

**STRESS OSSIDATIVO, ALTERAZIONI DELLA HMGC_oA
REDUTTASI E DEL METABOLISMO DEGLI ISOPRENOIDI
E PATOGENESI DELLA IPERCOLESTEROLEMIA SENILE**

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BIOMARKERS OF AGING

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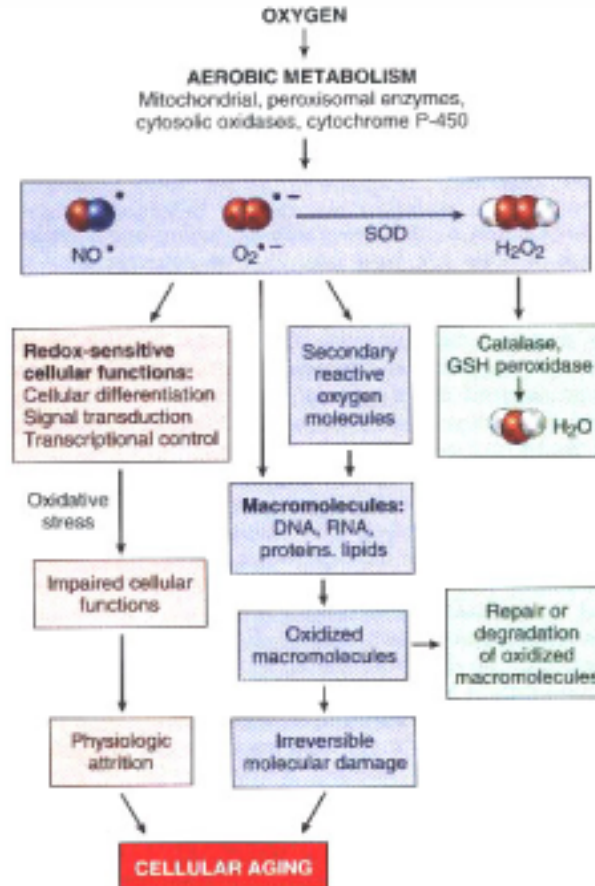
Centro di Ricerca sull'Invecchiamento

Università degli Studi di Pisa

A Biomarker of Aging is a biological parameter that can predict changes in function capacity with increasing age better than chronologic age, either alone or in a multivariate manner (Baker and Spratt, 1988)

Biochemical and molecular BA may be most interesting with regard to understanding of aging: accumulation of altered DNA, protein and lipid with increasing age may be measured and provide something like a sand-clock

Ageing is said to be the consequence of free-radical attack and age-related decline in cell-repair function



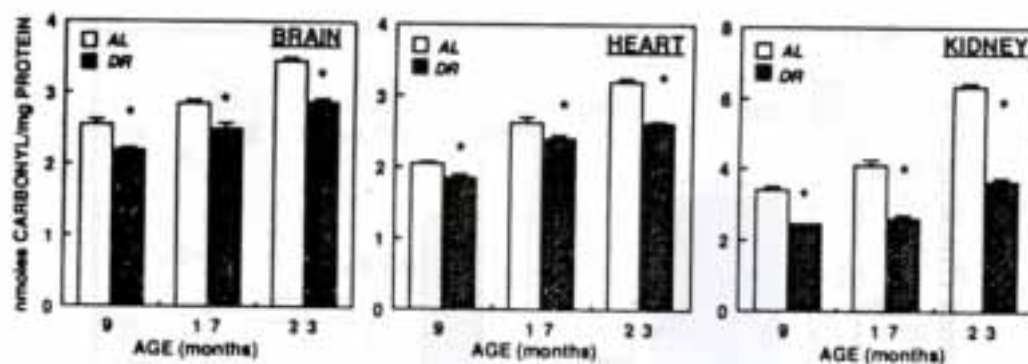


Figure 6-2. Effects of age and CR on the carbonyl content of proteins of the brain, heart, and kidney of male C57BL/6 mice. AL denotes *ad libitum*-fed mice and DR denotes mice on CR from 4 months of age. (From Sohal et al., 1994b.)

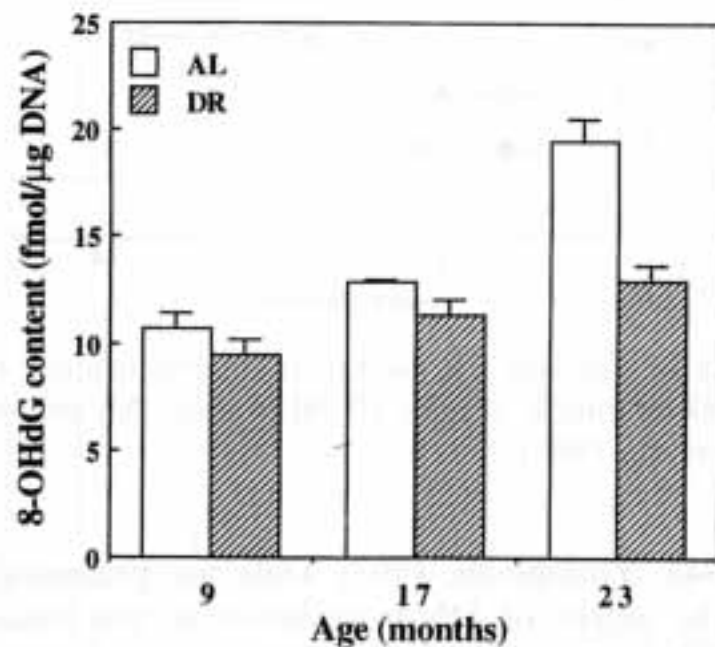


Figure 6-5. Effects of age and CR on the oxidation of hepatic DNA in male C57BL/6 mice. DR denotes CR. (From Sohal et al., 1994a.)

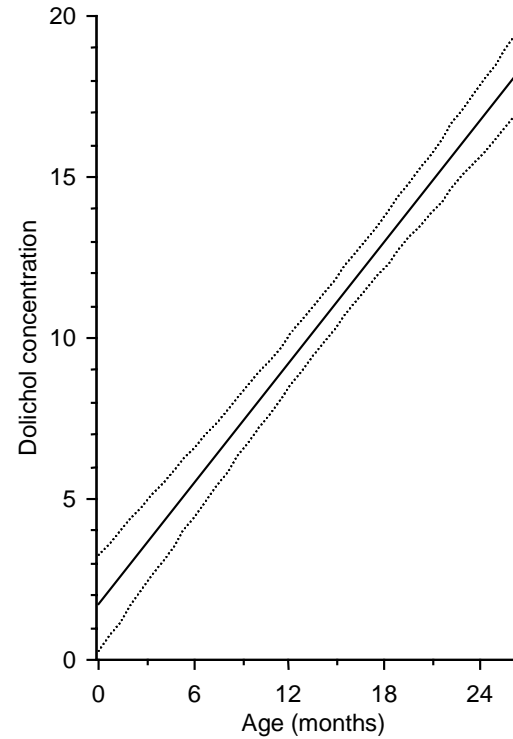
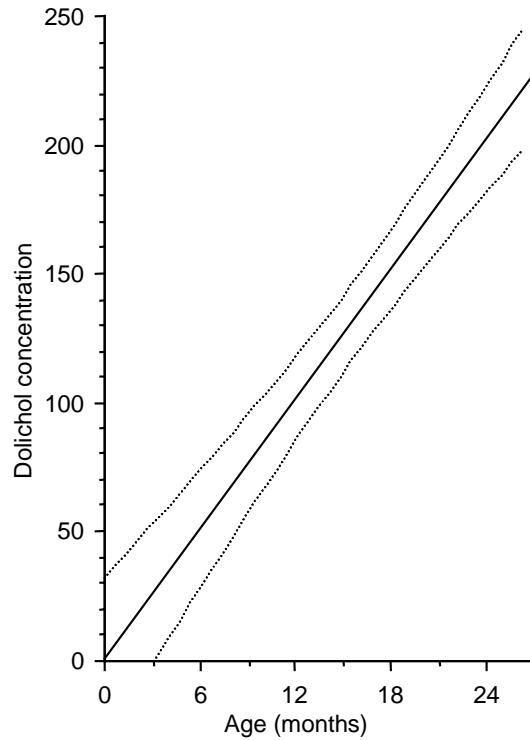
Parentini I, Cavallini G, Donati A, Gori Z, Bergamini E.

The accumulation of dolichol in older tissues satisfies the proposed criteria to be qualified a biomarker of aging.

Journals of Gerontology 2004, in the press.

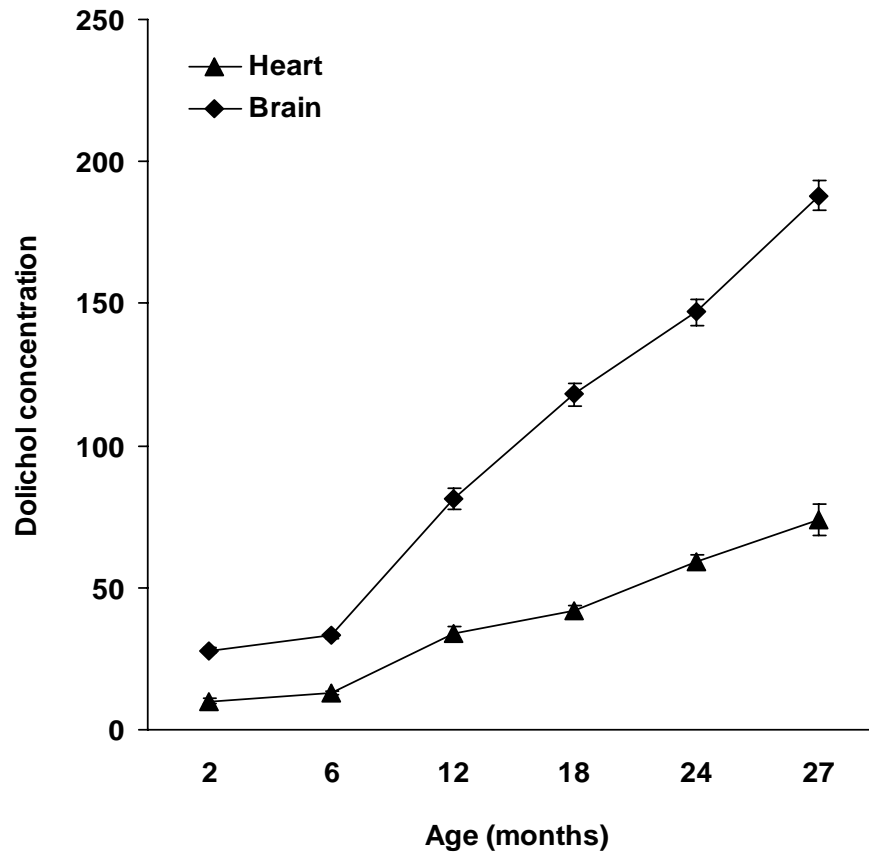
- * *Dolichol accumulates in mammalian tissues with increasing age.*
- * *The levels of dolichol are not altered with disease processes or alteration with disease is not in the same direction as that of aging.*
- * *The age-related alteration in the tissue levels of dolichol is not secondary to metabolic or nutritional changes of aging.*
- * *Caloric restriction, which modulates the aging rate, appropriately alters the levels of dolichol in the liver tissue.*
- * *As a biomarker of aging, dolichol is applicable to different tissues with similar replicative capacity and is generalizable across mammalian species.*
- * *Applicability of dolichol to syndromes of premature aging.*
- * *Application of the biomarker dolichol to aging studies.*

Aging related changes in dolichol concentration in the liver (A) and the soleum muscle (B) of male Sprague Dawley rats fed ad libitum



Linear regression and 5% fiducial limits are depicted. Regression equations are: liver: $y = 0.7 + 8.4x$; soleum muscle: $y = 1.8 + 0.6x$.

