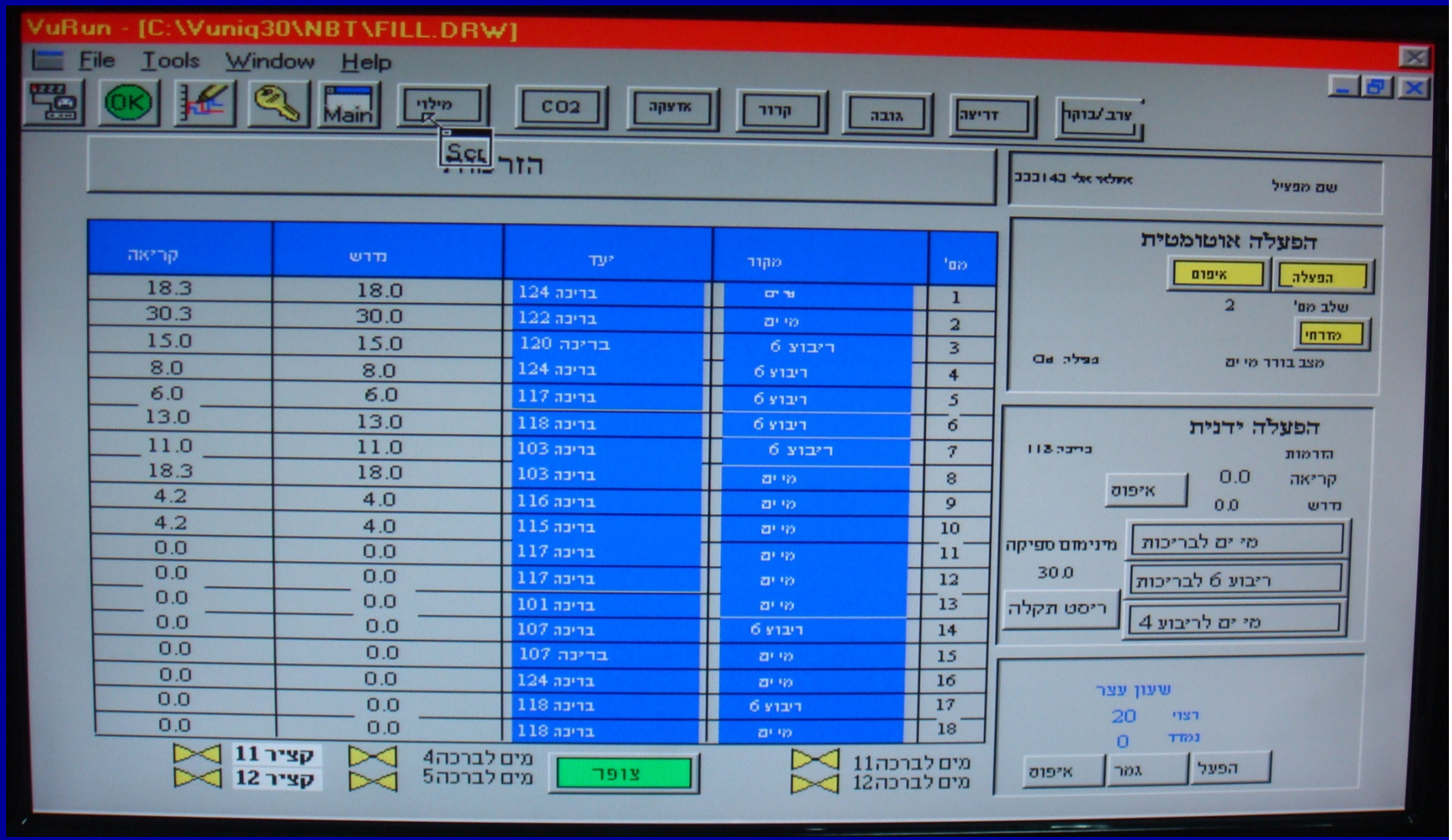
All Site PC pH Control



Depth Control (by sonic wave)



