TECHNICAL PAPERS

Calcium and Growth in Aging and Cancer[1]

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This paper is an attempt to bring together data on changes in intracellular calcium as an important factor in both aging and malignancy. A calcium increase with age has been demonstrated in a wide variety of organisms by many investigators, some of whom are Novi (*20*), Cahane (*5*), Burger and Schlomka (*4*), Simms and Stolman (*22*), Lansing (*12, 13*), and Blumenthal, Lansing, and Wheeler (*2*).

Between the cells calcium is increased in some locations and even decreased in others. It is with the cells themselves that we are concerned, both in aging and in cancer production. Lansing (*12, 13*) noted a marked increase with age in the calcium content of the cell periphery or cortex. This was demonstrated in a plant, a rotifer, a planarian, and a toad. At that time it was suggested that an increase with age in the calcium content of the cell cortex might produce a decrease in permeability of the cell membrane. It was further suggested that a permeability decrease might result in an accumulation of metabolic waste products in the cell which could, in turn, produce the changes associated with senescence.

This view is not without support. Benedict (*1*), as a result of his permeability studies in plants, suggested that aging is a result of decreased cellular permeability. Molisch (*17*) believed that there might be a relation between a calcium increase and a permeability decrease with age in plants. That senescence may be a consequence of progressive accumulation in the cell of materials which either are toxic or obstruct metabolism has been frequently suggested (Jickeli, *10*; Montgomery, *18*; Child, *6*; Seifriz, *21*; and Heilbrunn, *9*).

A series of experiments were conducted by the writer to determine the effect of experimental alteration of the calcium content of cells on longevity (*14*). It was found that, under carefully standardized environmental, nutritive, and genetic conditions, rotifers in a low calcium medium live longer than control animals. It was further shown that experimental removal of calcium from cells of the rotifer by sodium citrate results in a marked increase in longevity. The ability of sodium citrate to remove calcium from the cell cortex was demonstrated by microincineration and electron microscopy (*16*).

The mechanism of aging as set forth is obviously incomplete in that a serious gap exists in the postulated chain of events. Why does calcium increase with age in the cell cortex? This discussion of experiments on longevity and chemical studies on cancer may make it possible to relate the calcium change with age to a mechanism governing growth cessation of cells.

The results of a wide variety of experiments on the effect of starvation on longevity are in essential agreement that starvation increases longevity by extension of the period of growth. Northrop (*19*), in his experiments on *Drosophila*, showed that semistarvation resulted in increased total duration of life. Increased longevity was due to a prolongation of the period of growth; length of the adult period of life was unaffected. Similarly, Kopec (*11*), working with *Lymantria*, *Drosophila*, and tadpoles concluded that prolongation of total duration of life is effected by an extension of the period of growth and that the adult period is not increased. Comparable results with rats have been obtained by McCay and his associates in an extensive series of investigations (*8*). It would appear from these and similar experiments that the length of the period of growth may condition longevity and that cessation of growth may set off the aging mechanism.

Recent experiments by Lansing (*15*) and current unpublished data indicate that cessation of growth is, in fact, a critical turning point in initiation of age changes. Use was made of standardized nutritive and environmental conditions and genetically homozygous rotifers with parental age as the sole variable. Longevity of successive generations of rotifers was traced, parental age being uniformly maintained and lines of young and old parentage contrasted. Lines of old parentage showed progressive decline in mean life span, and all became extinct after a limited number of generations. However, lines of young parentage maintained and increased mean life spans.

Significant in the present discussion is the observation that there is a sharply defined transition between ages of lines that maintain and increase longevity or progressively reduce it. Lines of a parental age younger than that at which growth ceases show progressively increased longevity and are evidently free of any age change. *All* lines of parental age older than that at which growth ceases show progressive reduction of mean life spans to the point of nonviability. The conclusion is warranted that the process of aging is a consequence of changes which occur with cessation of growth.

If cessation of growth is a significant factor in aging and if a mechanism involving calcium increase is an integral part of this system, one would expect to find the very opposite situation in cancer. Cancer may be regarded as a very youthful and vigorous tissue with tremendous and uncontrolled growth capacity.

It has been shown repeatedly that cancer tissue is markedly low in calcium. Recently, Suntzeff and Carruthers (*23*) have shown that there is a 50 per cent reduction of total calcium in methylcholanthrene-induced epidermal hyperplasia from that of normal epidermis, and in squamous cell carcinoma a still further reduction of approximately 60 per cent from the precancerous hyperplastic epidermis. Brunschwig, Dunham, and Nichols (*3*) also discovered a sharp drop of calcium in cancer tissue.

In an extension of the observations of Suntzeff and Carruthers, Lansing, Rosenthal, and Au (to be published) studied the nature of the calcium drops in hyperplastic mouse epidermis and resulting carcinomas by means of ultrafiltration.

[1] Supported in part by grants from the Josiah Macy, Jr. Foundation.

It was found that the ultrafilterable calcium fraction of a squamous cell carcinoma was sharply reduced on both an absolute and relative basis. The conclusion was reached that the base-binding capacity of an organic fraction which binds calcium is altered in cancer. It is reasonable to expect that this calcium binding complex, which changes in cancer to limit calcium uptake, is located like that demonstrated by Heilbrunn in the cell cortex. The work of Heilbrunn and his associates, covering a number of years, are summarized in his book (*9*).

Further supportive but indirect evidence for the localization of the calcium change in cancer in the cell surface can be found in the work of Coman (*7*), who has found that normal cells are more readily separated by micromanipulation when in calcium-free medium than when in balanced salt solution. Further, he has demonstrated that cancer cells are more readily separated than are normal cells, and the suggestion was made that the decreased adhesiveness of cancer cells results from a local calcium deficiency which facilitates separation of these cells from one another.