Aging related changes in dolichol concentration in the heart and the brain of male Sprague Dawley rats fed ad libitum



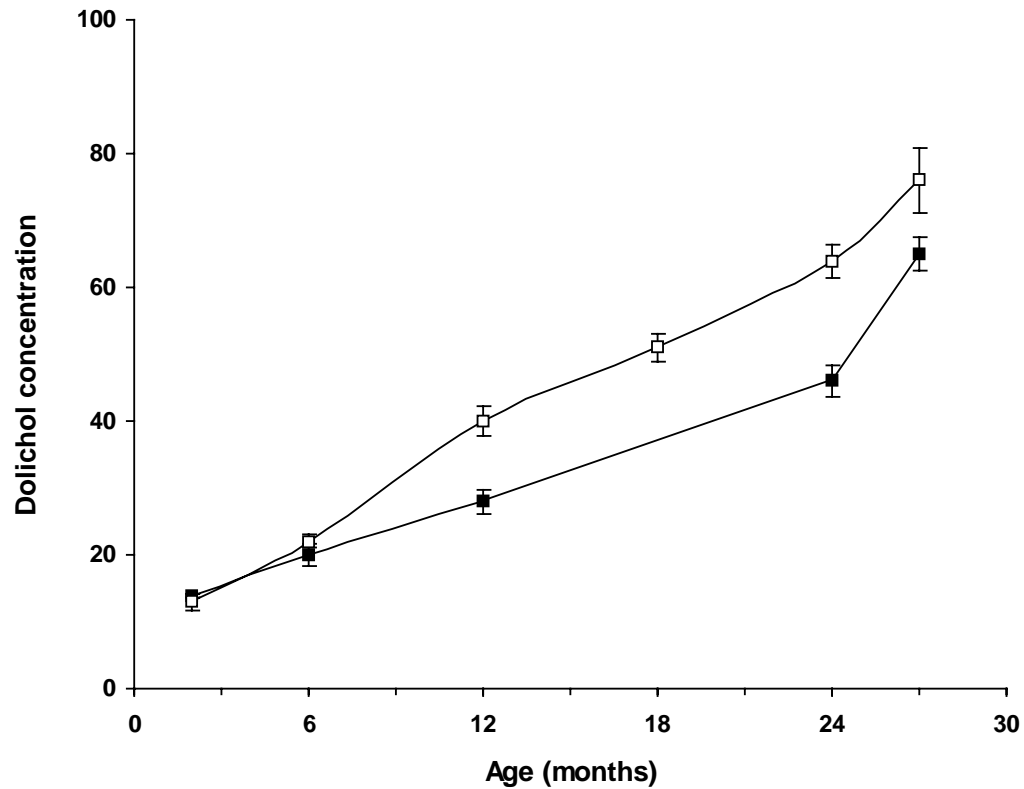
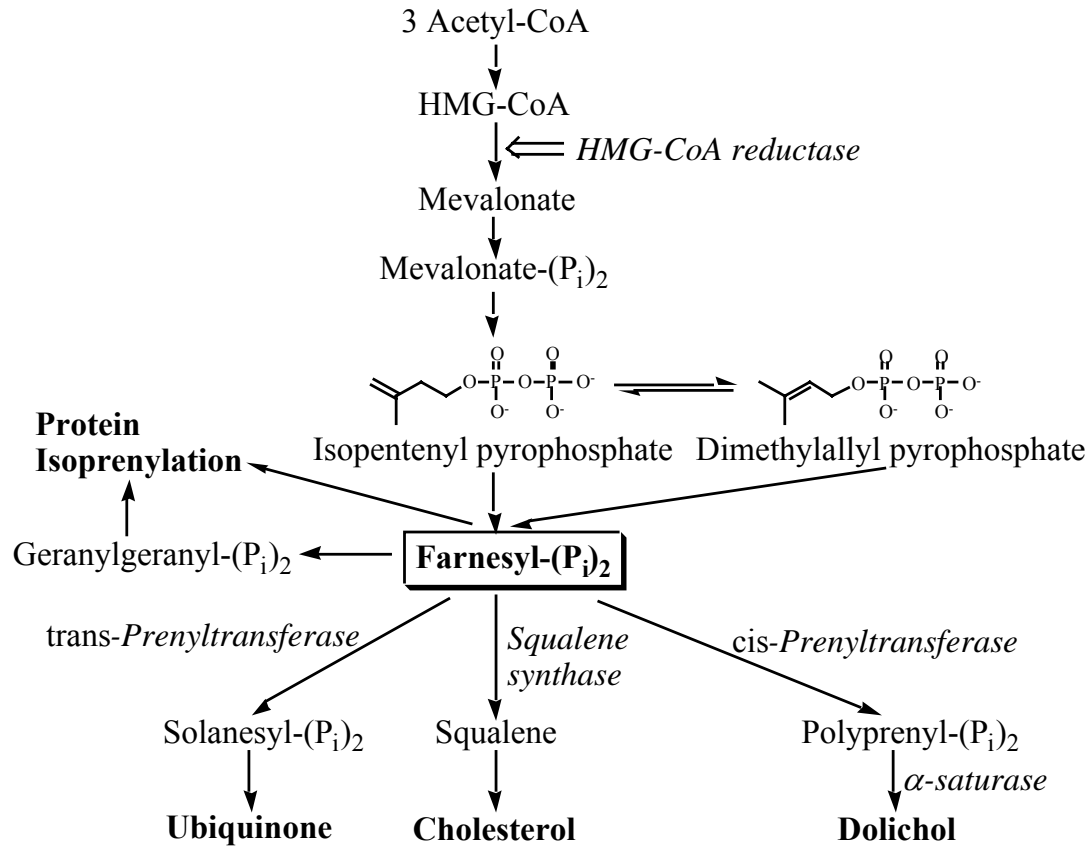


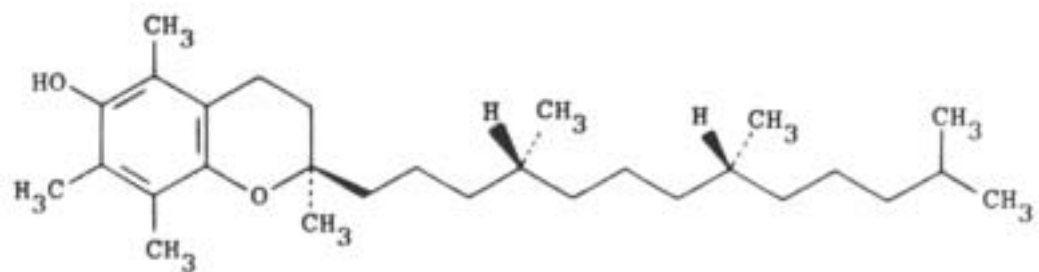
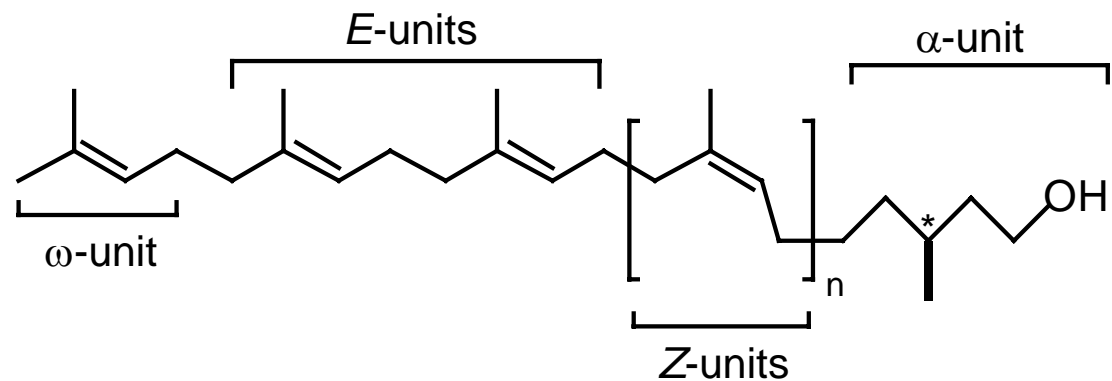
Figure 4. Effects of age on the concentration of dolichol in the liver of male (open squares) and female (closed squares) Sprague Dawley rats. Means \pm Standard Error of the Mean of at least 6 cases are given. Dolichol was extracted and assayed as described in (25).

Scheme 2. Biosynthetic pathway of dolichol.



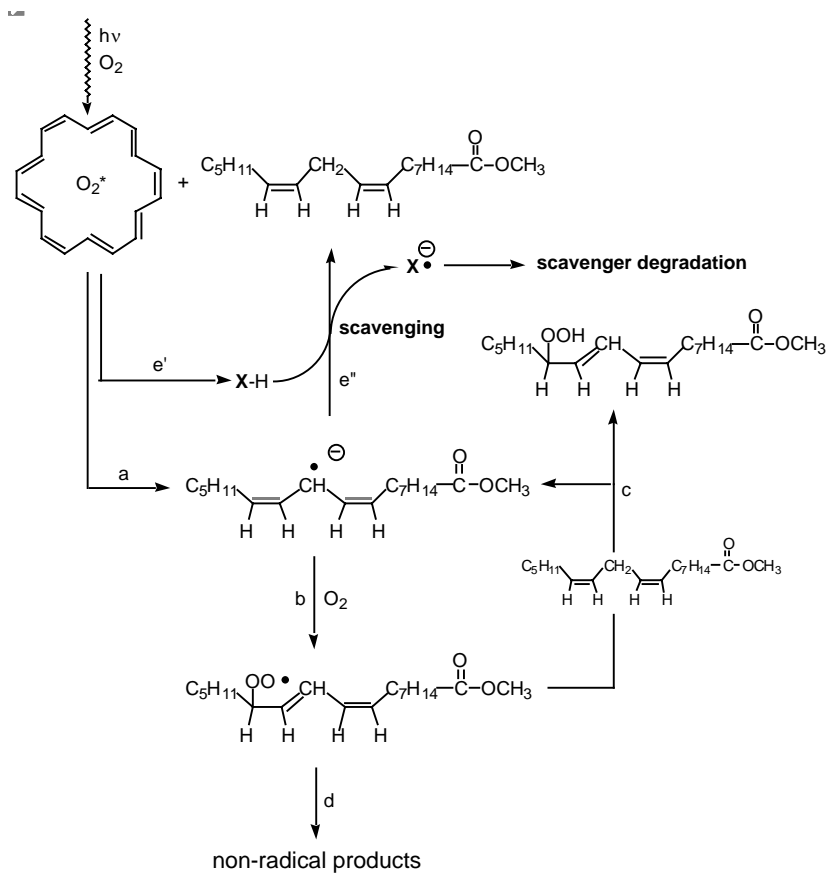
**A QUESTION IS: MAY DOLICHOL BE INVOLVED IN
FREE RADICAL METABOLISM ?**

Scheme 1. Structure of dolichol and of Vit E



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Peroxidation of Methyl Linoleate



- Solution of Photosensitizer, Methyl Linoleate, and Dolichol in Acetone- d_6
- Measurement of 1H -NMR spectra at different times

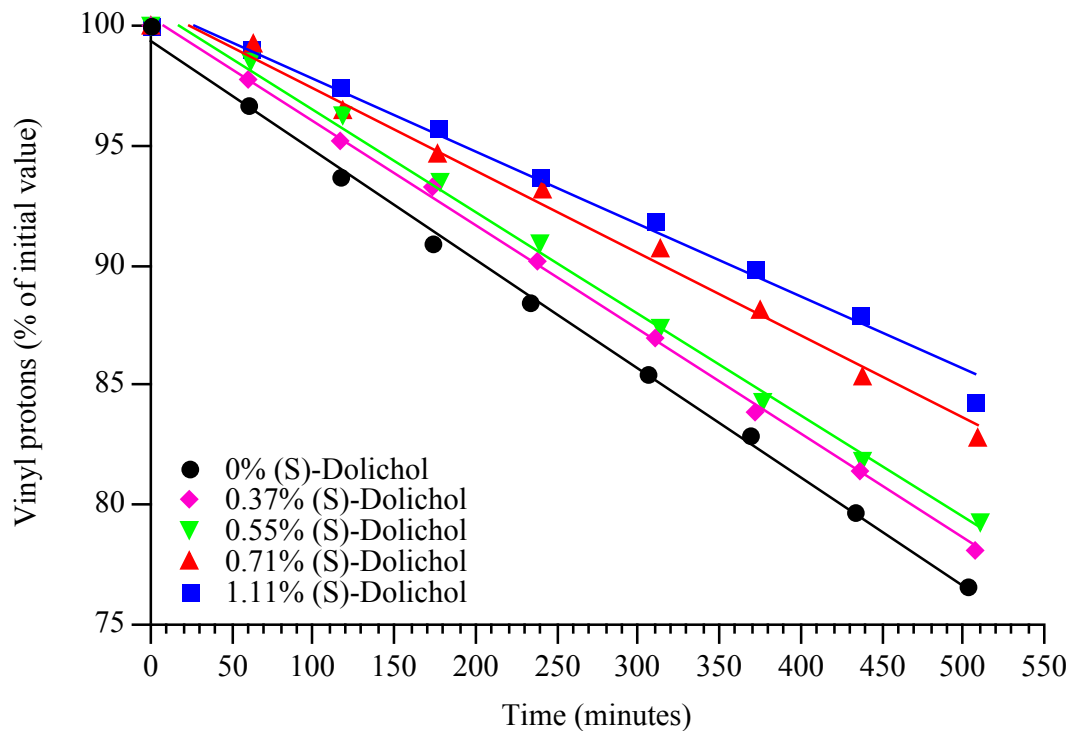


Figure 4. Normalized amount of vinyl protons of methyl linoleate as function of time upon photoinduced peroxidation.

