Electron micrographs were taken at the same magnification of both the control and experimental soleus muscles, care being taken to avoid areas in the tenotomized muscle that abounded with rod shaped or streaming Z-structures.

Direct measurements were taken (independently, by 3 of the authors) from 20×25 cm enlargements of the EM plates, using a metric ruler for sarcomere and A band length, and a desk-top magnifier containing a reticle with 0.1-mm gradations for Z-widths. As Z-lines tend to be very irregular across the width of the myofibril, measurements were taken at multiple intervals along each Z-line in a micrograph. A total of 1641 control Z-line widths, and 1735 Z-line widths in tenotomized muscle were measured; the data are contained in the table.

Results and discussion. The table presents the results of our analysis of paired muscles from 6 tenotomized animals. The mean Z-disc width of the control contralateral leg was 109 nm, the same as in the younger series of rats on which we previously reported[6]. Those of the tenotomized leg averaged 106 nm, ranging from 102 to 111 nm, and there was no significant difference between the control and tenotomized legs. In contrast to younger rats, we conclude that tenotomy is without effect on the general Z-line widths of older rats. As in the former experiment, however, occasional Z-line streaming and rod formation was observed in the experimental muscles. This has also been reported in aged rats by Fukisawa[10].

Note from the table that although there was no difference in the width of the control and experimental A-band, the sarcomere length of the experimental leg (mean = 1.78 μm) was significantly shorter than the control (mean = 1.99 μm). This of course is a consequence of tenotomy, but it might suggest that had we measured the Z widths only in those sarcomeres of identical length, the experimental Z-band width would have perhaps been wider. However, we have not found any statistical correlation between Z-disc width and sarcomere length in any group of experimental or control animals. Z-line width, in vertebrate muscle, is not a function of sarcomere length. We hold therefore with the conclusion that tenotomy does not produce a generalized widening of the Z-line in older rats. This may, in some way, be related to the flat growth curves of these animals. The finding that rod formation and streaming are still present, suggests that the stimulus or underlying cause for generalized widening may be different from that or those responsible for the other morphological changes.

1 This investigation was supported in part by NIH grants 1-RO1-NS-14033 and 1-T32-07224.
2 A.G. Engel, in: Exploratory Concepts in Muscular Dystrophy and Related Disorders, p.398. Excerpta Medica Foundation, Amsterdam 1967.
3 H.Y. Meltzer, R.W. Kunel, J. Click and U. Yang, Neurology *26*, 853 (1976).
4 S.A. Shafig, M.A. Gorycki, S.A. Asiedu and A.T. Milhorat, Archs Neurol. Chicago *20*, 625 (1969).
5 J.S. Resnick, W.K. Engel and P.G. Nelson, Neurology *18*, 737 (1968).
6 D. Chase and W.C. Ullrick, Experientia *33*, 1177 (1977).
7 S.P. Bishop and C.R. Cole, Lab. Invest. *20*, 219 (1969).
8 J. Auber, J. Microsc. *8*, 197 (1969).
9 M.J. Legato, J. molec. Cell Cardiol. *1*, 425 (1970).
10 K. Fukisawa, J. neurol. Sci. *24*, 447 (1975).

α-Tocopherol reduces fluorescent age pigment levels in heart and brain of young mice

Patricia Kruk and Hildegard E. Enesco

Department of Biology, Sir George Williams Campus, Concordia University, Montreal (Québec H3G 1M8, Canada), 10 February 1981

Summary. The levels of fluorometrically measured lipofuscin, or age pigment, were significantly lower in the brain and the heart of α-tocopherol (vitamin E)-injected mice as compared to untreated control mice at 3 and at 5 months of age.

Lipofuscin, or fluorescent aging pigment, accumulates progressively with age in mammalian post-mitotic tissues, especially in brain and heart[1,2]. It is present even in young animals[3]. Tappel et al.[4] have demonstrated that lipofuscin accumulation is reduced by feeding antioxidants, including vitamin E, to 9-month-old mice for periods ranging from 0.6 to 1.2 years.

