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SOME THERAPEUTIC IMPLICATIONS OF THE CROSSLINKAGE THEORY OF AGING

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INTRODUCTION

The concept of crosslinkages as the base for aging was suggested by Bjorksten (1941), who in 1942 expressed this theory as follows: "The aging of living organisms I believe is due to the occasional formation by tanning, of bridges between protein molecules, which cannot be broken by the cell enzymes. Such irreparable tanning may be caused by tanning agents foreign to the organism or formed by unusual biological side reactions, or it may be due to the formation of a tanning bridge in some particular position in the protein molecule. In either event, the result is that cumulative tanning of body proteins which we know as old age." If we add the words "and nucleic acids" to "proteins" this still covers the tenets of the crosslinking theory.

Bjorksten (1951) summarized the already then substantial literature on crosslinkages, predicting that their significance in processes of life--and death--would soon become appreciated.

In 1948 Haddow, Kon and Ross noted that substances having more than one alkylating group in the molecule--thus capable of forming crosslinkages--have a dramatically greater cytotoxic action than those having only one such group. Goldacre, Loveless and Ross (1949) proposed that the cytotoxic activity was due to their ability to crosslink. In 1955-1956 F. Verzár presented the same theory independently, in the context of collagen only, and D. Harman (1956) stressed the significance of free radicals, which Charlesby (1953) had proved to be powerful crosslinking agents. In 1957 Zinsser et al. showed the correlation between aging and the build-up of polyvalent metals as potential crosslinkers in aging aorta. Goldacre

et al. (1949) showed that chromosome abnormalities caused by alkylating agents are due to formation of covalent crosslinkages, and Alexander and Lett (1960) showed that crosslinking of DNA occurs in contact with bifunctional nitrogen mustards, an observation later extended to include a majority of carcinogenic compounds. That such reactions would be caused by all crosslinking agents (referred to in the technical and the older literature as "tanning agents") on the genetic matter is obvious in view of the presence in the latter of numerous reactive groups. The alkylating agents were conspicuous biologically only because of their ability to penetrate cell walls. On the other hand, for example, an aldehyde or peroxide formed by a reaction within the cell could be fully as effective as a crosslinker covalently changing DNA or RNA molecules (Bjorksten, 1963, p. 181; Bjorksten & Andrews, 1964, pp. 630-631).

The status of these developments was summarized by Bjorksten in 1962, and again in 1968 and 1971, with fairly comprehensive bibliographies. In 1965 Carpenter published his diffusion theory, based on crosslinkage concepts. It was apparent that the effect of crosslinking coincides with changes taking place in aging, and that no other single reaction known can on a qualitative basis equal its potential destructiveness to macromolecular systems.

More recently Piez, LaBella, Gross, Schmitt, Bensusan, Veis, Gallup, von Hahn and many others have added to our knowledge of the precise nature of the crosslinkages that occur, particularly in collagen. Since all proteins have basically related structures and similar reactive groups, their work has a significance extending much more broadly than to collagen specifically.

The significance of crosslinkage in proteins is its generality and ubiquity. It has long been known that extensively crosslinked proteins become resistant to enzymes (Thomas and Seymour-Jones, 1934; Gustavson, 1942; Lipsitz et al., 1949; Bjorksten and Gottlieb, 1954; Kohn and Rollerson, 1960). Extensively crosslinked protein cannot be broken down biologically. Some protein bound tritium administered at birth was found in rats 809 days after administration (Bjorksten and Ashman, 1970; Bjorksten et al., 1971). One mechanism which would prevent the breakdown by any possible enzyme would be the formation of aggregates of such density that steric hindrance prevents access of any large molecule. Crosslinking is the most obvious and prevalent cause of such steric hindrance.

The significance of crosslinkage in genetic substances as related to aging has been obvious since the work of Haddow, Goldacre and Alexander. That it is a large factor is conclusively proven by Hart and Setlow (1974) who have shown a clear correlation between the specific life span of a species, and its ability to rapidly repair induced damage to genetic molecules. It remains a moot

question which is more detrimental: whether it is the DNA crosslinkage or the more generalized protein-to-any-reactive-large-molecule crosslinkage, as discussed in 1964 (Bjorksten). For every DNA molecule there is a myriad of protein molecules. The DNA molecules are irreplaceable--but if enough of the other molecules accumulate in insoluble aggregates they will preempt space and disrupt transportation so that the damage may be as serious. In both cases, the bottleneck for possible repairs is the steric accessibility for repair of the aggregates. No repair can be made if access of the repair enzymes to the damage site is sterically barred. And the aggregates formed in old animals, which perinatally received radiotracer contain not only peptides, but also nucleotides, pointing to such steric hindrance by repetitive random crosslinkage as the identical limiting factor for repair and scavenging of both genetic and nongenetic immobilized material (Bjorksten et al., 1971).