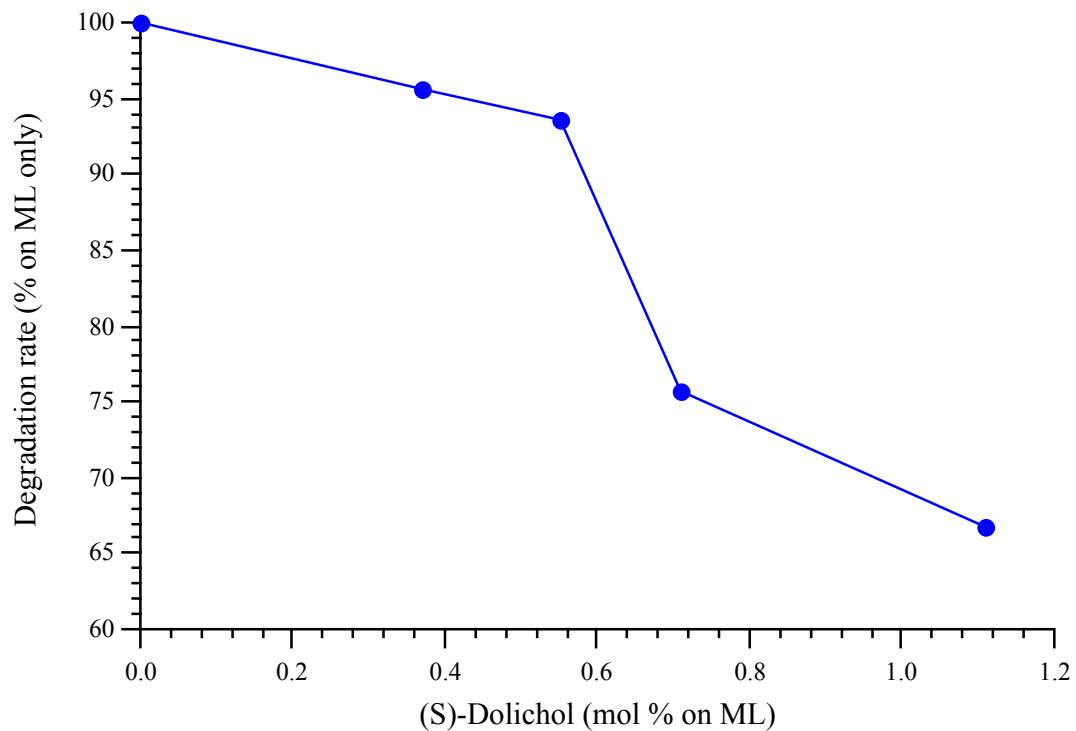
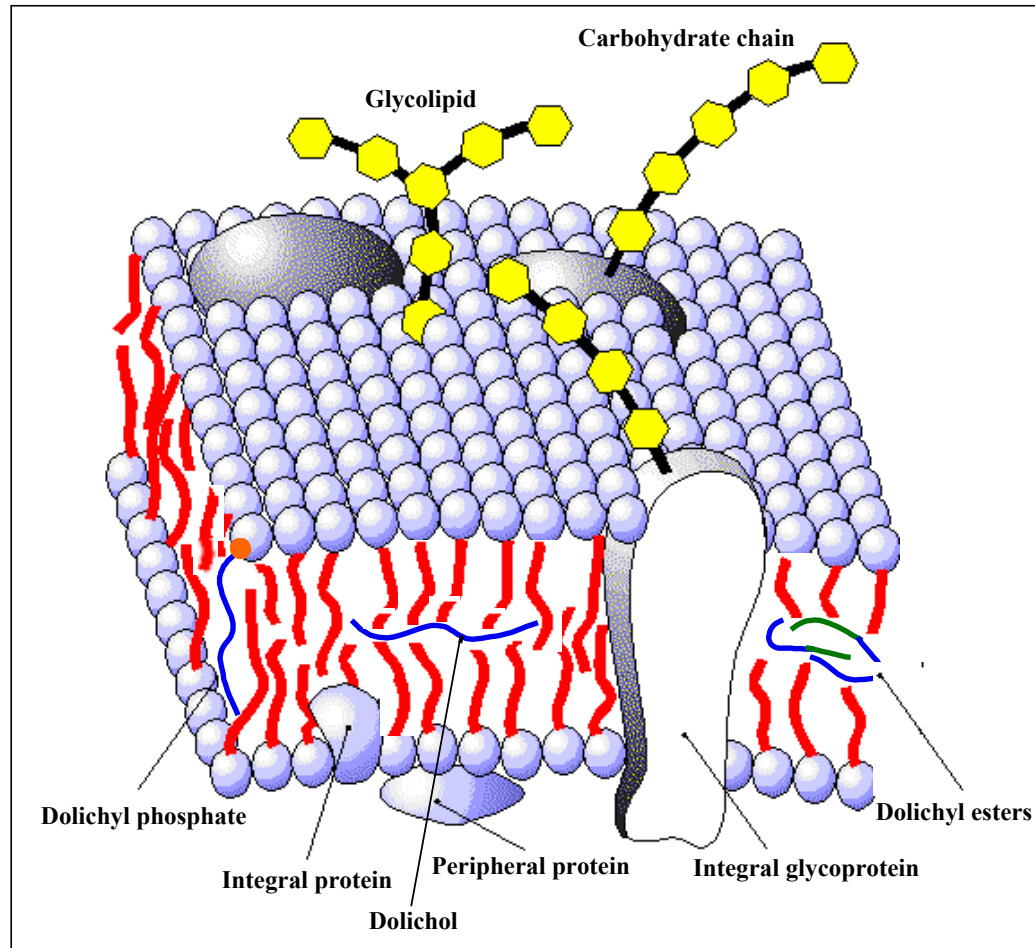
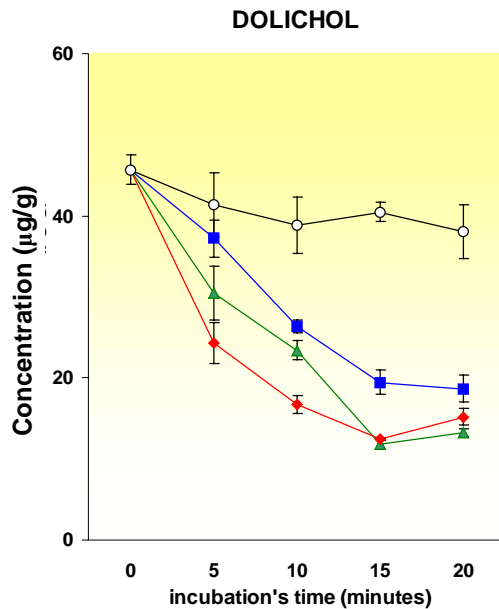


Figure 5. Experimental relationship between peroxidation rate of methyl linoleate and (S)-Dolichol concentration.

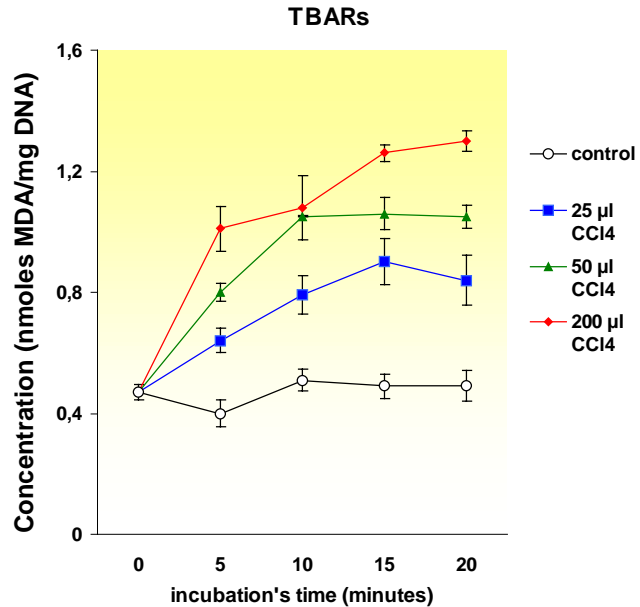
Dolichol: Localization in Membranes



► Treatment with CCl₄ caused a rapid, dose-dependent, decrease in Dolichol and simultaneous dose-dependent increase in TBARs. Release of TBARs is close to 0.1 nmoles/nmoles of Dolichol consumed.



Effects of CCl₄ on D in isolated liver cells in 3 months old rats. Means are given. Vertical bars represent SEM. Statistical analysis (ANOVA): time p<.0001, doses p<.0001, interaction time*dose p<.0001; Tukey test: time 0 minutes vs all times, 10-15 and 20 minutes vs 5 minutes; dose 0 µl vs all doses, 200 µl vs 25 and 50 µl

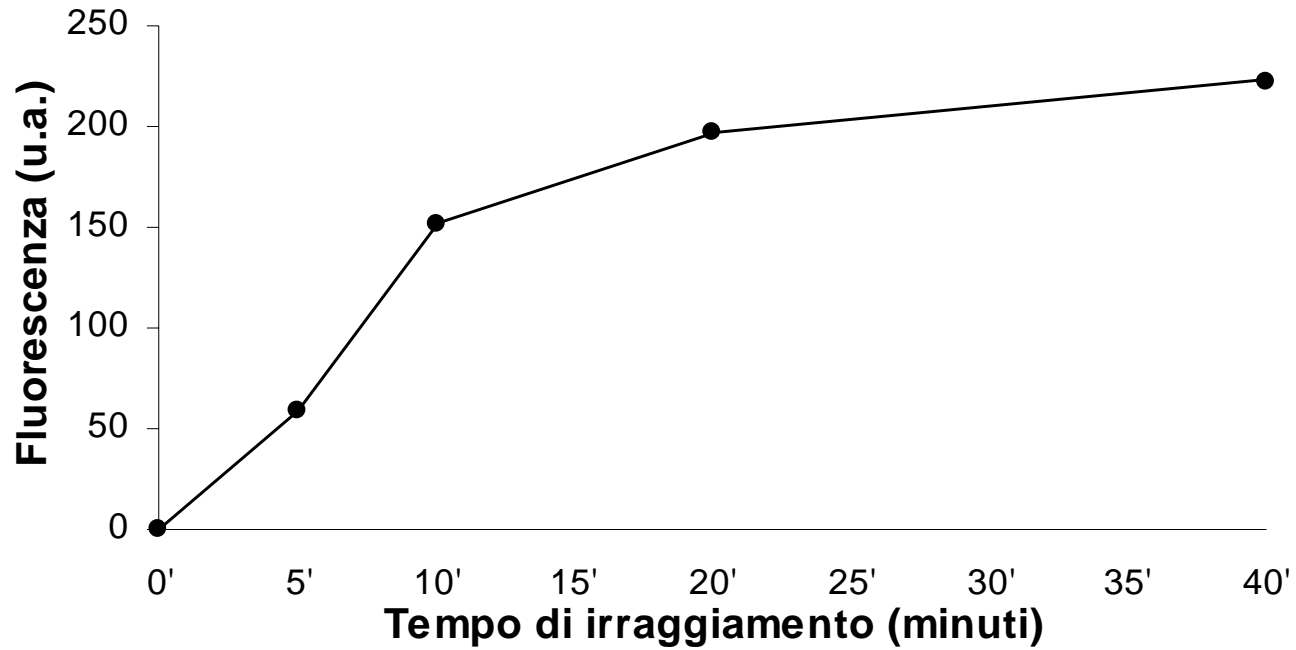


Effects of CCl₄ on TBARs production in isolated liver cells in 3 months old. Means are given. Vertical bars represent SEM. Statistical analysis (ANOVA): time p<.0001, doses p<.0001, interaction time*dose p<.0001. Tukey test: time 0 minutes vs all times, 10-15 and 20 minutes vs 5 minutes; dose 0 µl vs all doses, 200 µl vs 25 and 50 µl.