The questions which we asked in the present study were as follows: a) Can we alter lipofuscin accumulation in young mice at 3–5 months of age by α-tocopherol (vitamin E) administration? b) Can we alter lipofuscin levels if the α-tocopherol is administered by i.p. injections? c) Will the α-tocopherol have any effect on lipofuscin accumulation over relatively short periods in young animals?

Table 1. Data for heart. The level of lipid peroxidation fluorescence product in 0.2 g heart tissue is presented for mice injected with 1.1 IU α-tocopherol, as compared to control mice at 3 and 5 months of age. The standard deviation of the measurements is presented. The level of significance as determined by the t-test is presented as a function of age and as a function of treatment. N refers to number of measurements

Age	N	Fluorescence units (±SD) α-Tocopherol	Control	Difference (%)	Level of significance
3 months	3	43.00±2.82	53.00±5.10	23%	$0.10 < p < 0.05$
5 months	7	46.93±2.04	71.79±1.58	53%	$p < 0.001$
Level of significance		NS	$p < 0.001$		

Table 2. Data for brain. The level of lipid peroxidation fluorescence product in 0.2 g brain tissue is presented for mice injected with 1.1 IU α-tocopherol, as compared to control mice at 3 and 5 months of age. The standard deviation of the measurements is presented. The level of significance as determined by the t-test is presented as a function of age and as a function of treatment. N refers to number of measurements

Age	N	Fluorescence units (±SD) α-Tocopherol	Control	Difference (%)	Level of significance
3 months	6	47.75±3.29	56.75±3.51	19%	$p < 0.005$
5 months	14	47.18±4.13	73.64±5.43	56%	$p < 0.0001$
Level of significance		NS	$p < 0.001$		

Materials and methods. Female Swiss albino mice were obtained from Canadian Breeding Farms, St. Constant, Quebec. At 2 months of age 1 group of mice received an injection of α-tocopherol, while a 2nd group remained as controls. Measurements of lipid peroxidation fluorescent products were carried out when the mice were 3 and 5 months old. The Purina Chow used for the diet was standard mouse chow in pellet form.

In order to inject α-tocopherol, it was solubilized in carrier solution[5]. Experimental animals received 1 i.p. injection of 1.1 IU α-tocopherol in 0.1 ml of carrier. The dl-α-tocopherol used was in oil form, and was obtained from ICN Canada, Montreal, Quebec.

Lipid peroxidation levels were determined by fluorescence analysis. The brain and heart ventricles were excised and processed using the method of Tappel et al.[4]. Fluorescence measurements were made using an Aminco-Bowman spectrophotofluorometer standardized with quinine sulfate (1 mg/ml of 0.1 N H_2SO_4). Measurements were taken with the voltage set at 700 V and wavelength set at excitation 358 nm and emission 438 nm and a slit opening of 5. Measurements were obtained in fluorescent units per 0.2 g tissue sample.

Data were analyzed by means of the standard t-test.

Results and discussion. The results show that the level of lipid peroxidation fluorescent products is lower in the heart and the brain of tocopherol-treated mice as compared to control mice in both age groups examined. Table 1 illustrates the results for heart tissue. By 3 months of age the fluorescence level in the heart of the tocopherol-treated animals is clearly lower (by 23%) than that of the control animals, although there is only a borderline level of statistical significance. By 5 months of age, the level of fluorescence products in the heart is 53% lower than in the control animals, and the difference is highly significant.

Table 2 presents the data for brain. Both at 3 months of age and at 5 months of age, the level of lipid peroxidation fluorescent products is significantly lower in the animals injected with tocopherol than in the control mice.