The repair of damaged genetic molecules, particularly DNA, studied by Howard-Flanders and Boyce (1966), has received a great deal of attention in recent years. Hart et al. (1976) cites 87 references relevant to this work. The establishment by Hart and Setlow (1974) that a good correlation exists between the speed of DNA repair and the life span of various species of mammals confirms strongly the significance of DNA repair speed as an important factor in aging. The postulated repair mechanisms are enzymic. However, any repair of DNA is impossible when the access of repair enzymes is prevented by extensive crosslinking. The aggregate must first be broken down to the extent necessary to permit access of repair enzymes to the DNA. In this manner, and to this extent, crosslinkage is and remains a key mechanism to aging (Bjorksten, 1955), regardless of which factor in the end proves to dominate: the scarcer but more damaging crosslinkage of genetic molecules, or the individually less damaging, but much more frequent, crosslinkage of nongenetic molecules.

COULD VITALITY BE PRESERVED IN A MAJOR WAY?

The lack of progress in countering the destructive impact of the years, which now steadily reduces our ability to maintain health, is apparent from Fig. 1.

In other fields breakthroughs have resulted when enough well directed massive effort was applied.

It should be possible to bend the curve in Fig. 1 into a breakthrough similar to those shown in Fig. 2. It seems a stain on the administration of our science that life expectancy at 60 is still within 2 years of where it was 184 years ago. Where should

Fig. 1. The life expectancy at age 60 for 1789-1973. (From data of Historical Statistics of the Metropolitan Life Insurance Company, and from Vital Statistics of the United States, 1973).

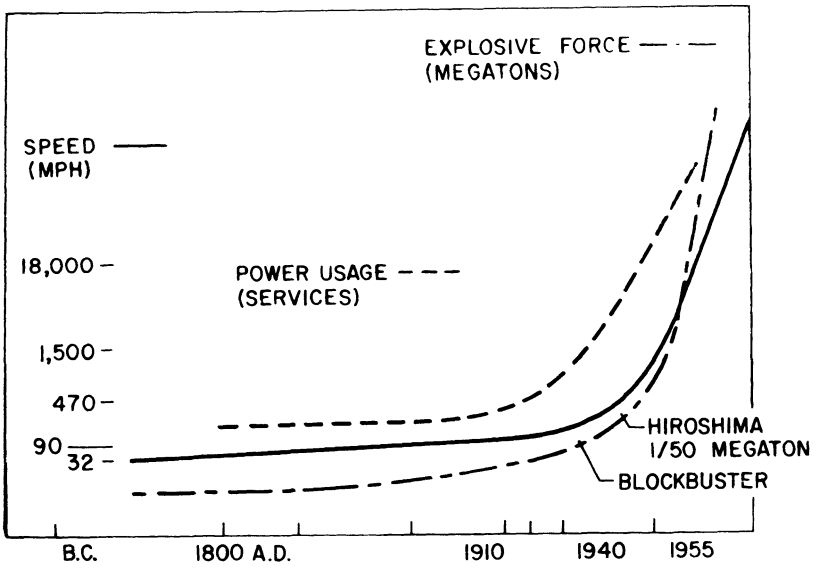

Fig. 2. Other important problems, properly researched and supported, have decisively broken away from past inhibitions. Why not aging? After Powell, J. L., The Chemist 35, 351 (1958).

our efforts be centered? What guidelines do we have? First of all, we need full recognition of the primary cause of aging so that our efforts can be directed by logic as contrasted with convenience or mere chance!

RANDOM CROSSLINKAGE A PRIMARY CAUSE OF AGING

Many theories have been advanced to explain the loss of strength and vitality (Bjorksten, 1969). Of these, only the crosslinkage theory is primary and has answered all objections and met all criteria (Bjorksten, 1971, pp. 33-36). The crosslinkage theory has now withstood the test of time for 35 years; it has been rediscovered independently at least 4 times; there has been enough testing and evaluating. Since it is basic, I shall again briefly restate the way random crosslinking can initiate aging on a molecular scale. The following illustration (Figures 3, 4, 5, 10) and explanations are reproduced with permission from my article in THEORETICAL ASPECTS OF AGING (ed. by M. Rockstein, Academic Press, Inc., 1974).

Figs. 3, 4 and 5 show schematically what crosslinkages between strands can do to DNA (Bjorksten, 1974).