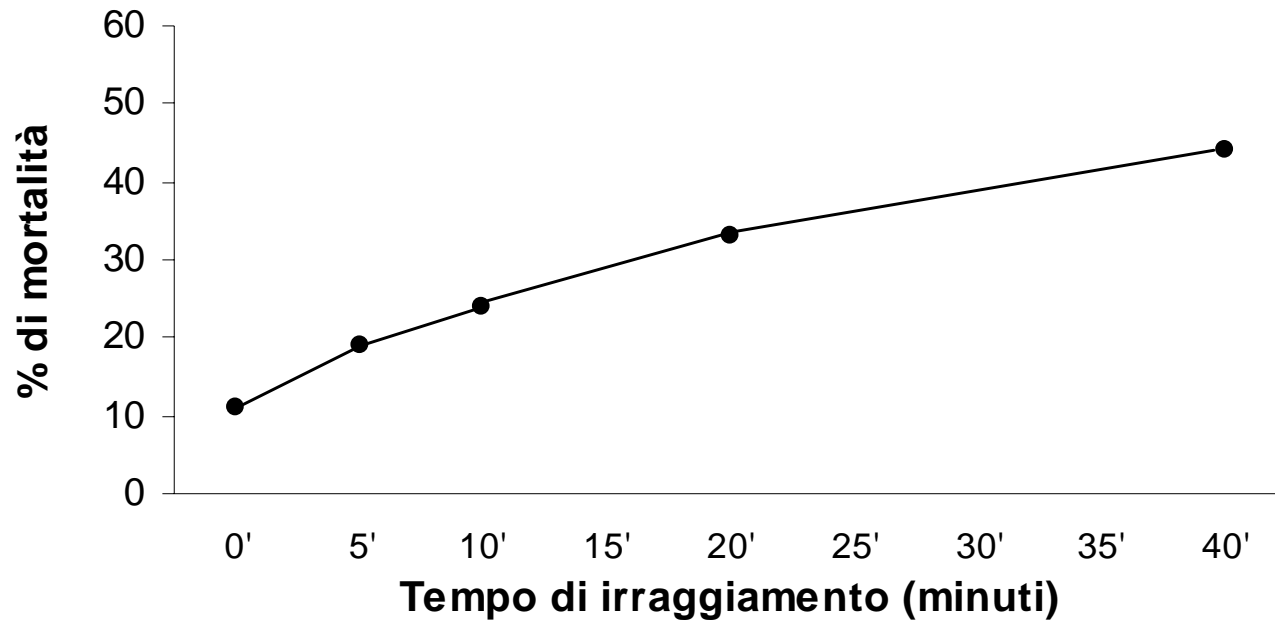
► Phospholipid fatty acids were assayed, in the same cells, after 10 and 60 minutes incubation. The obtained values are given in next table as percent abundance of fatty acids. No significant effects of CCl₄ were detected.

PUFA(%)	Control	10 minutes		60 minutes	
		25 µl CCl ₄	200 µl CCl ₄	25 µl CCl ₄	200 µl CCl ₄
14:0	0.13 ± 0.023	0.13 ± 0.016	0.11 ± 0.019	0.09 ± 0.002	0.09 ± 0.004
14:1	0.23 ± 0.020	0.23 ± 0.010	0.22 ± 0.007	0.16 ± 0.032	0.18 ± 0.016
16:0	13.70 ± 0.071	13.44 ± 0.303	13.32 ± 0.292	13.04 ± 0.162	13.35 ± 0.200
18:0	31.26 ± 0.885	30.53 ± 0.674	30.63 ± 0.728	29.34 ± 0.634	29.98 ± 0.401
18:1 ₉	1.65 ± 0.017	1.65 ± 0.077	1.61 ± 0.065	1.68 ± 0.006	1.72 ± 0.010
18:1 ₇	1.54 ± 0.030	1.53 ± 0.023	1.64 ± 0.024	1.44 ± 0.026	1.46 ± 0.031
18:2 ₆	12.38 ± 0.312	12.13 ± 0.284	12.20 ± 0.169	12.52 ± 0.209	12.27 ± 0.0.05
20:0	0.08 ± 0.005	0.09 ± 0.005	0.07 ± 0.012	0.07 ± 0.016	0.06 ± 0.003
18:3 ₆	0.14 ± 0.001	0.14 ± 0.009	0.14 ± 0.008	0.14 ± 0.019	0.16 ± 0.0002
20:1 ₉	0.08 ± 0.003	0.10 ± 0.011	0.09 ± 0.018	0.10 ± 0.006	0.09 ± 0.006
20:2 ₆	0.18 ± 0.001	0.16 ± 0.027	0.19 ± 0.008	0.17 ± 0.001	0.17 ± 0.003
22:0	0.27 ± 0.022	0.31 ± 0.020	0.27 ± 0.030	0.24 ± 0.011	0.15 ± 0.004
20:3 ₆	0.46 ± 0.017	0.50 ± 0.036	0.48 ± 0.039	0.43 ± 0.014	0.43 ± 0.015
20:4 ₆	22.78 ± 0.326	22.54 ± 0.196	22.43 ± 0.343	23.57 ± 0.299	23.55 ± 0.287
20:5 ₃ /22:4 ₆	0.24 ± 0.015	0.25 ± 0.024	0.24 ± 0.025	0.311 ± 0.004	0.30 ± 0.004
24:0	0.83 ± 0.038	0.89 ± 0.022	0.78 ± 0.057	0.73 ± 0.042	0.46 ± 0.014
22:5 ₃	0.77 ± 0.024	0.76 ± 0.040	0.73 ± 0.030	0.90 ± 0.017	0.90 ± 0.013
22:6 ₃	12.25 ± 0.024	12.02 ± 0.368	12.31 ± 0.486	12.81 ± 0.484	12.77 ± 0.430
Other	2.14 ± 0.056	2.58 ± 0.262	2.55 ± 0.308	2.23 ± 0.100	1.90 ± 0.145

Effetti dell'irraggiamento UV sull'intensità di fluorescenza DCF



Effetto della durata dell'irraggiamento sulla percentuale di cellule non vitali



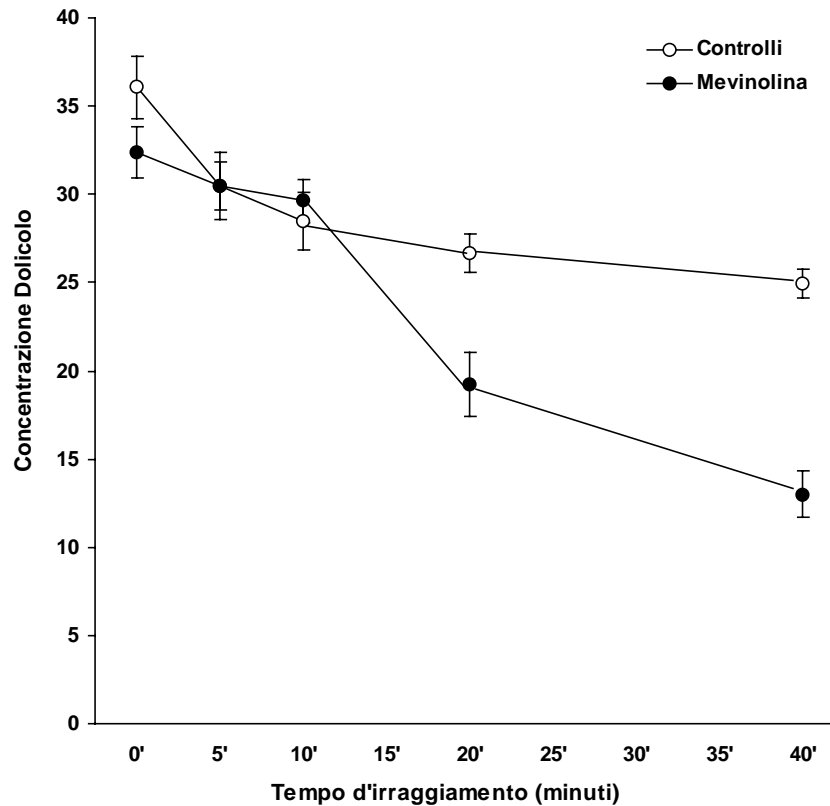
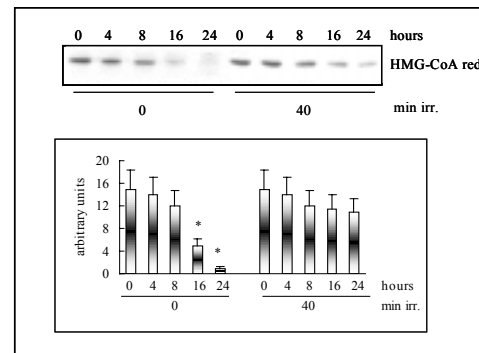
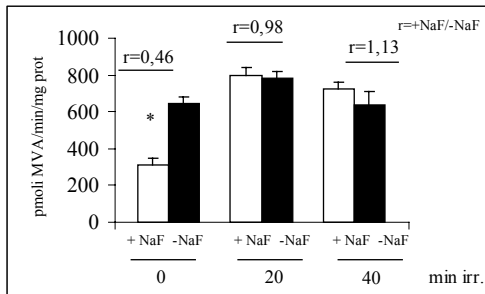
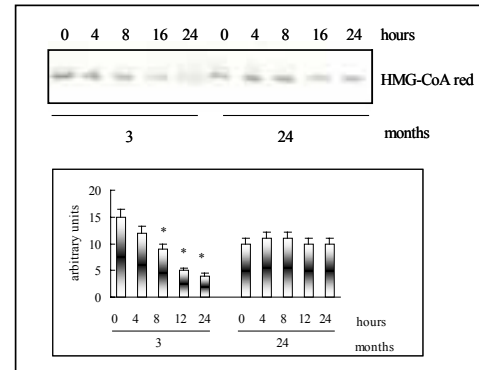
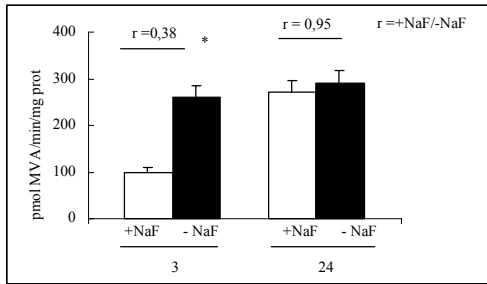
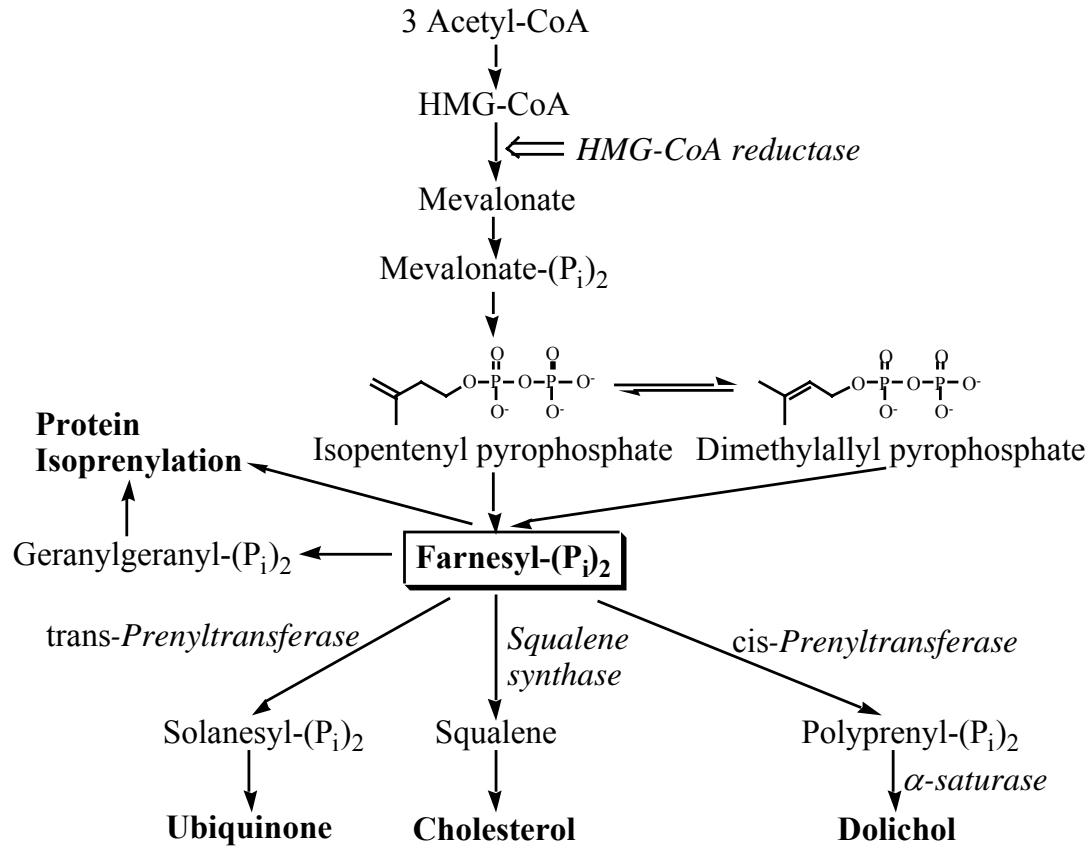


Figura 41: Effetti dell'irraggiamento UV-B e della mevinolina (3µM) sui livelli di Dolicolo di epatociti di ratto di 3 mesi d'età. Nel grafico sono riportate le medie \pm ESM della concentrazione di Dolicolo (μg Dolicolo/g di cellule) in funzione del tempo d'irraggiamento. Analisi statistica (Anova) *Effetti del tempo* Test F $p < .0001$, Tukey Test ($p < .05$) 0' vs 10', 20' e 40'; 5' e 10' vs 20' e 40' *Effetti del trattamento* Test F $p < .0001$. *Interazione tempo*trattamento* Test F $p < .0003$.

a
Fig 2 HMG-CoA reductase activity and degradation rate in younger and older not-irradiated or UV-radiated liver cells



Scheme 2. Biosynthetic pathway of dolichol.