Volume and Salinity Control

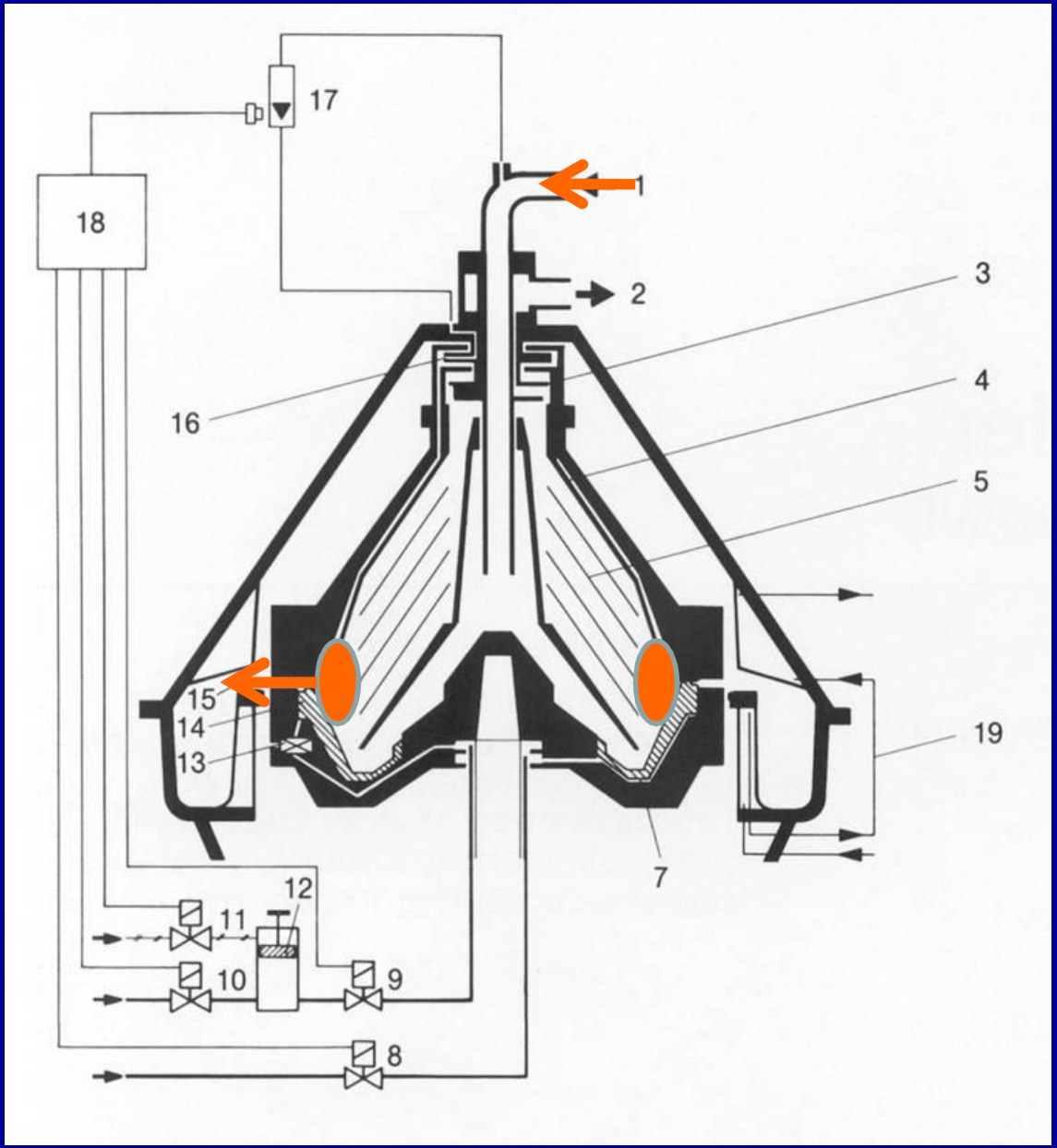


DUNALIELLA Harvesting

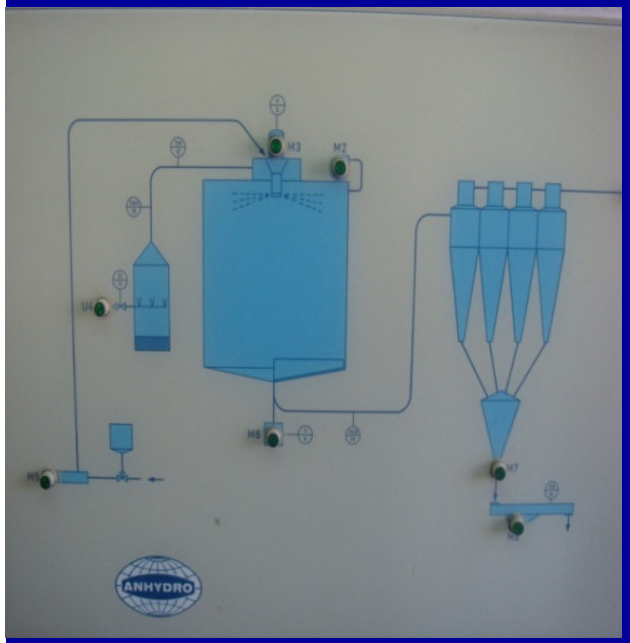
Westphalia Ltd., continuous "Clarifier Centrifuges"

Dunaliella Paste

Clarifier Centrifuges



Common Spray Dryer



Dunaliella
Spray Dried High β-Carotene Algal Powder

β-Carotene *Dunaliella* Powder Vacuum Packing

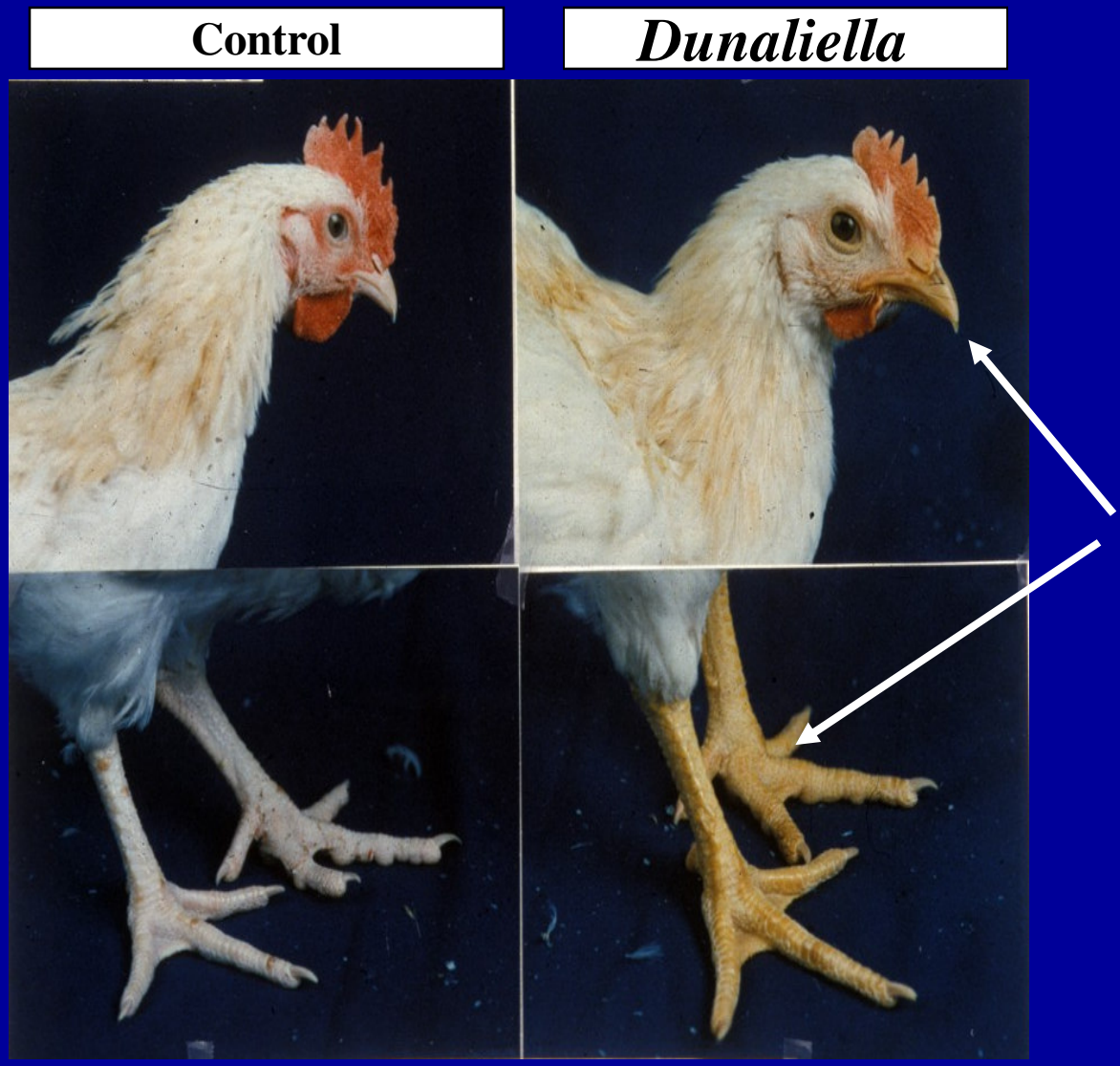
Natural β-Carotene
Dunaliella
Feeding Studies
Chickens
1990-2000

Chicks Feed

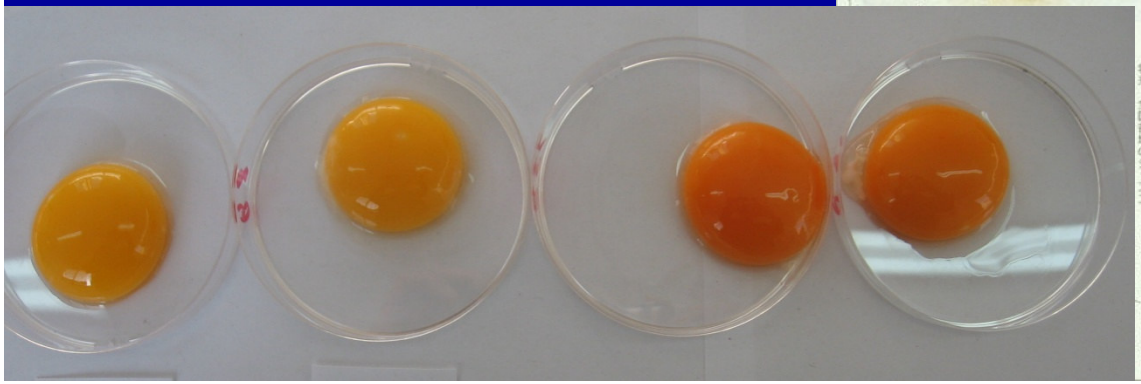
No Vitamin A | **+ Vitamin A** | **+ Synthetic β-carotene** | **+ *Dunaliella***