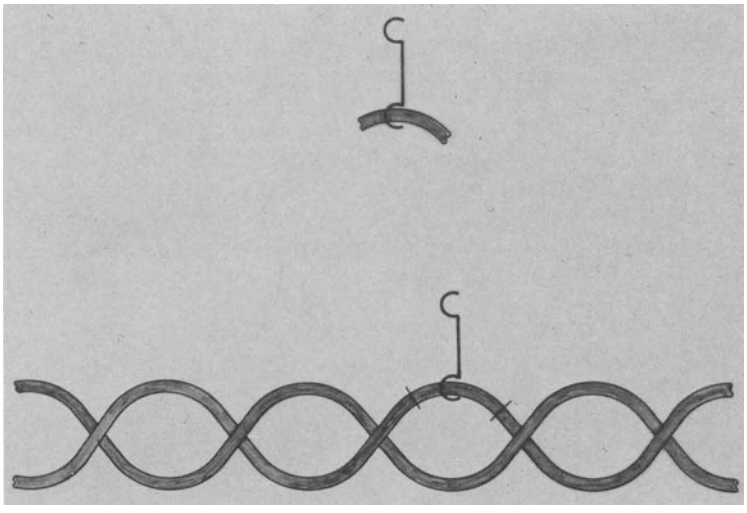

Fig. 3. A crosslinking agent attaches itself at one point of a DNA molecule, involving one strand only. Right, the agent has been excised by defense mechanisms together with a piece of the DNA affected. The damage is then repaired, the unaffected strand being the template.

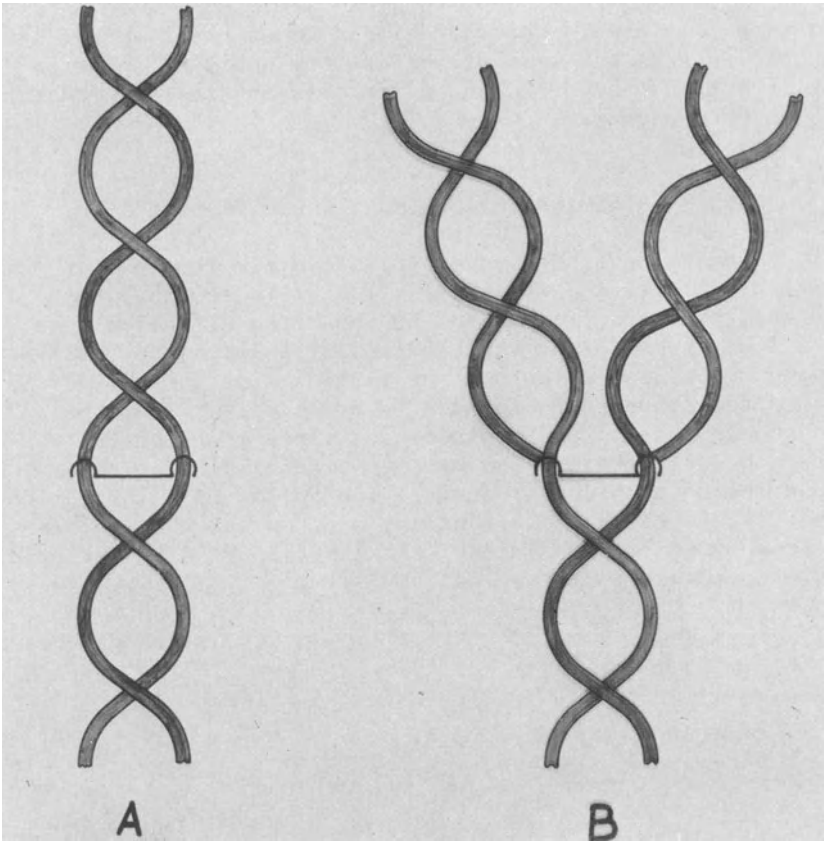

Fig. 4. In A, the crosslinking agent has become attached to the second strand of DNA before the defense mechanism could excise it. When this has happened, the cell is doomed. If the crosslinker is excised, there will be no template for repair as both strands are involved at the same point. If the crosslinker remains as shown in B, it will block the normal parting of strands in mitosis at a stage where the resultant DNA can neither return to normal nor complete the division.

More complex, but nonetheless often fatal damage is done when the crosslinker already attached to one strand of DNA reacts with another different macromolecule, such as a histone (von Hahn, 1964), an RNA, an enzyme, peptide or any other large molecule which has reactive groups (Bjorksten et al., 1971; Acharya et al., 1972).

In spite of the key role which DNA plays in biology, and the proven sensitivity of DNA to crosslinkage, damage to DNA may not be the most important factor in aging. Virtually every large molecule which has more than a purely structural function is susceptible to crosslinkage with any other similarly susceptible molecule. In a lifetime billions of crosslinkages will thus unavoidably be formed. Most of these can be reversed, but some of them cannot. These latter will accumulate over the years. The resultant aggregates are composed of proteins, nucleotides, polymeric fats, polysacharides, any available large molecule at all which can react with any crosslinking agent at all, or which can be directly interlocked, will form parts in the resultant aggregates, as illustrated in Fig. 5.