When we examine the data for both brain and heart in relation to the age of the animals, we note that there is no increase in lipofuscin level between 3 and 5 months of age in the tocopherol-treated animals. In contrast, there is a highly significant increase in the level of fluorescent products between 3 and 5 months of age in the control animals.

The data presented show that the level of lipid peroxidation fluorescent products, or lipofuscin, can be significantly lowered in both brain and heart tissue of young mice by a single i.p. injection of tocopherol. In analyzing these results it is doubtless important to remember that tocopherol can be stored in various body tissues[6] and that parenteral doses are eliminated slowly[7]. Thus the tocopherol that was injected may have gone into a storage pool to be utilized gradually over a period of time. Or, once the level of tocopherol was elevated by the supplementary dose, it may have remained at a new and higher plateau since the animals were receiving an accepted level of tocopherol through the Purina Chow diet. The results can be interpreted in relation to the free radical theory of aging[8]. The presence of tocopherol is believed to reduce the level of free radical reactions at the tissue level, thus retarding the build up of lipofuscin[9], although it is not yet clear whether antioxidants alter the rate of aging in mice.

1 J.F. Munnell and R. Getty, J. Geront. *23*, 154 (1968).
2 W. Reichel, J. Geront. *23*, 145 (1968).
3 K.R. Brissee, B. Kaack and P. Klara, in: Neurobiology of Aging, p.463. Ed. J.M. Ordry and K.R. Brizzee. Plenum Press, New York 1975.
4 A. Tappel, B. Fletcher and D. Deamer, J. Geront. *28*, 415 (1973).
5 H.L. Newmark, W. Pool, J.D. Baurenfeind and E. Deritter, Pharm. Sci. *64*, 655 (1975).
6 H.E. Gallo-Torres, in: Vitamin E: A Comprehensive Treatise, p.193. Ed. L.J. Machlin. Marcel Dekker, New York 1980.
7 J. Green, in: The Vitamins, 2nd edn, vol.5, p.259. Ed. W.H. Sebrell and R.S. Harris. Academic Press, New York 1972.
8 P. Gordon, in: Theoretical Aspects of Aging, p.61. Ed. M. Rockstein, M.L. Sussman and J. Chesky. Academic Press, New York 1974.
9 D. Harman, Age *3*, 100 (1980).

Fever and survival in the rat. Metabolic versus temperature response

M. Banet[1]

Physiologisches Institut der Universität, Deutschhausstr. 2, D-3550 Marburg (Federal Republic of Germany), 6 October 1980

Summary. In infected rats, survival was directly proportional to the metabolic cost of fever ($\Delta O_2\%/\Delta$ °C) during its rising phase and inversely proportional to the height of fever. It is suggested that some febrile, metabolic response may be beneficial, while the rise in temperature may be harmful for the host animal.

Although fever is the most common symptom of illness, its role in infectious disease in mammals is not clear. In rats infected with *Salmonella enteritidis*, cooling the spinal cord enhances the febrile, metabolic response to cold without affecting body temperature and increases survival[2]. On the other hand, cooling the preoptic area raises the febrile temperature without essentially increasing metabolic rate and decreases survival[3]. It may then be speculated that in salmonellosis some febrile, metabolic response to cold increases survival, whereas the increase in temperature decreases it. The present work, in which the natural course of this illness was studied, shows that this hypothesis is tenable.

Materials and methods. The results were obtained in 156 specific pathogen free, male Wistar rats, weighing about 350 g and kept at 23 °C with natural illumination and food and water freely available. The animals were anesthetized with pentobarbital and fixed to an antirotatory device that otherwise allowed freedom of movement[3]. At this time, either a preoptic[4] or a spinal[5] thermode was implanted in 70 of the animals but, except for the implantation, these animals were handled like the ones without thermodes.

2-3 weeks after the operation, the animals were i.p. infected with 1 ml of a suspension of live *S. enteritidis*[3]. Body temperature was measured at least once a day (at 12.00 h) with a thermocouple inserted about 60 mm beyond the