Chicken Feed
Vitamin A deficiency & *Dunaliella*

Dunaliella
Chicken Study



Chicken Feed, Egg Color

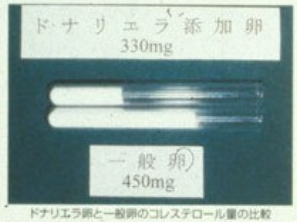


平成11年1月20日(水曜日)　THE ALGAE　VOL.219 (2)

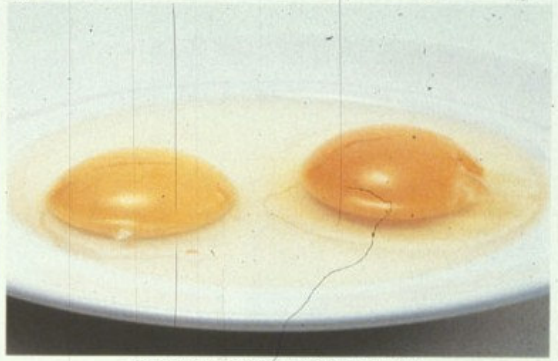
通常の10倍(150μg)の
ベータカロチンを含む

大きな期待が膨らむ
ドナリエラ卵の実力!!

ドナリエラ卵と一般卵のサルモネラ菌発生実験



ドナリエラ卵と一般卵のコレステロール量の比較



一般卵とドナリエラ卵の比較(左側一般卵 右側ドナリエラ卵)

ドナリエラ卵の特徴

- 天然ベータカロチンが一般の卵と比べて10倍以上ある
- コレステロールが一般の卵より少ない
- 食中毒の原因となるサルモネラ菌の繁殖を抑える
- 卵の生臭さがほとんどない
- 味が非常に濃く、美味である
- 黄身の色が大変濃く、艶がある
- 黄身は半円状に盛り上がり、摘めるほどの弾力がある

ドナリエラ卵が、ジャスコの店頭で発売開始されたということは、それなりの学術的な実証があってからこそ、現実のものと成り得たのです。

ドナリエラ飼料をニワトリに与え、そのニワトリが産んだ卵は、一目で一般の卵とは異なるものだと認識することが出来ます。そんなドナリエラ卵の特徴を次のようにまとめてみました。

日健総本社のドナリエラを飼料として利用すると、ニワトリも元気になり、その元気なニワトリから、このような素晴らしい卵が誕生するというわけです。

ジャスコ店頭で販売されたドナリエラ卵

養父への追い風になる

イスラエルN.B.T.で培養された飼料用ドナリエラを、ニワトリに与え、そのニワトリから産まれた卵が、「ドナリエラ卵」です。当初、ジャスコ側から提示された卵の名前がありましたが、「ドナリエラ卵」という名前に強いこだわりを持ち、交渉を重ねた結果、商標が「ドナリエラ卵」になりました。

将来、全国各地のジャスコ

今、国内外の様々な企業から我々の天然ベータカロチン「ドナリエラ」に対する関心が高まり、アプローチがきています。世界中から注目を浴びているこのドナリエラという商品に携わることが出来るのは、我々クロスタニングループだけなのです。そのことに自信と誇りを持って、今以上の頑張りで我々の商品を普

ます。そして、写真(左上)のようにサルモネラ菌の発生を抑制するという抗菌作用があること、また、コレステロールを抑えることが出来るのも魅力的です。

これらの新鮮な情報をホームパーティーの場で活かせば、話が盛り上がり、会話が弾むことなるでしょう。

このような商品を研究・開発し、生み出すことが出来る日健総本社ですから、発売から20余年、現在皆様に愛用していただいているクロスタニンゴールド、ドナリエラ、エバヤングをはじめとする40数種類もの我々の商品群が、いかに優れたものであるかがお分かりいただけると思います。

Dunaliella
Human Commercial Products

Japan & Far East Market Price
~$4,000/kg algal AFDA
(β-Carotene Health Food)
Total sale ~$US 100 million/year