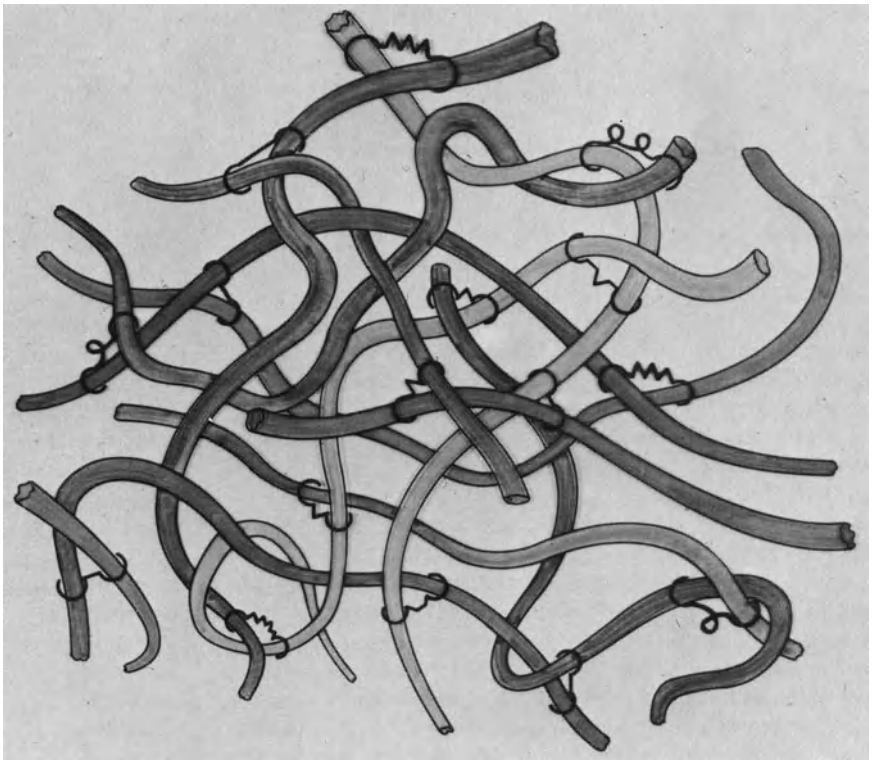

Fig. 5. Random, accidental, mostly nonenzymatic crosslinkages accumulate over a lifetime to form dense aggregates of any available large molecules with any crosslinking agent.

Such essentially nongenetic agglomerates will be whittled away from the surface by proteolytic and other catabolic enzymes, but when the crosslinkages have reached a certain density, the aggregates will become sterically inaccessible to the enzymes available to human metabolism. Because of their much greater quantity, these aggregates may play as large a part in the aging syndrome as does crosslinking of DNA. Tanzer (1973) suggests that crosslinking may prove the "Rosetta Stone of Aging." I could not agree more, having expressed the same conclusion 18 years earlier (Bjorksten, 1955).

The principal group of enzymes which is preferentially increased with age, is that of enzymes which have preponderantly catabolic function, in other words, those most adapted to break down these aggregates (Bjorksten, 1966). It would be very interesting to know if the increase of the activity of these enzymes with age correlates with the life-span in various species in the same way as the speed of repair of DNA studied by Hart and Setlow (1974).

CROSSLINKAGES AS A COMMON BASE FOR DEGENERATIVE DISEASE SYNDROMES

A few major instances will be briefly discussed.

Athero-Arteriosclerosis

It has been fully established (Fishman et al., 1975; Freidman et al., 1975; Constantinides and Wiggers, 1973) that the endothelium becomes permeable to blood serum before infiltration or deposits take place. It has also been established that elasticity of the arteries diminishes with progressive years. Loss of elasticity and embrittlement are well known primary effects of crosslinkage (Bjorksten, 1951, pp. 343, 349).

When loss of elasticity has progressed so far that the endothelium no longer can follow the pulsations, it may rupture at the point of highest hydrostatic pressure, usually at a bend or branching point of the artery, or the media hardens to the point that the intima no longer can stand the squeeze repeated every second between noncompressible liquid and hardening media, or the continuity of cellular adhesion in the endothelium is damaged and gives rise to leaks. Whichever takes place first, the effect is the same: penetration of the endothelium and subintimal deposition of suspended substance. If this is easily resorbable, the lesion may heal, but if it consists of difficult-to-remove particles, for example, of cholesterol or triglycerides, it cannot be resorbed. Such a sequence is shown in Figs. 6, 7, 8, which are copied with permission from a recent article (Bjorksten, 1976).

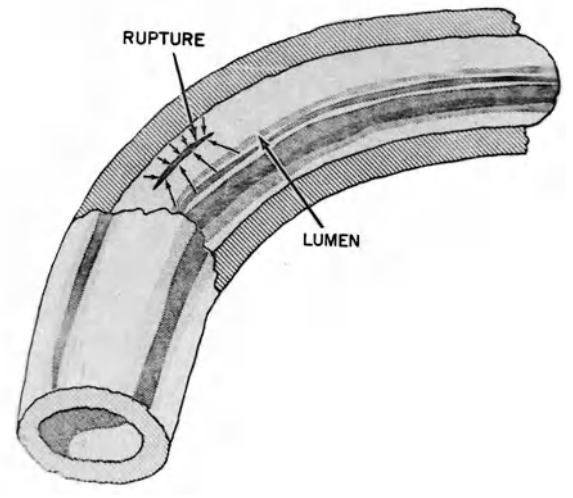

Fig. 6. A rupture has formed in the intima; blood serum filters through and deposits particulate matter in the subintimal region.