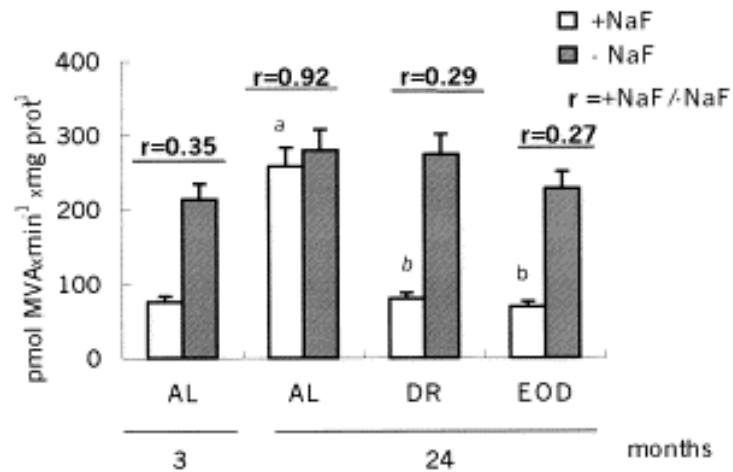


Fig. 1. HMGCoA reductase activity in liver of 3- and 24-month-aged AL or DR or EOD fed rats. Microsomes were prepared in presence or in absence of NaF (50 mM). The data represent the mean±S.D. of at least four different experiments. *r* represents the ratio between HMGCoA reductase activity in presence or in absence of NaF. $P < 0.001$ performed by Bonferroni's *t*-test: (a) significantly different from 3 months, (b) significantly different from AL 24 months.

months.

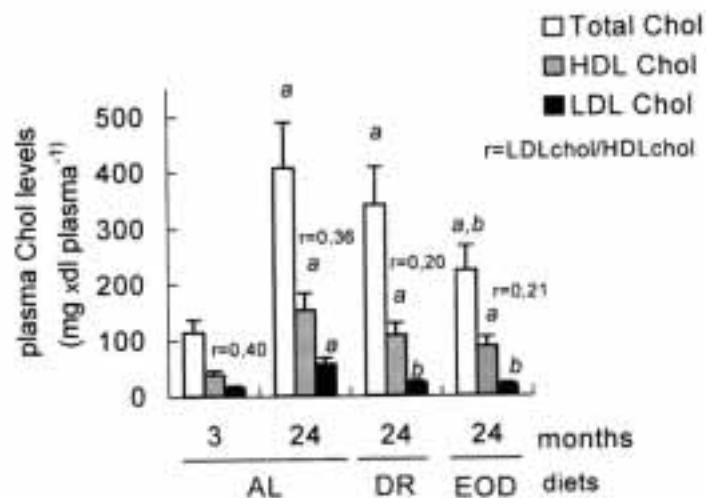
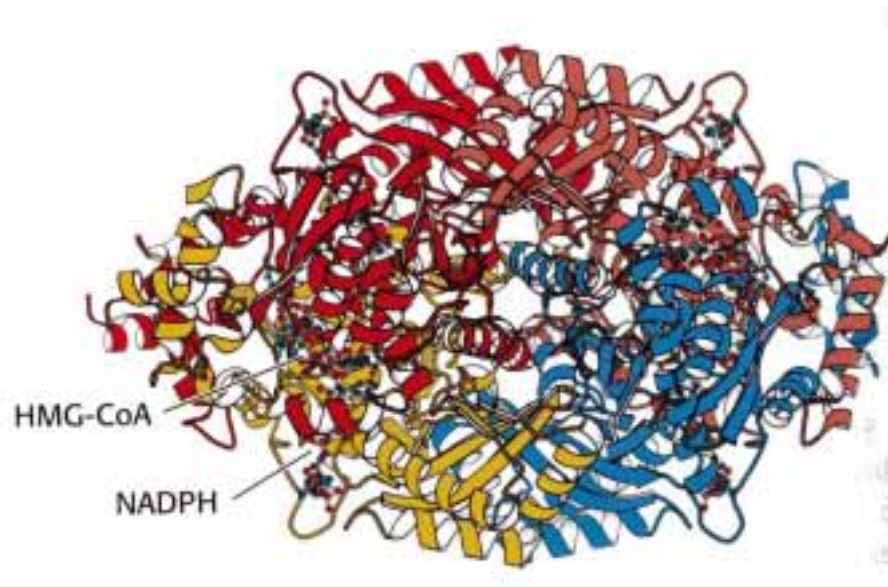


Figure 5. Total Chol, HDL-Chol and LDL-Chol levels in plasma of 3-month-old AL and 24-month-old AL or DR or EOD fed rats. The data represent the mean \pm S.D. of at least four different experiments. 'r' represents the ratio between LDL/chol and HDL/chol. $P < 0.001$ performed by Bonferroni's *t*-test: (a) significantly different from 3 months, (b) significantly different from AL 24 months.



BOTH ACUTE FREE-RADICAL GENERATION AND AGING MAY AFFECT THE MEMBRANE-DOMAIN OF HMGCoA REDUCTASE AND MODULATE FUNCTION AND DEGRADATION IN ORDER TO DESENSITIZE THIS BIPARTITE ENZYME TO CONTROL BY CHOLESTEROL AND KEEP SYNTHESIS OF DOLICHOL AND UBIQUINONE HIGHER, AND ENHANCE MEMBRANE ANTI-OXIDANT DEFENSES.

INFORMATION MIGHT PERHAPS HELP CARDIOLOGISTS TO MAKE BETTER AND SAFER USE OF STATINS

HMGCoA reductase is controlled in multiple ways (Berg JM et al., Biochemistry 5th ed, Freeman Co, 2002, p 726-727) via:

- 1. Rate of synthesis of mRNA (controlled by the sterol regulatory element binding protein, sensing cholesterol levels)**
- 2. Rate of translation of reductase mRNA, inhibited by metabolites of mevalonate and dietary cholesterol.**
- 3. Rate of enzyme degradation (controlled by cholesterol concentration in membranes, sensed by the membrane sensing domain)**
- 4. Phosphorylation by AMP-activated kinase (inhibitory)**

Novel findings indicate that steps 3 and 4 may be inhibited by Free-radical attack and aging.

THERE IS EVIDENCE THAT HYPER-
CHOLESTEROLEMIA IS A SIGN OF
AGING AND THAT ATHEROSCLEROSIS
IS A COMPLICATION OF AGING

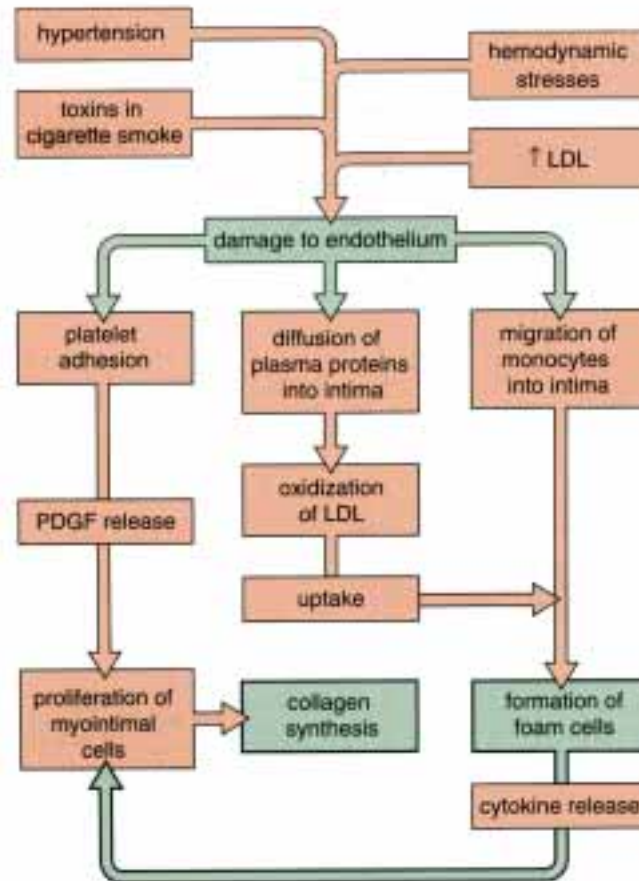
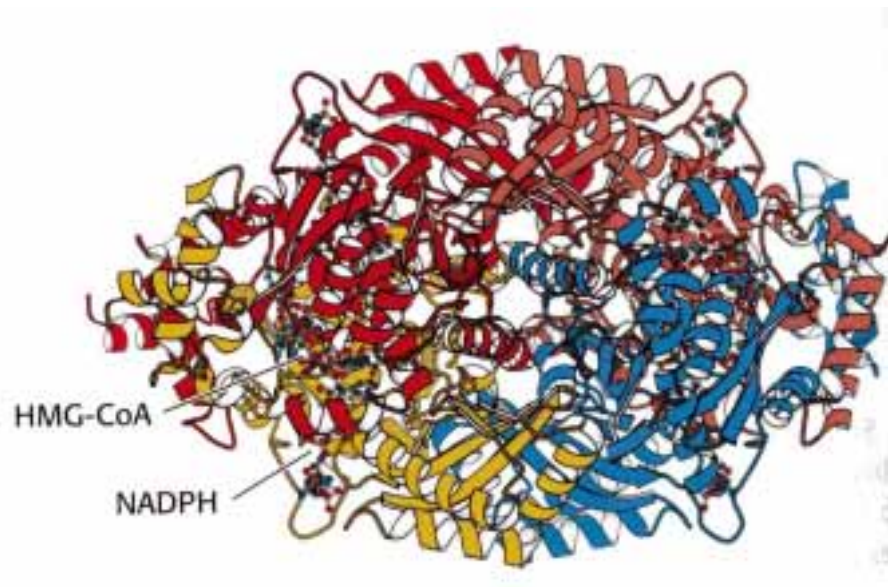


Fig.10.13 Flow-chart summarizing events involved in pathogenesis of atheroma.