Final Product
Japan

Raw algae
Israel

Dunaliella Capsules

300 mg dry powder/cap
20 mg β-carotene
9-cis/all-trans 1:1

Commercial *Dunaliella*

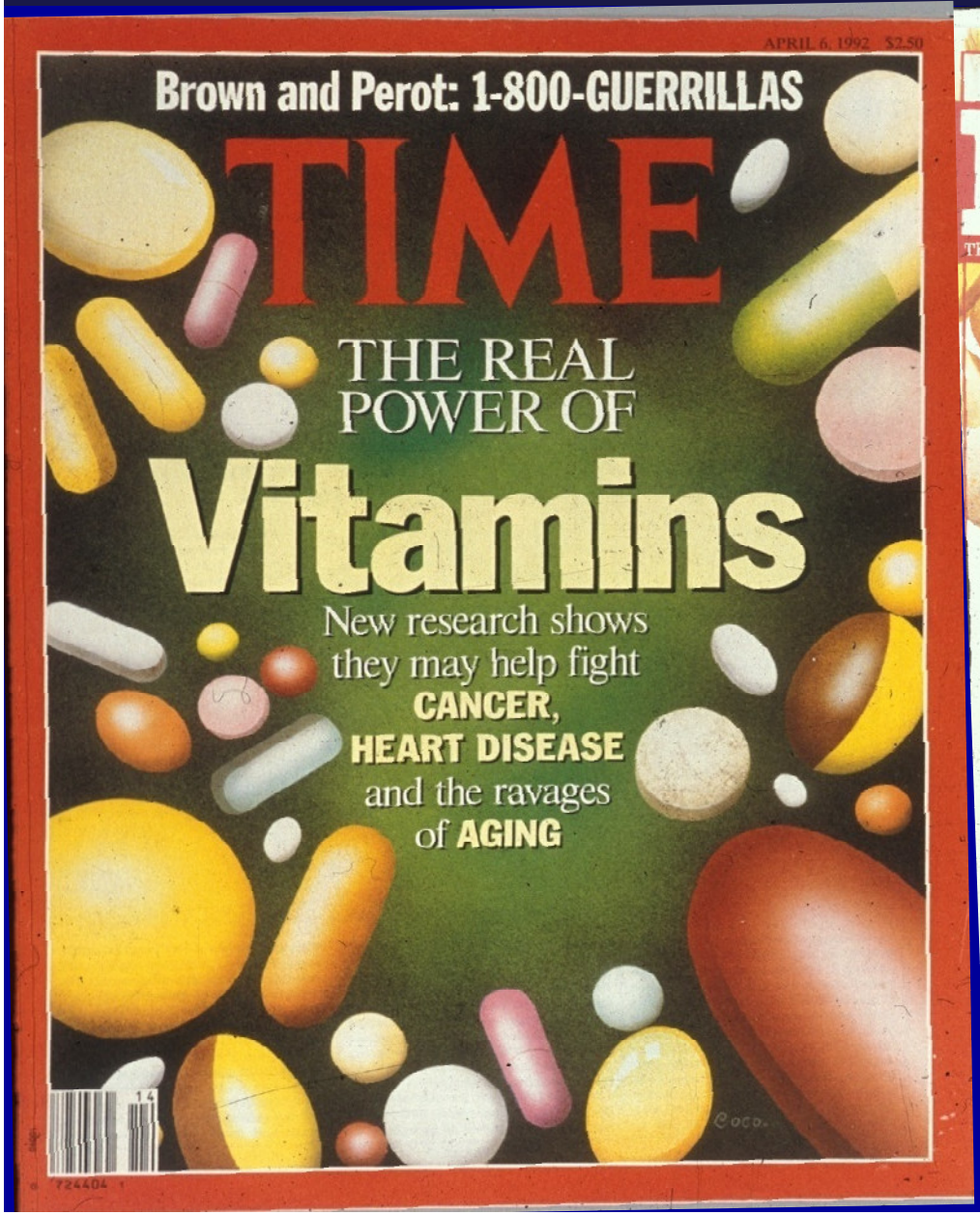
Dunaliella Health Food Powder

Door to door marketing by 500,000 sale agents in the Far East

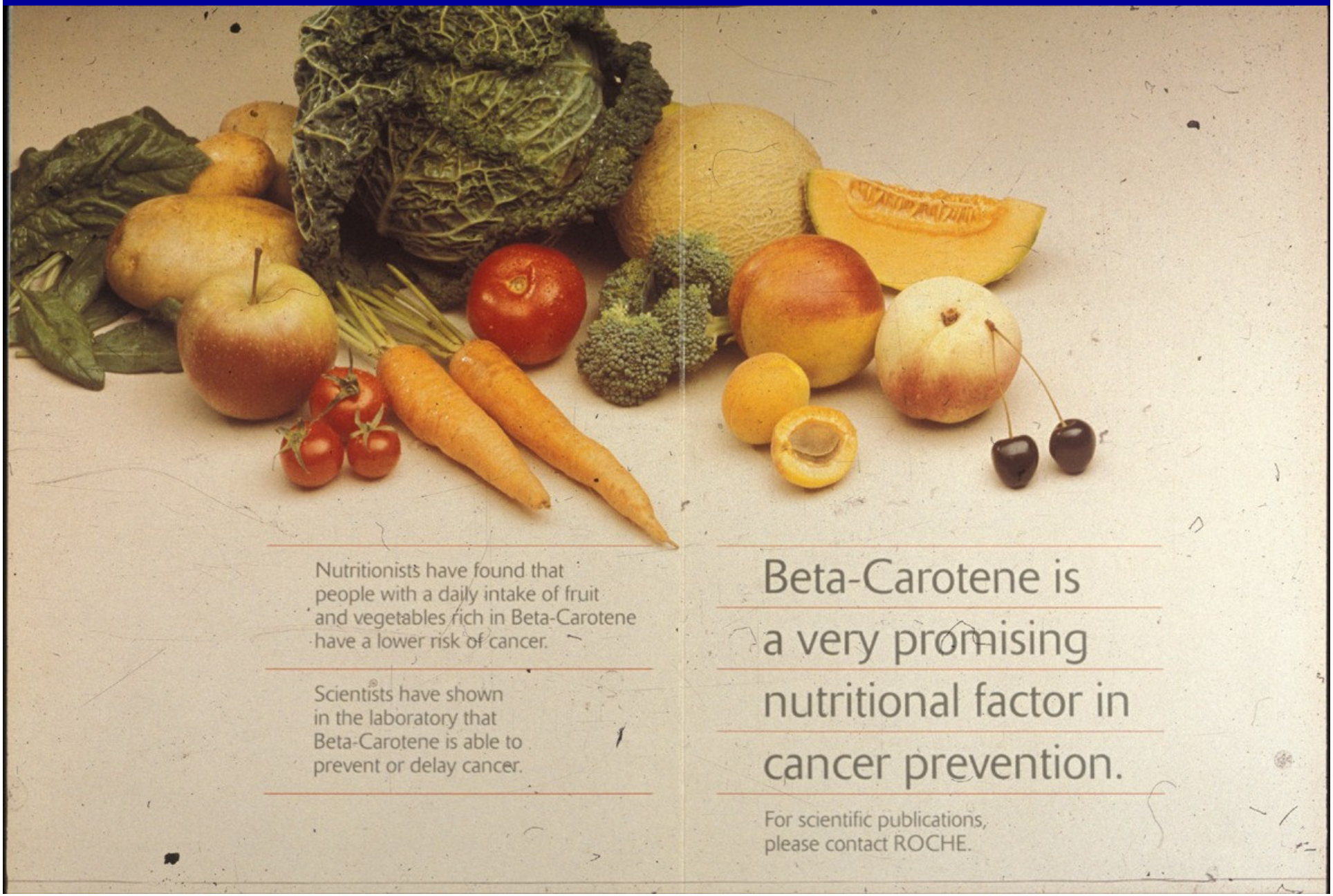
Dunaliella β-Carotene Cosmetics

β-Carotene
Friend? 1980-90
or
Foe? 1990-2000

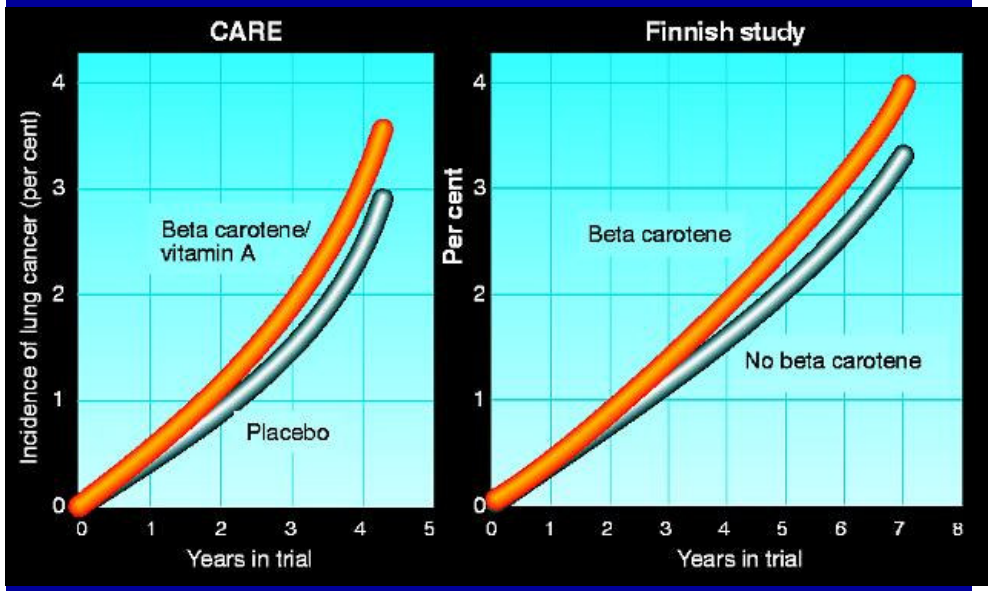
Vitamins, Market Boom 1980-1990



"β-Carotene Prevents or Delays Cancer", Roche 1986



The Rise and Demise of β-Carotene
Omenn 1998



Annu. Rev. Public Health. 1998. 19:73–99

CHEMOPREVENTION OF LUNG CANCER: The Rise and Demise of Beta-Carotene

Gilbert S. Omenn
Former address: School of Public Health & Community Medicine, University of Washington, Seattle, Washington 98195–7230 and Fred Hutchinson Cancer Research Center, Seattle, Washington 98109–1024; Current address: University of Michigan, Ann Arbor, Michigan 48109–0624; e-mail: gomenn@umich.edu

KEY WORDS: fruits/vegetables, carcinogenesis, vitamin-supplements, antioxidants, vitamin A

ABSTRACT

Beta-carotene and retinoids were the most promising agents against common cancers when the National Cancer Institute mounted a substantial program of population-based trials in the early 1980s. Both major lung cancer chemoprevention trials not only showed no benefit, but had significant increases in lung cancer incidence and in cardiovascular and total mortality. A new generation of laboratory research has been stimulated.