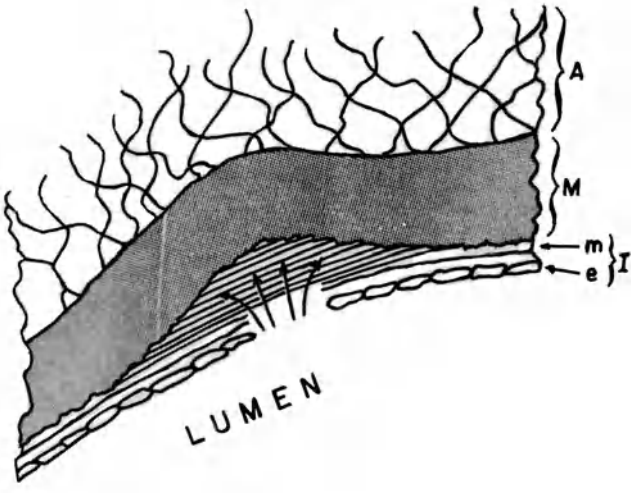

Fig. 7. Diagram of the rupture in section. A = adventitia, M = media, m = membrane, e = endothelium, i = intima

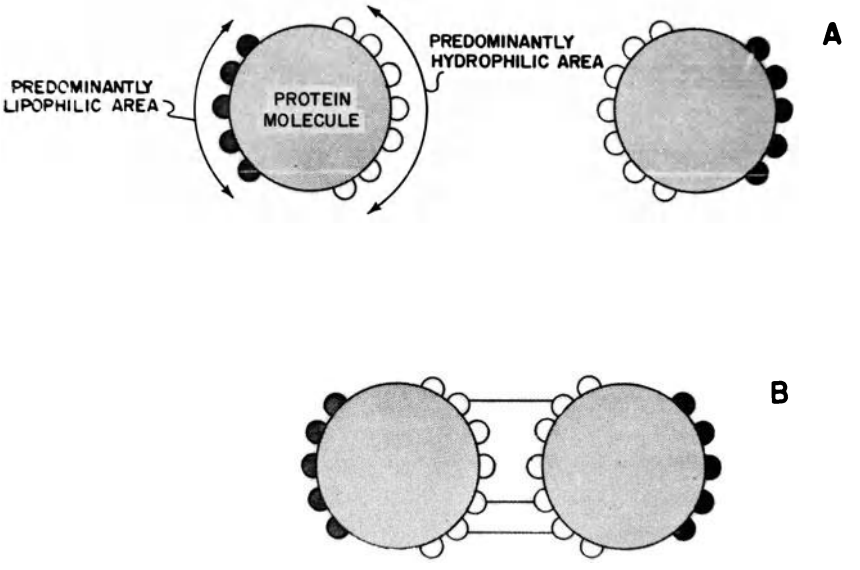

Fig. 8. A. Two protein molecules having both lipophilic (dark) and hydrophilic (white) sites. B. The same molecules cross-linked preferentially at the hydrophilic sites, so as to turn more of the lipophilic sites outward on the average, thus becoming lipophilic.

The affinity to tissue proteins for lipids increases with progressive crosslinkage, for reasons shown in Fig. 8.

On the average, the hydrophilic areas of a macromolecule are more susceptible to crosslinkage than the lipophilic areas, because more crosslinking agents are water soluble, and also because many of the reactions occur more easily in the aqueous phase. Since a preponderance of crosslinkages in both participating macromolecules will be apt to involve their more hydrophilic areas, these will tend to become oriented towards each other, leaving the preponderantly lipophilic areas preferentially exposed. The result is the increasing lipophilicity with progressive crosslinkage phenomena well known and utilized both in the leather manufacture, and in graphic arts to prepare preferentially oil ink receptive printing plates.

This increased lipophilicity of the proteins generally will favor fat deposit on the soft tissues with progressing age.

It may also affect the immunological recognition of the molecules, and could thus well be a factor in autoimmunity (Raff, 1976).

These last mentioned effects of crosslinking may explain the frequent occurrence above age 70 of extensive arterio-atherosclerosis where lipidemia is conspicuously absent. Blumenthal (1975) states in a penetrating article: "There are many curves representing functional declines with age, or changes in the incidence of disease states, which show a middle age peak followed by a decline, and for many of these there is no evident causal link with lipid metabolism; some cancers represent a case in point. All may have a common fundamental underlying cause, not yet elucidated. In any event, these observations raise doubt as to how essential hyperlipidemia is in the genesis of arterio-atherosclerosis, when so many individuals with advanced arterio-atherosclerosis after age 70 do not manifest this biochemical abnormality." Goldstein et al. (1973) found in a study of 500 survivors of myocardial infarct that only 25% of the women and none of the men above 70 showed elevated lipid levels.