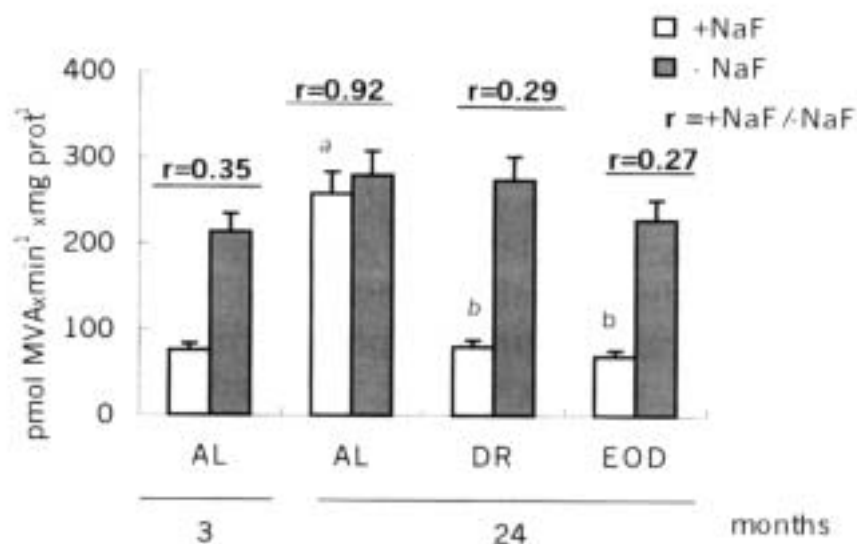
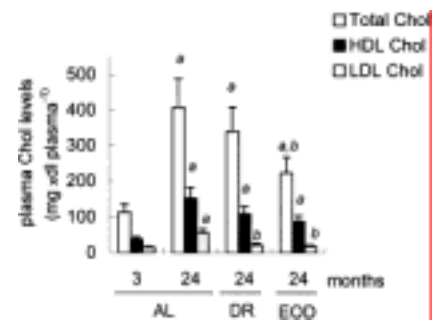
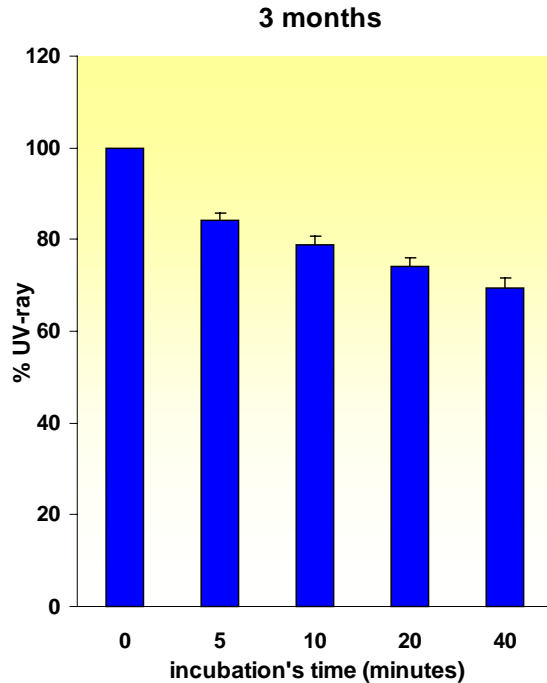


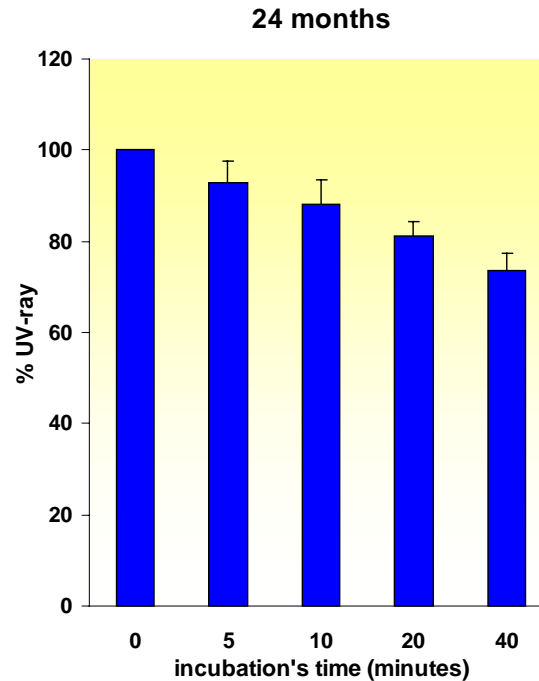
Fig. 1. HMGCoA reductase activity in liver of 3- and 24-month-old AL or DR or EOD fed rats. Microsomes were prepared in presence or in absence of NaF (50 mM). The data represent the mean \pm S.D. of at least four different experiments. r represents the ratio between HMGCoA reductase activity in presence or in absence of NaF. $P < 0.001$ performed by Bonferroni's t -test: (a) significantly different from 3 months, (b) significantly different from AL 24 months.



► Radiation caused a significant breakdown of Dolichol
Here again it appears that in older cells breakdown after a free radicals attack might be smaller and delayed.



Means are given. Vertical bars represent SEM.
Statistical analysis (ANOVA): $p < .0001$; Tukey test: time 0 minutes vs all time, 10 minutes vs 40 minutes, 20 and 40 minutes vs 5 minutes.



Means are given. Vertical bars represent SEM.
Statistical analysis (ANOVA): $p < .002$; Tukey test 0 minutes vs 20 and 40 minutes, 40 minutes vs 5 minutes

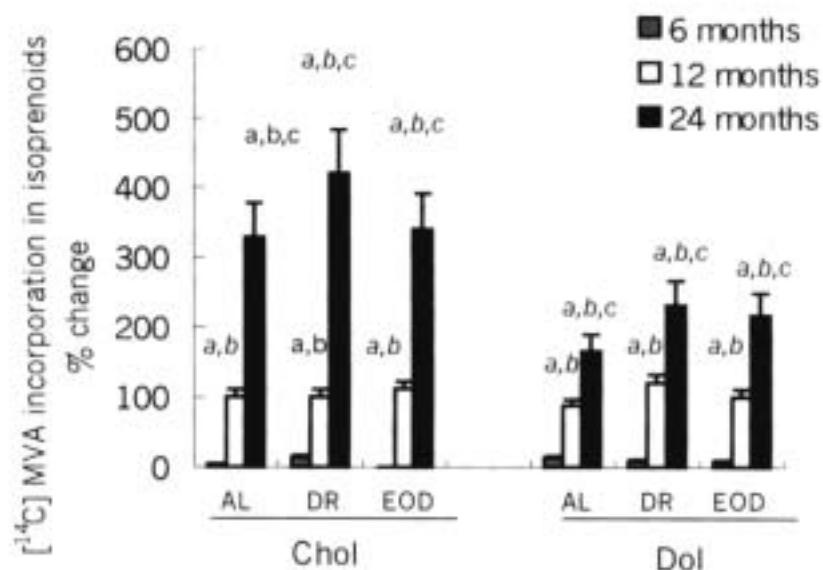


Fig. 2. Incorporation of [^{14}C]MVA in liver Chol and Dol of 6-, 12-, and 24-month-aged AL or DR or EOD fed rats. Data are expressed as percentage change with respect to the control rats (3-month-aged). The data represent the mean \pm S.D. of at least four different experiments. $P < 0.001$ performed by Bonferroni's t -test: (a) significantly different from 3 months, (b) significantly different from 6 months, (c) significantly different from 12 months.

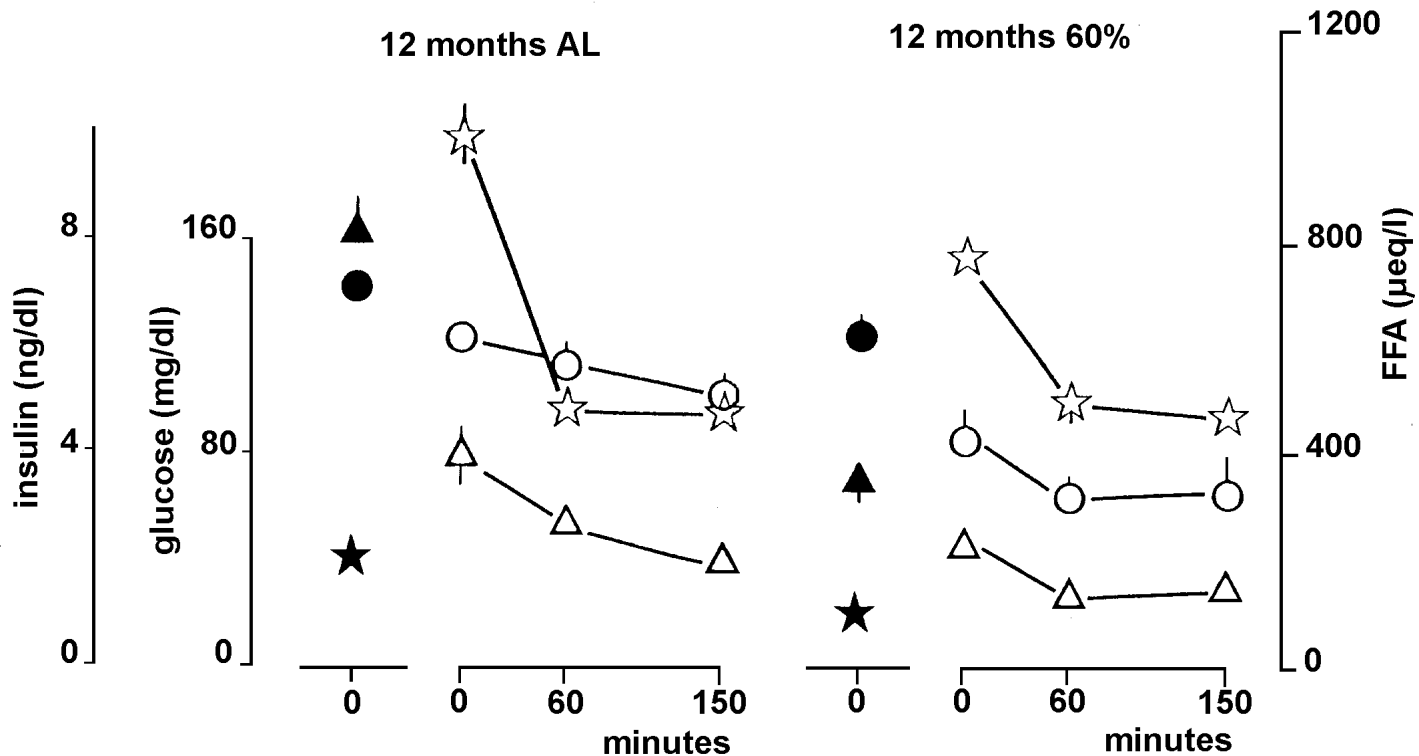
EPATOCITI ESPOSTI A RADIAZIONE UV DI RATTI SPRAGUE-DAWLEY MASCHI DI 3 MESI DI ETÀ

Acidi grassi	Controlli 0'	Trattati 40'	Controlli carenti di Vit E 0'	Trattati 40'	Controlli carenti di PUFA 0'	Trattati 40'
14:0	0.14±0.029	0.12±0.007	0.15±0.011	0.14±0.007	0.35±0.016	0.39±0.027
16:0	17.0±0.25	17.5±0.14	17.2±0.45	17.8±0.37	15.4±0.24	16.3±0.09 ^A
18:0	22.8±0.32	23.1±0.28	23.5±0.39	24.4±0.44	24.3±0.27	24.2±0.41
20:0	0.08±0.003	0.08±0.003	0.05±0.014	0.07±0.003	0.20±0.013	0.19±0.013
22:0	0.22±0.006	0.23±0.004	0.24±0.005	0.26±0.005 _B	0.35±0.057	0.37±0.039
24:0	0.69±0.016	0.71±0.002	0.66±0.033	0.75±0.017 _B	0.63±0.011	0.64±0.023
Saturi	41.0	41.8^A	41.8	43.4	41.3	42.0
14:1	0.17±0.017	0.15±0.004	0.10±0.006	0.10±0.002	0.08±0.004	0.08±0.004
18:1 ₇	2.2±0.14	2.3±0.06	2.5±0.04	2.6±0.07	3.3±0.10	3.4±0.08
18:1 ₉	1.9±0.10	2.0±0.09	2.0±0.08	2.1±0.08	6.8±0.07	7.2±0.16 ^B
20:1 ₉	0.08±0.022	0.07±0.017	0.03±0.017	0.06±0.018	0.00±0.000	0.00±0.000
18:2 ₆	13.0±0.08	13.6±0.43	13.6±0.24	13.7±0.33	6.2±0.05	6.2±0.12
20:2 ₆	0.31±0.012	0.31±0.007	0.17±0.006	0.18±0.004	0.03±0.004	0.04±0.013
Oligo Insaturi	17.7	18.4	18.4	18.8	16.4	16.9
Mono-/Di-Insaturi	0.33±0.017	0.33±0.016	0.34±0.003	0.35±0.003_B	1.6±0.03	1.7±0.05
18:3 ₃	0.08±0.018	0.09±0.014	0.08±0.016	0.05±0.022	0.03±0.004	0.03±0.007
18:3 ₆	0.07±0.004	0.07±0.003	0.12±0.005	0.11±0.002	0.24±0.024	0.30±0.013
20:3 ₆	0.45±0.006	0.47±0.017	0.31±0.010	0.32±0.017	0.96±0.040	0.96±0.039
20:4 ₆	27.2±0.25	24.7±0.35 ^A	27.8±0.34	24.7±0.37 ^A	22.7±0.26	20.4±0.17 ^A
20:5 ₃ / 22:4 ₆	0.17±0.005	0.15±0.010	0.09±0.008	0.07±0.006	0.14±0.006	0.11±0.019
22:5 ₃	0.95±0.014	0.95±0.027	1.1±0.04	0.98±0.034	0.29±0.013	0.30±0.013
22:6 ₃	9.7±0.09	8.0±0.39 ^A	7.6±0.20	6.2±0.28 ^A	5.6±0.12	4.8±0.12 ^A
Poli Insaturi	38.6	34.4^A	37.1	32.4^A	30.0	26.9^A
Picchi non identificati	2.7±0.16	5.4±0.14^A	2.7±0.10	5.4±0.16^A	12.3±0.24	14.2±0.44^A
Famiglie						
n-9	2.0	2.1	2.0	2.1	6.8	7.2 ^B
n-7	2.2	2.3	2.5	2.6	3.3	3.4
n-6	41.0	39.1 ^A	42.0	39.0 ^A	30.2	27.9 ^A
n-3	10.8	9.0 ^A	8.8	7.3 ^A	10.8	5.1 ^A

I risultati sono dati come composizione % di acido grasso sugli acidi grassi totali dei fosfolipidi in epatociti di ratto. Medie ± S.E della media di almeno 5 casi. Significatività della differenza da ratti di controllo: ^A P < 0.01; ^B P < 0.05.