Rational public health recommendations at this time include: 1. Five-A-Day servings of fruits and vegetables, a doubling of current mean intake; 2. systematic investigation of the covariates of extremes of fruit and vegetable intake; 3. discouragement of beta-carotene supplement use, due to adverse effects in smokers and no evidence of benefit in non-smokers; 4. multilevel research to develop and evaluate candidate chemoprevention agents to prevent lung and other common cancers; and 5. continued priority for smoking prevention, smoking cessation, and avoidance of known carcinogens in the environment.

Could Synthetic β-Carotene Be the Real Problem?

Could Synthetic Beta-Carotene Be the Real Problem?

This article is from the September 1996 issue of The Nutrition Reporter■ newsletter.

Smokers who took beta-carotene supplements in recent experiments may have faced a greater risk of lung cancer because they took the synthetic form of the nutrient, a new study suggests. The subtle differences between synthetic and natural beta-carotene do appear to influence how the body uses the nutrient.

Synthetic beta-carotene consists of just the "all-trans" isomer of the nutrient, whereas natural beta-carotene consists of two different isomers, "9-cis" and "all-trans." Isomers have the same molecular formula, but a different arrangement of atoms. They're a little like anagrams, in which the letters of one word can be rearranged to form another, such as "star" and "rats."

It turns out that the natural 9-cis isomer is a more potent antioxidant than the all-trans, according to Ami Ben-Amotz, Ph.D., and Yishai Levy, Ph.D., in the American Journal of Clinical Nutrition (May 1996;63:729-34). That means the natural form has something the synthetic does not.

In experiments at Israel's National Institute of Oceanography, the researchers gave young, healthy men supplements of either natural beta-carotene from Dunaliella algae or synthetic beta-carotene. Blood analyses showed the presence of the all-trans isomer of beta-carotene, but not the 9-cis form found in natural beta-carotene. However, the researchers looked for and found 9-cis metabolic byproducts, indicating the presence and activity of the natural isomer.

Ben-Amotz and Levy reported experiments showing that the natural 9-cis isomer was rapidly used up in quenching free radicals and preventing oxidative damage to cell fats. In contrast, much of the all-trans isomer was converted to vitamin A, which is a very weak antioxidant.

Ben-Amotz and Levy wrote that the differences between natural and synthetic beta-carotene "should provoke a shift in scientific attention to natural sources of carotenoids and their role in cancer prevention." They urged that researchers pay more attention to the different isomers of beta-carotene.

An earlier study, in Free Radical Biology & Medicine (1994;17:77-82) also showed that the natural 9-cis isomer is a more potent antioxidant than the all-trans form.

The information provided by Jack Challem and The Nutrition Reporter■ newsletter is strictly educational and not intended as medical advice. For diagnosis and treatment, consult your physician.

[Vitamins Natural vs. Synthetic]

Posted on: 09-11-2009 , by: admin, in: Nutrition

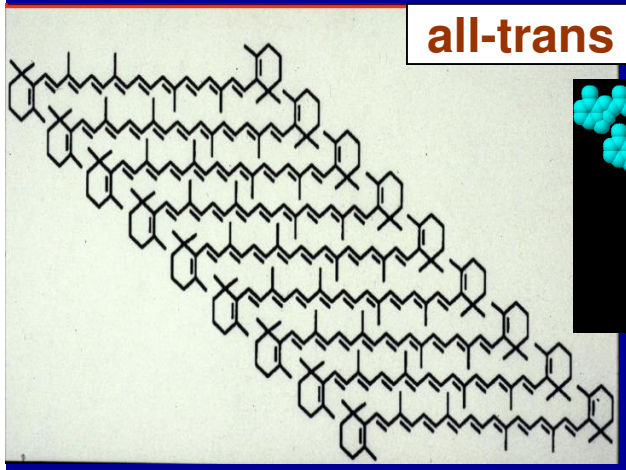
Beta-Carotene: The Controversy
Alternative Medicine Review, Dec, 2000
by Lyn Patrick

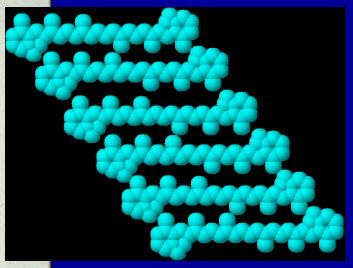
Carotenoids
Nutritional & Medical Effect

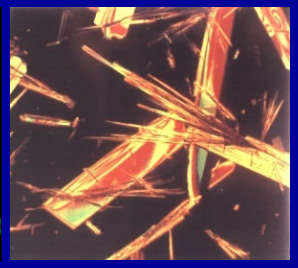
1. Antioxidants, cellular level?
2. Pro-retinoic acids, molecular level?
3. Structural?
4. Other?

β-Carotene, the Packaging Structural Model

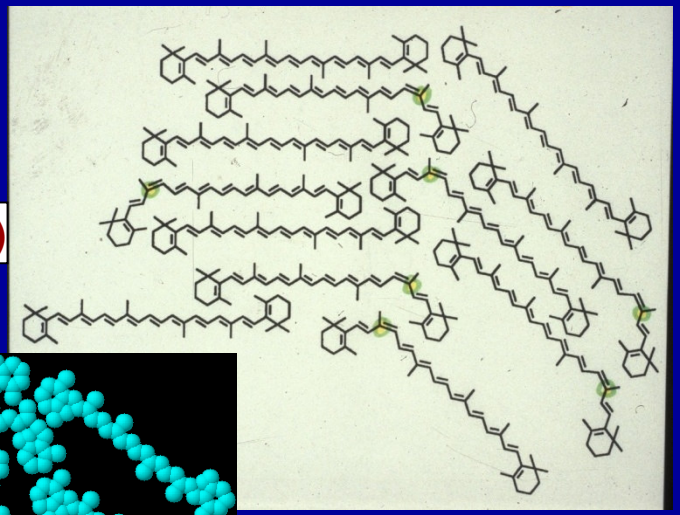
all-trans (crystals, not absorbed)