The increase in lipophilicity on aging, may explain this. It can be demonstrated easily and even simulated in vitro in gelatin treated with crosslinking agents. Cholesterol adsorption has been induced in vivo by uranium salts, also effective crosslinking agents (Campbell and Longebaugh, 1950). Protein rendered lipophilic by blockage of a sufficient part of its hydrophilic sites, as illustrated in Fig. 8, will absorb lipids even when these are present in low concentrations. Cholesterol and triglycerides will then remain fixed more tenaciously than lipids of a lower melting point or having additional reactive sites.

A crosslinking agent, attached with one end to any surface is capable of capturing with the other end a lipoprotein molecule where the lipid is cholesterol, regardless of the nature of the surface to which it is attached. If the crosslinking agent is strongly ionic, it is even capable of capturing cholesterol connected with nonproteinic colloidal carriers (Bjorksten, 1952); Bjorksten and Gottlieb, 1954). Cholesterol and probably also triglycerides so captured will then remain fixed more tenaciously than lipids of a lower melting point or having additional reactive sites.

Other Clinical Manifestations of Crosslinking

1. Senile Cataract.

In a recent paper, Bellows and Bellows (1976) conclude that crosslinking is the common denominator in past theories of

senile cataracts. Due to its transparency, the lens is an ideal organ for the systematic observation of aging phenomena, and the hardening process can be analyzed in convincing detail. This has been done, and the conclusions stated.

2. Diabetic Sclerosis.

Diabetes is characterized by the inability of the organism to process carbohydrates in the normal pathways. Instead, alternate, and less efficient pathways are taken, with the result that incomplete oxidation products are favored. Some of these are known crosslinking agents.

The shifting of pathways for energy generation which takes place in diabetes is schematically shown in Fig. 9.

Basically, glucose is the optimal biological energy source, because it is the only carbohydrate which can be utilized directly by all tissues. The brain cannot utilize fatty acids or fats under any circumstances, but it can as a poor second choice to glucose, utilize ketone bodies such as pyruvic acid or gamma hydroxy butyric acid.

The muscles can utilize fats or fatty acids for energy production. Therefore, when the glucose available is short of abundance, the brain gets first call on the glucose. This is the evolutionary reason why fats cause an inhibition of insulin function and/or formation. It is the means for forcing the muscles into a fat combustion metabolism in order to secure that the brain gets all of the scarce glucose (Miettinen, 1969). At the same time, protein-amino acid catabolism generates pyruvic acid, which in turn goes to fatty acid to increase the supply of this for the muscles, and in the process also steps up the cholesterol production. This is the reason why cholesterol and fatty acids in the blood increase in diabetes.

It is thus apparent that in diabetes not only the formation of very reactive pyruvic acid is stimulated. Other keto-bodies are formed, not all of which have been defined. Fatty acids and cholesterol have a well known positive correlation to atherosclerosis. The oxidation products of the fats comprise both aldehydes and peroxides, of which many are particularly active crosslinkers (Gustavson, 1956; Bjorksten, 1963; Milch, 1964).

The diabetic metabolism is thus particularly unfavorable from the standpoint of the crosslinking theory of aging. It appears desirable to avoid inhibiting insulin function with diets containing substantial and avoidable quantities of any fats. Several investigations have substantiated the practical soundness