Effects of the intraperitoneal injection of 3,5-dimethylpyrazole (DMP, 12 mg/Kg b.w.) on the FFA (★,☆), glucose (●,○) and insulin (▲,△) plasma levels.

Data obtained with ad libitum fed non fasted rats are given as an additional control (black symbols)



No Truth to the Fountain of Youth

Fifty-one scientists who study aging have issued a warning to the public: no anti-aging remedy on the market today has been proved effective. Here's why they are speaking up

By S. Jay Obshansky,
Leonard Hayflick
and Bruce A. Carnes

Illustrations by
J. W. Stewart



Efforts to combat aging and extend human life date at least as far back as 1900 B.C., and self-proclaimed experts have found anti-aging elixirs ever since. Indeed, the prospect of immortality has always had universal appeal, spurring Alexander the Great and Ponce de León to search for the legendary Fountain of Youth and leading alchemists' desire to transmute gold (once believed to be the most potent anti-aging substance in existence). But the hawking of anti-aging "therapies" has taken a particularly troubling turn of late. Disturbingly large numbers of entrepreneurs are luring gullible and frequently desperate customers of all ages to "longevity" clinics, claiming a scientific basis for the anti-aging products they recommend and, often, sell. At the same time, the Internet has enabled those who seek face from supposed anti-aging products to reach new consumers with ease.

Alarmed by these trends, scientists who study aging, including the three of us, have issued a position statement containing this warning: no carefully marketed inter-

Scheme 2. Biosynthetic pathway of dolichol.

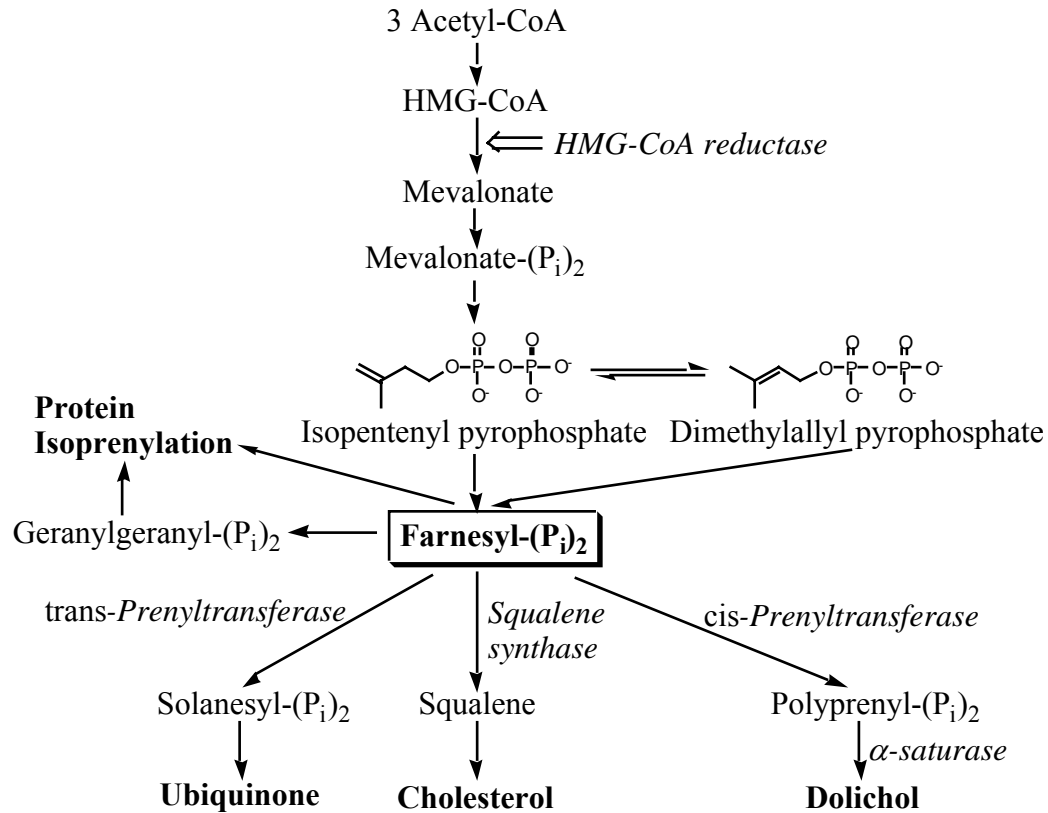


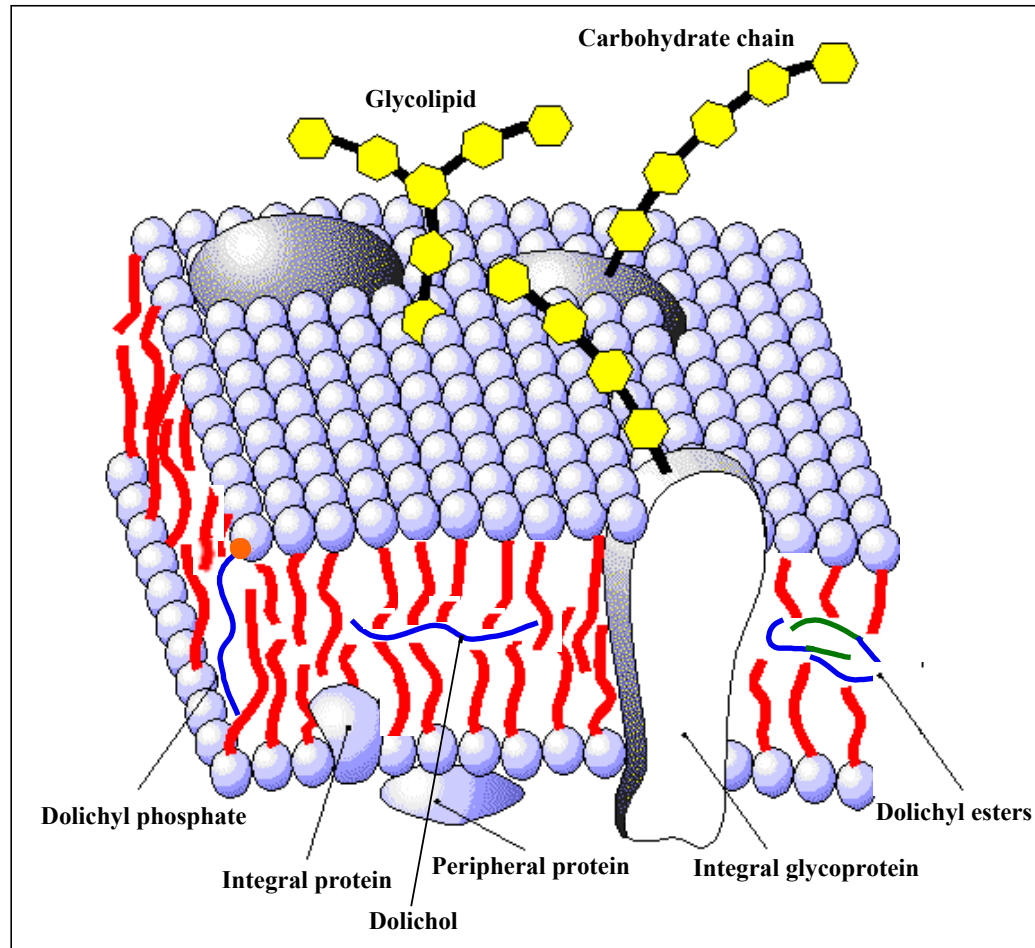
TABLE I

Abundance of Vitamin E, Dolichol and unsaturated fatty acids in the liver tissue of ad libitum fed male Sprague Dawley rats.

Lipid	3-mo-old rat	24-mo-old rat
Vitamin E	1	1
Dolichol	1.3	2.3
Unsaturated Fatty Acids in Phospholipid	617	898
Fatty acids in Phospholipid	1087	1591

Molar ratios are given. The concentration of Vitamin E was significantly higher in older than in younger liver tissue, but values are given equal to one in both cases, Vitamin E being the reference. These data were obtained by Drs Arianna Manfrini, Ilaria Parentini, and Ilaria Tamburini, in this Centro di Ricerca di Biologia e Patologia dell'Invecchiamento.

Dolichol: Localization in Membranes



Probability of survival in ad libitum and 10%, 25% and 40% food restricted male Sprague-Dawley rats (from Duffy et al., 2001)

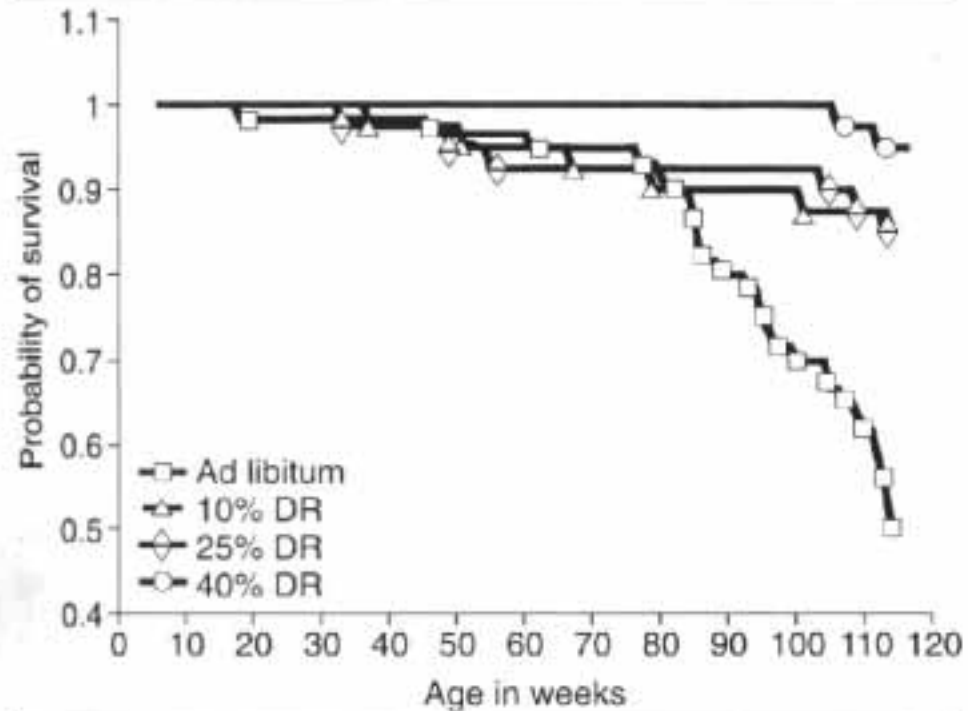
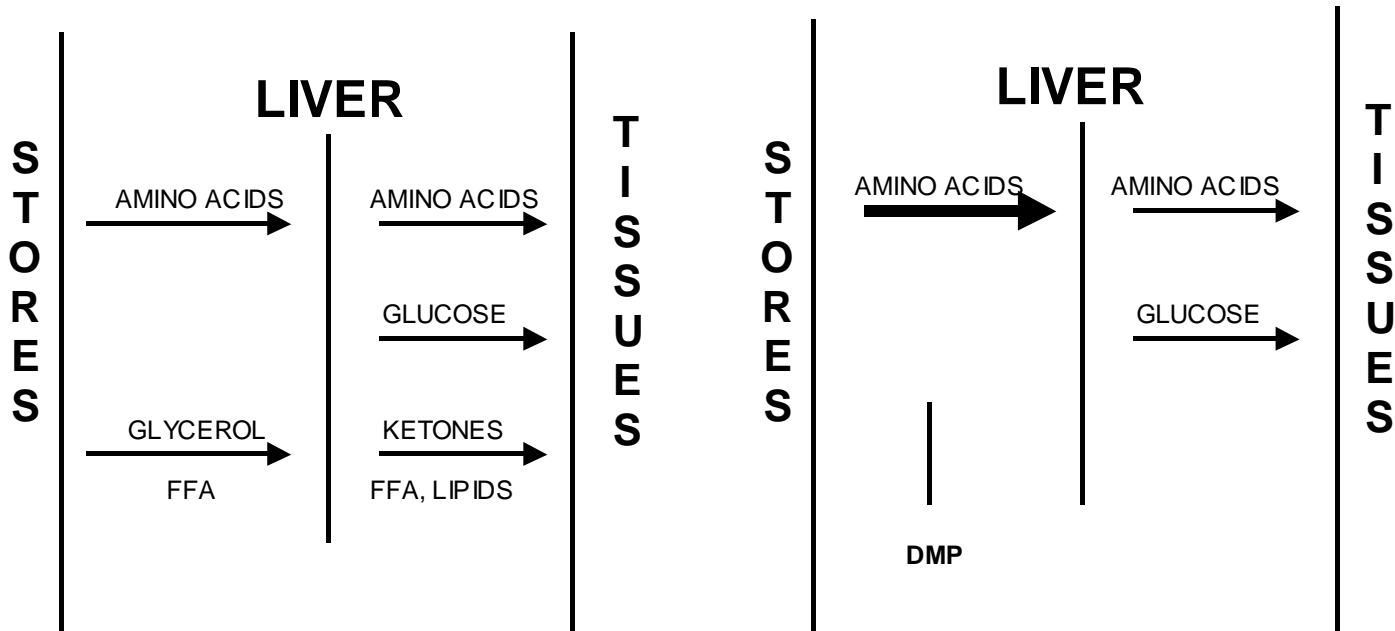


Figure 1 - Kaplan-Meier curves representing the probability of survival in ad libitum (AL) and 10%, 25%, and 40% dietary restricted (DR) male SD rats.