Thus, the evidence presented, while undoubtedly weak, makes it possible to correlate the various changes described in a single hypothesis. It seems quite likely that an organic calcium-binding complex of the cell cortex plays an integral part in the growth regulatory mechanism of cells and that at the time of cessation of growth this calcium-binding complex, presumably a protein, is altered or reoriented in such a way as to increase calcium-binding capacity. Thus, a mechanism is offered which at least in a general way synchronizes the changes that occur with age. On the other hand, when the calcium-binding system alters in such a manner as to decrease calcium binding, growth does not cease, age changes do not occur, and, in effect, the state of affairs exists that is associated with cancer. This hypothesis is also compatible with Coman's concept of the invasiveness of cancer.

References

1. Benedict, H. M. *Cornell agric. exp. Sta. Mem.*, 1915, **7**, 273.
2. Blumenthal, H., Lansing, A. I., and Wheeler, P. *Amer. J. Path.*, 1944, **20**, 665.
3. Brunschwig, L., Dunham, L., and Nichols, S. *Cancer Res.*, 1946, **6**, 233.
4. Burger, M., and Schlomka, G. *Z. exp. Med.*, 1927, **55**, 287.
5. Cahane, M. *C. R. Soc. Biol.*, 1927, **96**, 1168.
6. Child, C. M. *Senescence and rejuvenescence.* Chicago: Univ. Chicago Press, 1915.
7. Coman, D. R. *Science*, 1947, **105**, 347.
8. Cowdry, E. V. *Problems of ageing.* Baltimore: Williams & Wilkins, 1942. Chap. 26.
9. Heilbrunn, L. V. *An outline of general physiology.* Philadelphia: W. B. Saunders, 1943.
10. Jickeli, C. F. *Die Unvollkommenheit des Stoffwechsels.* Berlin: Friedländer, 1902.
11. Kopec, S. *Brit. J. exp. Biol.*, 1928, **5**, 204.
12. Lansing, A. I. *Biol. Bull.*, 1942, **82**, 385.
13. Lansing, A. I. *Biol. Bull.*, 1942, **82**, 392.
14. Lansing, A. I. *J. exp. Zool.*, 1942, **91**, 195.
15. Lansing, A. I. *J. Gerontol.*, 1947, **2**, July.
16. Lansing, A. I., and Scott, G. H. *Anat. Rec.*, 1942, **84**, 91.
17. Molisch, H. *The longevity of plants.* (Engl. trans.) Lancaster, Pa.: Science Press, 1938.
18. Montgomery, T. H. *Trans. Tex. Acad. Sci.*, 1906, **9**, 75.
19. Northrop, J. H. *J. biol. Chem.*, 1917, **32**, 123.
20. Novi, I. *Arch. Ital. Biol.*, 1913, **58**, 333.
21. Seifriz, W. *Protoplasm.* New York: McGraw-Hill, 1936.
22. Simms, H. S., and Stolman, A. *Science*, 1937, **86**, 269.
23. Suntzeff, V., and Carruthers, Christopher. *Cancer Res.*, 1943, **3**, 431; *J. biol. Chem.*, 1944, **153**, 521.

Coproantibody Excretion During Enteric Infections[1]

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The immune mechanism responsible for initiation of recovery from infections confined essentially to the lumen and mucosa of the intestinal tract, viz., cholera, bacillary dysentery, and certain *Salmonella* infections, is suggested by the recent work of Burrows and co-workers (*1*), who showed that antibody is excreted in the feces by both infected and immunized animals and immunized human volunteers, and that immunity to experimental infection is associated with its presence. They showed that the appearance of coproantibody precedes that of serum antibody, reaches peak titer and declines while serum antibody is still rising, and completely disappears despite the persistence of serum antibody. They suggested that the independent behavior of serum and fecal antibody might be explained on the assumption that the latter represents intracellular antibody, possibly transported to the lumen of the bowel by lymphocytes. However this may be, the late appearance and persistence of serum antibody strictly limits its diagnostic utility, but the behavior of coproantibody suggests that it might be useful both in the rapid diagnosis of specific acute enteric infection and in providing a clue to the possible etiology of chronic diarrheal disease such as chronic ulcerative colitis.

TABLE 1

Coproantibody Excretion From Patients Suffering From a Variety of Enteric Infections

Clinical diagnosis	No. cases	Pathogens isolated	Copro-antibody present
Acute bacillary dysentery	20	*S. sonnei* from 6; *S. flexneri* from 5; none from 9	20*
Chronic bacillary dysentery	18	*S. sonnei* from 3; *S. flexneri* from 5; none from 10	18†
Acute diarrhea	14	Various *Salmonella* from 10; none from 4	14†
Chronic diarrhea	35	Para A from 9; Para B from 3; none from 23	32‡
Chronic ulcerative colitis	5	*S. flexneri* from 1; *S. sonnei* from 2; none from 2	5§

* Eleven, 1:640 or above; 8, 1:320; 1, 1:160 (maximum titers obtained).

† Coproantibody titer of 1:160 or above obtained against homologous organism, or against one or more species of enteric pathogens, on several occasions from each patient.

‡ Three failed to show coproantibody at any time during the observation period.

§ Usually obtained in highest titer during episode of exacerbation of disease.

[1] Aided in part by a grant from the M. D. Anderson Foundation of Houston.

[2] The authors are indebted to William Burrows for making available copies of translations from the Russian (*4*) furnished him by the Translation Unit of the U. S. Public Health Service.