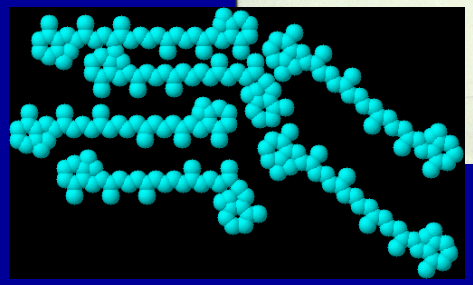




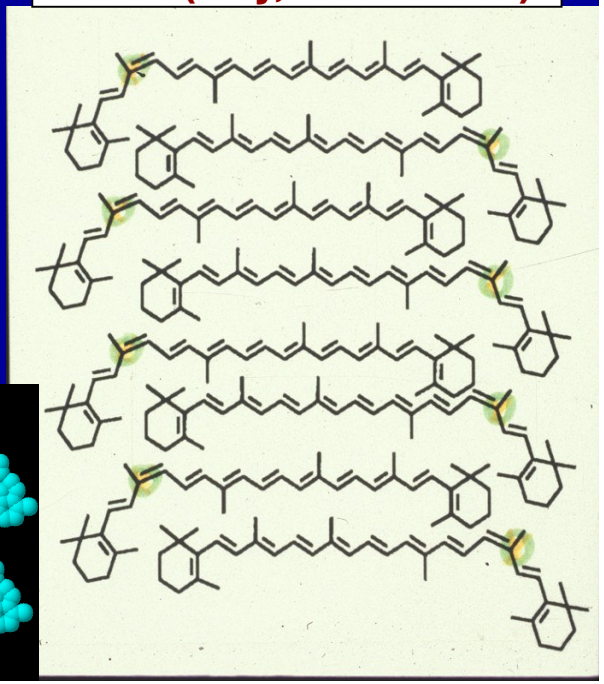


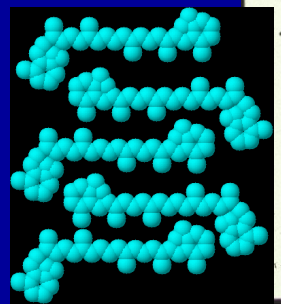
all-trans/9-cis (oily, absorbed)



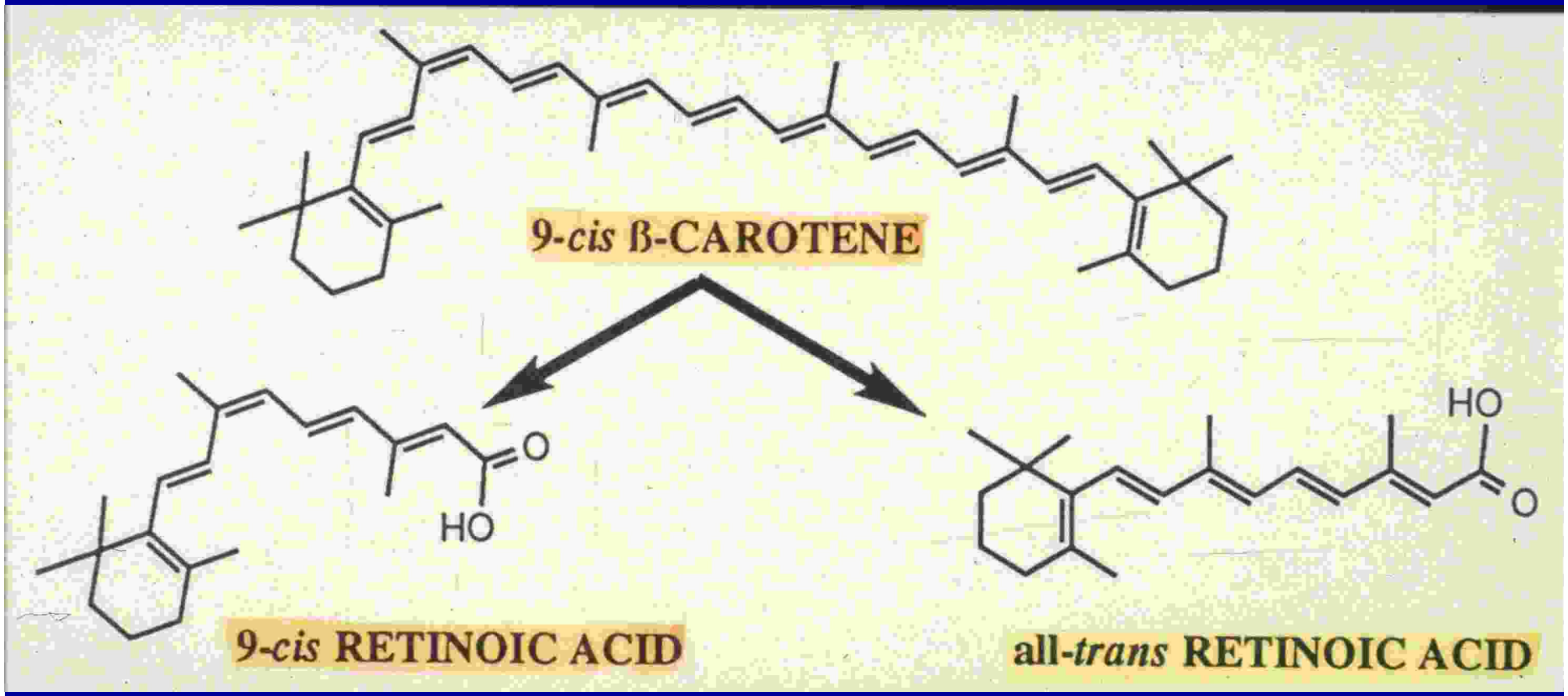


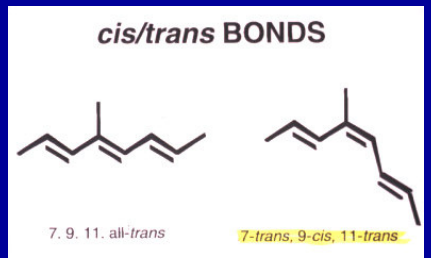
9-cis (oily, absorbed)