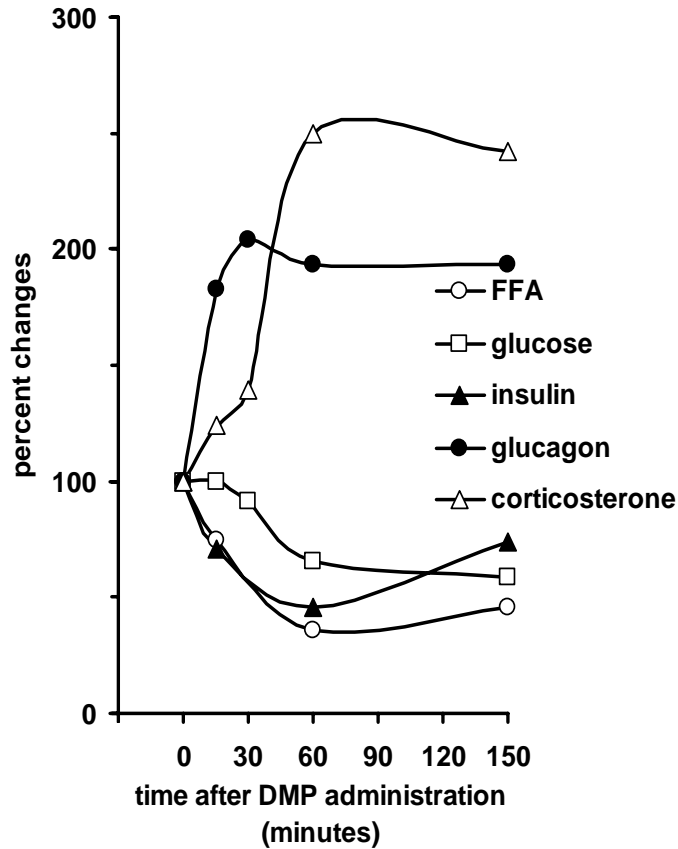
Table 1. Anti-ageing cell repair mechanism

<i>Mechanism</i>	<i>Predictable effects or malfunctioning</i>	<i>Direct effect by nutrition</i>
<u><i>Molecular level</i></u>		
DNA repair	Accumulation of altered DNA	Not proved
Proteolysis	Accumulation of altered protein	Not proved (proteosomal) Yes (autophagic-lysosomal)
Fatty acid replacement	Changes in phospholipid	Not proved
<u><i>Subcellular level</i></u>		
Autophagy	Abnormal accumulation of membrane lipids (dolichol); accumulation of altered organelles (mitochondria, peroxisomes) and increased oxidative stress	Yes (stimulated by lower amino acids, insulin, IGF-1 plasma levels)
<u><i>Cell and Tissue level</i></u>		
Apoptosis	Accumulation of defective cells in tissues	Not proved

Autophagy can be stimulated *in vivo* by the administration of antilipolytic drugs to starved animals



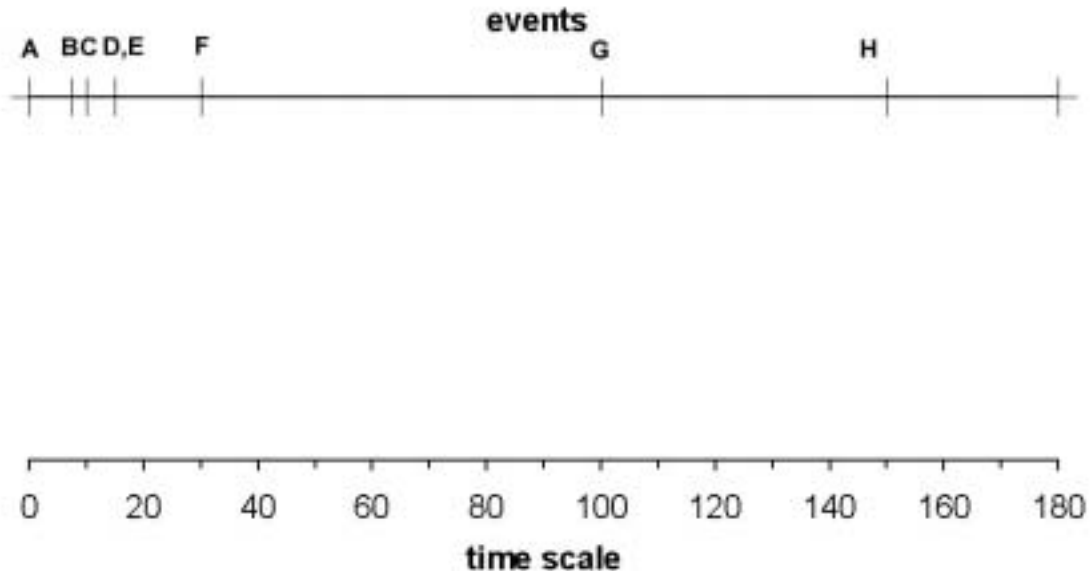
Changes in plasma levels after the administration of antilipolytic drugs to starved animals



E. Bergamini et al., A new method for the investigation of endocrine-regulated autophagy and protein degradation in rat liver. *Exp Mol Pathol.* 1993 Aug;59(1):13-26.

E. Bergamini et al. Endocrine and amino acid regulation of liver macroautophagy and proteolytic function. *Am J Physiol.* 1994 Jan;266(1 Pt 1):G118-22.

Time-sequence of the events after the administration of DMP



- A:** injection of the antilipolytic agent
- B:** decrease in plasma FFA levels
- C:** decrease in blood glucose levels
- D:** changes in glucagon and insulin plasma levels
- E:** the intraperitoneal administration of glutamine still prevents H
- F:** liver cells lysosomes are vacuolated
- G:** peroxisomes were detected in autophagic vacuoles
- H:** peroxisomal enzyme activities are significantly decreased in the liver tissue (from: Bergamini and Segal, 1987)

Aging: The Reality

Biomarkers of Aging: From Primitive Organisms to Humans

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Michael Forster,⁶ Howard Fillit,⁷ S. Mitchell Harman,⁸ Michael Hewitt,⁹ Mark Hyman,¹⁰
Kathleen Johnson,⁹ Evan Kligman,¹¹ Gerald McClearn,¹² James Nelson,¹³ Arlan Richardson,¹⁴
William Sonntag,¹⁵ Richard Weindruch,¹⁶ and Norman Wolf¹⁷

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³Biology of Aging Program, National Institute on Aging, Bethesda, Maryland.

⁴Metagenics, Inc., Gig Harbor, Washington.

⁵National Center for Toxicology Research, Jefferson, Arkansas.

⁶Texas College of Osteopathic Medicine, Fort Worth.

⁷The Institute for the Study of Aging, New York.

⁸Kronos Longevity Research Institute, Phoenix, Arizona.

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¹¹Desert Life Medical Plaza, Tucson, Arizona.

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¹³The University of Texas, San Antonio.

¹⁴The University of Texas, Health Science Center, San Antonio.

¹⁵Department of Physiology/Pharmacology, Wake Forest University School of Medicine, Winston-Salem, North Carolina.

¹⁶University of Wisconsin, Madison.

¹⁷University of Washington, Seattle.

Leading biologists and clinicians interested in aging convened to discuss biomarkers of aging. The goals were to come to a consensus, construct an agenda for future research, and make appropriate recommendations to policy makers and the public-at-large. While there was not total agreement on all issues, they addressed a number of questions, among them whether biomarkers can be identified and used to measure the physiological age of any individual within a population, given emerging information about aging and new technological advances. The hurdles to establishing informative biomarkers include the biological variation between individuals that makes generalizations difficult; the overlapping of aging and disease processes; uncertainty regarding benign versus pathogenic age-related changes; the point at which a process begins to do damage to the organism, and, if so, when does it occur; and when to distinguish critical damage from noncritical damage. Finally, and significantly, it is difficult to obtain funding for this research.

PUFA	5'	10'	20'	40'
20:4(n-6)	99	99	95	91
22:5(n-3)	99	98	95	100
22:6(n-3)	98	97	90	82

Tabella 2: Effetti dell'irraggiamento sui livelli di acidi grassi. **I risultati sono espressi come percentuale di variazione rispetto al tempo 0'**

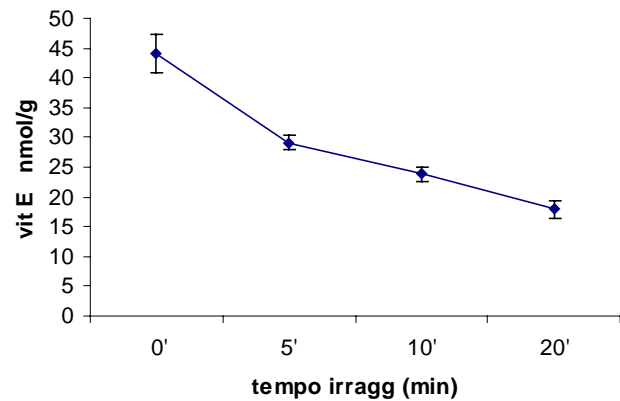


Figura 1: Effetti dell'irraggiamento sui livelli di Vitamina E in cellule epatocitarie di ratti. Nel grafico sono riportate le medie \pm ESM di Vitamina E in funzione del tempo d'irraggiamento. Analisi statistica (Anova) *Effetti del tempo* Tukey Test ($p < .05$) 0' vs tutti, 5' vs 20'

CALORIC RESTRICTION'S EFFECT IN RODENTS AND MONKEY

- a** EFFECTS INDICATIVE OF ALTERED GROWTH, DEVELOPMENT OR METABOLISM
 - Lower body temperatures
 - Later sexual maturation
 - Later skeletal maturation

- b** EFFECTS INDICATIVE OF IMPROVED HEALTH
 - Lower weight
 - Less abdominal fat

- c** EFFECTS INDICATIVE OF REDUCED RISK FOR AGE-RELATED DISEASES (SUCH AS DIABETES AND HEART DISEASE)
 - Greater sensitivity to insulin
 - Lower fasting insulin levels
 - Lower fasting glucose levels
 - Lower cholesterol and triglyceride levels
 - Lower insulin-like growth factor 1 levels
 - Higher levels of "good" (HDL) cholesterol
 - Slower decline in levels of the hormone DHEAS

- d** EFFECTS FOUND IN RODENTS BUT STILL UNDER INVESTIGATION IN MONKEYS
 - Later onset of age-related diseases (including cancer)
 - More cell suicide (which may help limit tumor growth)
 - Longer average life span
 - Longer maximum life span (a strong sign of slowed aging)

**EFFETTI DELL' IRRAGGIAMENTO UV-B E DELLA MEVINOLINA (3 μ M)
SUL CONTENUTO DI DOLICOLO DI EPATOCITI DI RATTO DI 3 MESI**