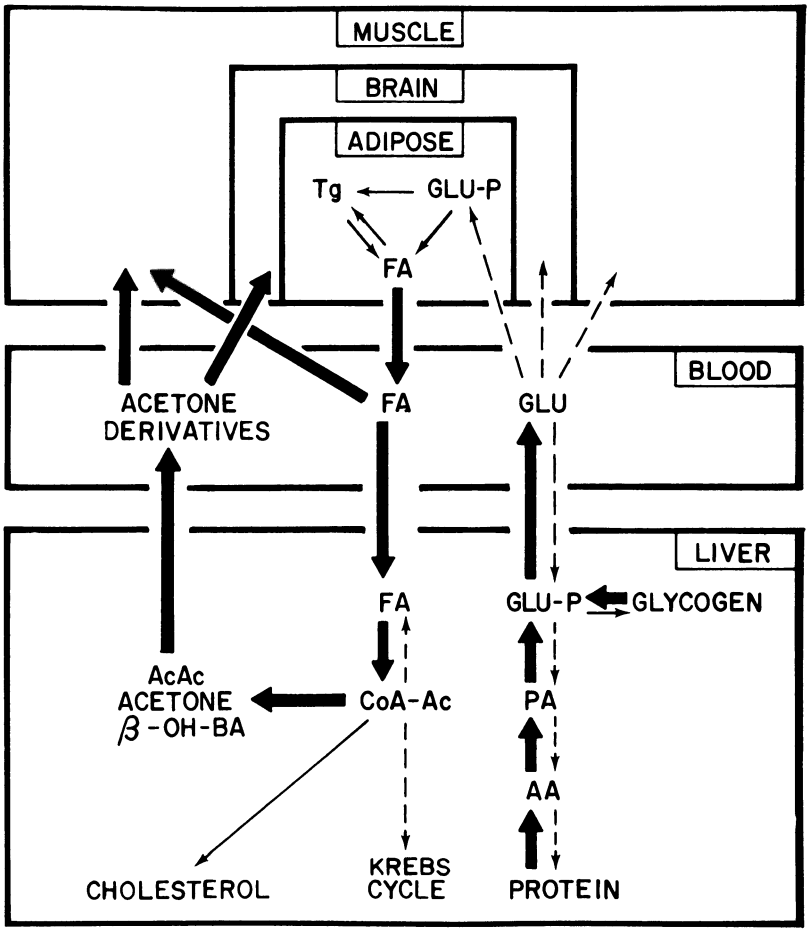

Fig. 9. (After T. Miettinen, Medical Diabetes Symposium, 1969, p. 39.) Diagram showing diabetic metabolism. The metabolic pathways which are restricted in diabetes are shown as dotted arrows; pathways increased in diabetes with heavy arrows. AA = Amino acids; AcAc = Acetoacetic acid; β-OH-BA = β Oxybutyric acid; CoA-Ac = Coenzyme A acetate; FA = Fatty acids; Glu = Glucose; Glu-P = Glucose phosphate; PA = Pyruvic acid; Tg = Triglycerides.

of this general conclusion (Rabinowich, 1935; Buber, 1968; Bierman et al., 1971; Brunzell et al., 1971; Anderson et al., 1973; Weinsier et al., 1974). After a review of the relevant literature, Pritikin (1974) concludes that more than ten % of fat in the diet is disadvantageous. Unquestionably, excessive fat in the diet will force even a normal metabolism part way into diabetic pathways by reducing insulin effects, thus shifting muscle metabolism from carbohydrates to fats as an energy source with consequent increase in the formation of deleterious by-products.

Slow release of glucose is favored by the use in the diet of its polymeric forms, as, in particular, boiled starch, dextrins. Heinonen (1969) states it is well known that persons who eat many small meals show less sclerosis than those who take few large meals. This too avoids release of nutrients at a faster rate than metabolism can handle smoothly, without build-up of reactive intermediate products.

3. Spontaneous Fractures.

Progressive loss of elasticity is a well known effect of extensive crosslinking. Crosslinking on a molecular scale may be likened to the crossbracing which a welder or a carpenter resorts to when he wishes to make a structure very rigid and nonyielding. The result is the same. A bone or a blood vessel or any other tissue is thus rigidized by extensive crosslinkage, any mechanical energy input is dangerously concentrated at the point where it occurs instead of being divided and absorbed over a much larger area as it would be if the resilience of youthful tissue were still present.

The loss of elasticity of the collagenous substance in the bones thus facilitates breakage. The loss of elasticity in arteries has already been discussed. Corresponding phenomena may increase the susceptibility to microfractures in glandular or intestinal organs, where they could become the starting point for continual irritations or ulcerations, and perhaps even malignancies.

4. Cancer

The material at hand is too complex to permit any conclusions at this time. However, a few points may be worth considering:

a. DNA anomalies can be caused by crosslinkages, which could even be their major cause. Such anomalies have been associated with cancer (Failla, 1958; Alexander and Lett, 1960).

b. Almost all of the known carcinogens either are themselves potential crosslinking agents, or have metabolites which are (Alexander et al., 1952, 1959).

c. There is a high correlation between cytotoxicity and crosslinking capacity (Haddow et al., 1948).

d. There is a high correlation between incidence of cancer and exposure to known crosslinking influences, including both ionizing radiation (Upton, 1960) and chemical carcinogens, including "natural" carcinogens such as the aflatoxins. Such relationships are increasingly referred to (for example, Conney, 1973; Nietert et al., 1974; Zeldin et al., 1975).

5. Immunological impairment.

There can be no question at all of the significance of the immunological effects in aging and at least in part in the formation of crosslinked deposits, as in amyloidoses. The primary effect of a considerable part of these changes could very well depend on the effect of crosslinking in changing the antigenic properties.

That changes as profound as those occurring in crosslinking must necessarily result in immunologic recognition, cannot be doubted, although the precise quantitation remains to be worked out.

The illustrations in the paper by Raff (1976) show clearly the significance of crosslinkage in the immune system.

Walford (1964) discussed this in a positive sense, and the questions raised by him (1969) have been answered (Bjorksten, 1974).

The present state of knowledge - and lack thereof in some crucial regards - have been aptly summarized by J. D. Stobo and T. B. Tomasi in an editorial in Journal of Chronic Disease (1975). The crosslinkage concept might well prove rewarding as a guideline in further exploration of the gaps pointed out by these authors.