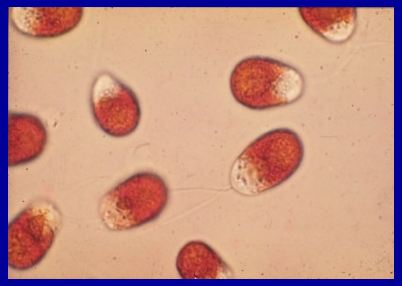


Conversion of cis-Carotenoids to cis-Retinoids



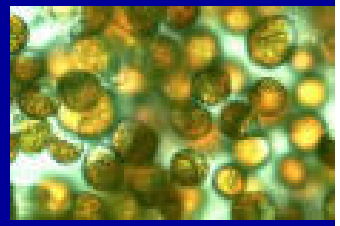


9-cis β-Carotene → 9-cis Retinoic acid



Dunaliella

9-cis phytoene → acyclic (pro)-retinoid



Phyto-*Dunaliella*

Nucleus

RXR & pro-RXR

PPAR

AGGTCA TAGGTCA

Target Gene

Metabolic function

PPRE

Dunaliella carotenes

Metabolic Molecular Function

マイクロアルジェは、どれだけ人間を幸せにできるだろう。

The Blessing Of Nature

自然の贈り物

天然ベータカロチン含有食品

NATURAL BETA CAROTENE

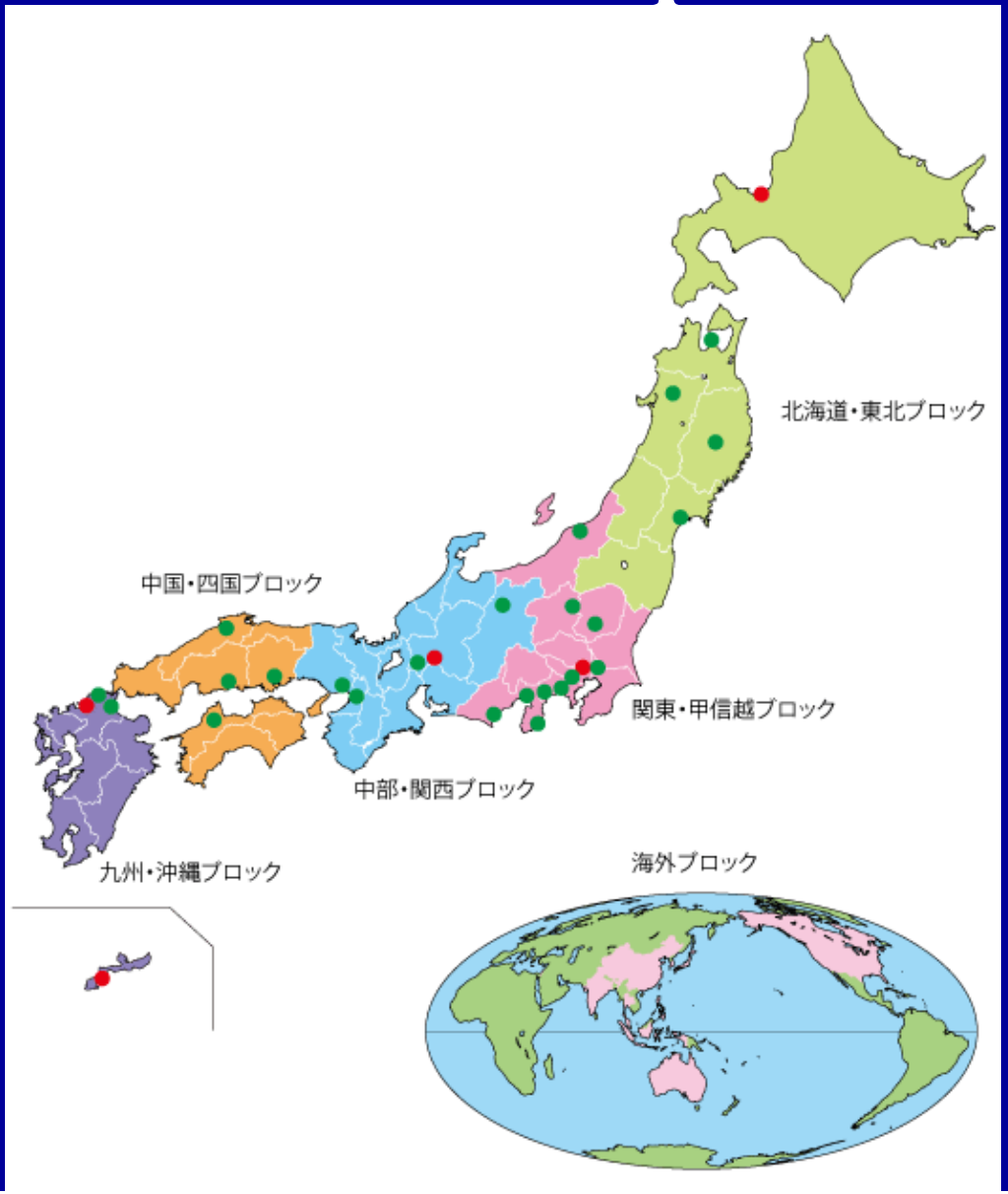
Dunaliella（ドナリエラ）

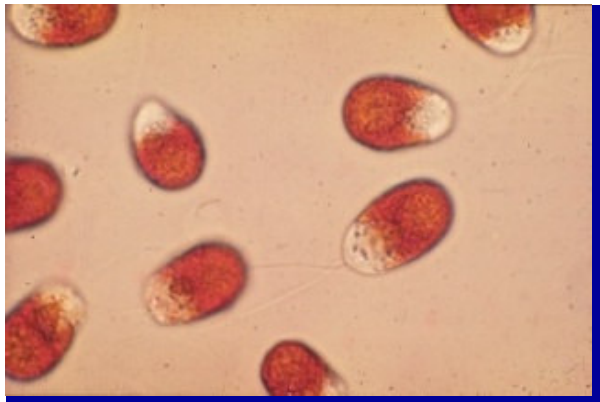
ハードカプセル

私たちが健康に生きるためには、健康な食物を毎日食べる事が必要です。
化学物質や環境ホルモンなど、現在の生活習慣病を引き起こすものは、化学合成品によるものが多いのではないでしょうか。
天然の良さは、様々な必要な成分が混ざっておりこれらが互いに働きあって、安全でかつ幾つかの作用が発揮されます。

合成でだいじょうぶ!?

Nikken Sohonsha Co. Network
The Far East & Japan Markets





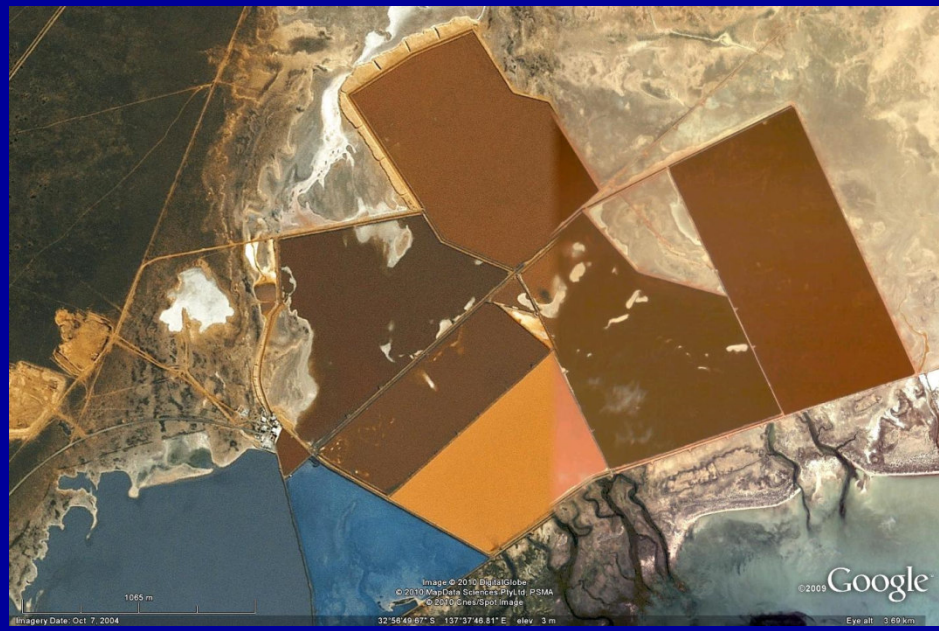
Natural Carotenoids

Production Cost

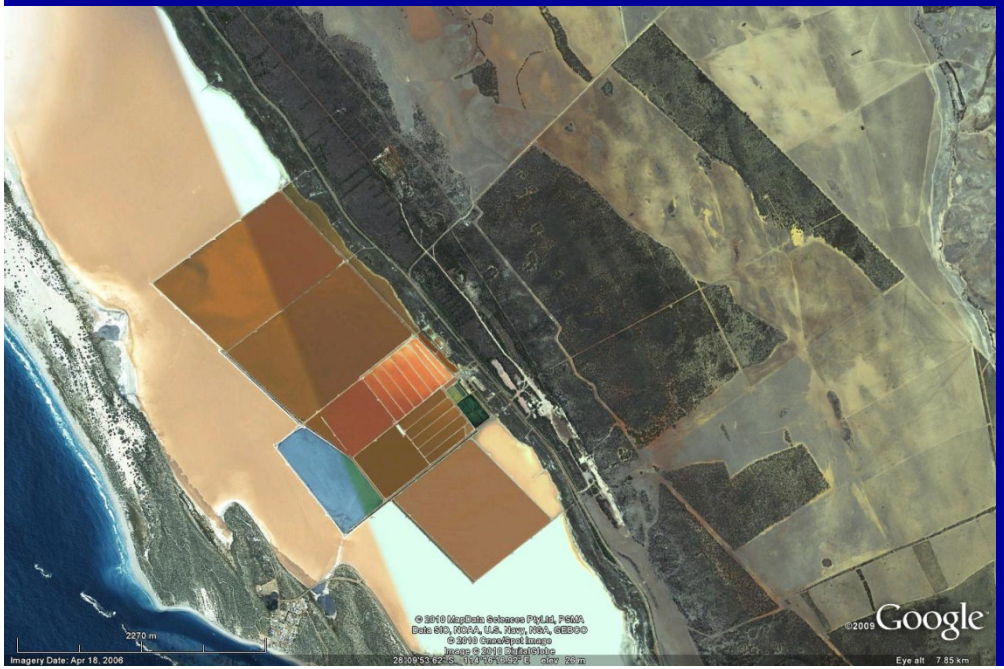
NBT *Dunaliella* Plant
(10 Hectares Plant)

Item	*Dunaliella* NBT Ltd., Eilat, 2009
	Cost in US$/year
Manpower	500,000 (20 workers)
Electricity ($0.125/KW)	180,000
Fertilizers (N,P,K, Fe) and other chemicals	36,000
Domestic Land City Taxes	50,000
CO_2 ($400/ton)	120,000
Sea Water ($0.25/m3)	200,000
Fresh Water	20,000
Other supplies and Miscellaneous	30,000
Total	1,136,000
Yearly production of dry algae biomass	70 tons (2g/m2/day)
Cost of 1Kg dry *Dunaliella*	$16.23/kg

Cognis, Whayalla (Adelaide), Australia
400 hectares



Cognis, Hutt Lagoon, West Australia
400 hectares



About the Book

The green microalga *Dunaliella* has received renewed attention due to its biotechnology applications including production of high-value compounds as well as generation of biofuels. The scope of the present volume 'The Alga *Dunaliella*: *Biodiversity, Physiology, Genomics, and Biotechnology*' is wider than the first book on *Dunaliella* by Avron and Ben-Amotz published in 1992, and presents a broad collection of state-of-the-art reviews on a variety of fundamental and applied research fields involving *Dunaliella*. The 21 contributions written by leading international experts cover not only basic science regarding cultivation of *Dunaliella*, biodiversity, life-cycle, pigments and photosynthesis, but also the applied fields of bioreactor design, production of valuable compounds, biofuels, bioremediation, genetic engineering.

This book is an essential read for researchers in the field, while at the same time providing a valuable resource for teaching of advanced undergraduate, graduate, and postgraduate students who are interested in algae.

About the Editors

Dr. Ami Ben-Amotz is Emeritus Professor of Marine Phycology at the National Institute of Oceanography (NIO) in Haifa, Israel. He initiated research on the biology, physiology, biochemistry and biotechnology of *Dunaliella* in long collaboration with the late Prof. M. Avron of the Weisman Institute of Science (WIS). The fruitful cooperation led the way to establishment of the commercial *Dunaliella* production plant in Eilat. Prof. Ben-Amotz served as Head of the Department of Marine Biology at NIO and Head of the *Dunaliella* Section at the WIS, Chief Scientist of Nature Beta Technologies Ltd., (NBT) and recently as Chief Scientist of Nikken Sohonsha Co., Japan. Prof. Ben-Amotz served as President of the 8th International Marine Biotechnology Conference, held in Eilat in March 2007.

Dr. Jürgen Polle is an Associate Professor in the Department of Biology and Associate Director of the Aquatic Research & Environmental Assessment Center at Brooklyn College of the City University of New York. He received his Doctorate in Biology from the Georg-August University in Gottingen, Germany and he is an alumnus of the German National Merit Foundation. He performed postdoctoral research from 1997 to 2002 at the University of California at Berkeley. At Brooklyn College, Dr. Polle's focus on the alga *Dunaliella* resulted in creation of the *Dunaliella* Culture Collection at Brooklyn College (DCCBC) with the webpage www.dunaliella.org. Dr. Polle is a member of the consortium responsible for the genome sequencing of *Dunaliella salina*. Currently, he mentors several graduate students in his laboratory.

Dr. D.V. Subba Rao is a biological oceanographer, and Emeritus Scientist at the Bedford Institute of Oceanography (BIO), Canada. His research and teaching activities took him to CSIRO, Australia. The Johns Hopkins University, Baltimore, and Kuwait Institute for Scientific Research. Based on data collected on natural populations and algal cultures he is author/co-author of over hundred papers in peer reviewed journals on phytoplankton ecology, physiological ecology, primary production, picoplankton, red tides, phycotoxin episodes, ballast water introductions of exotic organisms, tsunamis and marine life, and impact of mega engineering projects on marine environment. He currently serves as editor of Journal of Oceanography. Dr. Subba Rao has edited two books "Pelagic Ecology Methodology" (Balkema, 2002) and "Algal Cultures, Analogues of Blooms and Applications" (Science Publishers, 2006).

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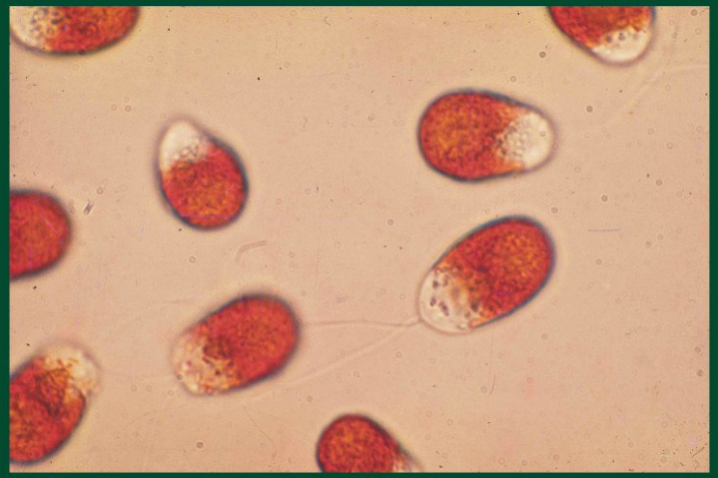
The Alga *Dunaliella*

Biodiversity, Physiology, Genomics and Biotechnology

Editors
Ami Ben-Amotz
Jürgen E.W. Polle
D.V. Subba Rao

The Alga *Dunaliella*

Biodiversity, Physiology, Genomics and Biotechnology



Editors
Ami Ben-Amotz
Jürgen E.W. Polle
D.V. Subba Rao


Thank You