Prevalence of crosslinked aggregates will certainly greatly disturb intracellular transportation, and might, for example, in this manner cause defects in the interaction between helper T and B cells stressed by Price and Makinodan (1973) as well as interfere with regulatory functions, considerably before the syndrome becomes more general.

COUNTERMEASURES

Various steps for minimizing exposure to random crosslinking have been discussed recently (Bjorksten, 1976). These include nutrition planning to avoid sudden overloads or any "bottleneck" in any metabolic path, favoring paths which involve the minimal amount of chemical processing in the body. Sound guidelines, to which I subscribe as far as they go, have been presented by R. Williams (1976).

What can be done along these lines is necessarily limited, and can be expected to effect life extensions only in the approximate range 5 - 15 years. This, however, can be done now and would gain for some of us the time needed to master the more difficult but also potentially far more rewarding problem of reversing damage already done, beyond what can be done by proper dieting and exercising (Pritikin, 1975; Haeger, 1973, 1974; Williams, 1976).

SOLUBILIZING CROSSLINKED AGGREGATES

These considerations apply both to DNA or RNA molecules repaired within amitotic cells such as neurons, and to breakdown and removal of complex crosslinked aggregates which have been formed during a lifetime and have withstood all enzymic attacks the host organism has been able to mount. Yet, there must be in existence enzymes which can cope with these, for otherwise large fossil deposits of such crosslinked proteins would have been found.

Starting on this premise, suggested by Bjorksten (1955), Bjorksten, Weyer and Ashman (1971) isolated insoluble aggregates from old human brains, suspended them in agar and inoculated them with various microbe rich infusions. Figure 10 shows how a few cultures were capable of dissolving the grey suspended matter surrounding them, forming "halos" in the Petri dishes.

Subcultures were made, grown in 14000 ml fermenters. In 4 years of work an enzyme was separated and purified chromatographically, which was considerably superior to any enzyme previously known to us in solubilizing highly crosslinked protein aggregates, whether natural or artificially prepared by treating gelatin with powerful crosslinking agents.

The enzyme in question was derived from a mutant strain of Bacillus cereus. At least some traces of similar enzymes were found in every spore forming organism examined. Their content of such enzymes appears connected with sporulation. Evolution might have developed this class of very low molecular weight enzymes

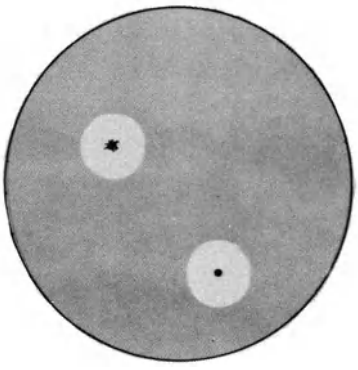

Fig. 10. The dense structures of Fig. 5 were isolated from old human brain, pulverized and dispersed in agar, rendering the agar grey. A suspension of mixed soil organisms was used as inoculum. Two colonies have developed clear halos, showing that these colonies excrete enzymes capable of dissolving the dispersed "gerogenic aggregates.

in order to make possible the dissolution, and perhaps also the synthesis, of the extremely dense and resistant spore shells where access of larger enzyme molecules are sterically hindered. So far only one of these enzymes has been fairly thoroughly studied (Schenk and Bjorksten, 1973). It has only been tested toxicologically, as quantities so far available to us have been insufficient for extensive animal tests. However, enterically coated granules fed to rodents did pass the acidity of the stomach. Some evidence indicates that it has had effects in the bloodstream. It would be almost a miracle if the first member of a family of enzymes should be found to provide the solution to all problems. We have encountered at least 7 promising members of this family of enzymes present in other microorganisms. One of these is now available to researchers from Worthington Biochemical Corporation, Freehold, New Jersey, under the designation "Microprotease, Bac. cereus" (Worthington, 1976).

If these low molecular enzymes are indeed capable of penetrating to organs beyond the circulatory system, they may well provide the key to the long overdue breakaway in the curve of life expectancy at 60, and thus give all of us many additional happy years in good health. Since they appear to destroy most other blood enzymes, their application should probably be intermittent and/or the dosage carefully controlled. The blood enzymes can be

replaced rapidly, the crosslinked aggregates have a vastly slower cycle.

The work along this line has been slowed by the governmental pressures on the pharmaceutical industry, which was supporting our work 1966-1970.

This long term work was one of the first casualties of the reallocation of funds, forced upon the pharmaceutical industry by congressional investigations. Funds allocated for our long range research on aging were among those preempted for defensive research to disprove allegations made. This situation is general and we all have merely begun to pay for it.

In 1961, for example, 39 new single chemical entities appeared--most of these generated by American pharmaceutical industry. In 1974 the number of these was only 18, largely originated abroad, and I see no sign of a reversal.

While hamstringing the most effective mechanism for pharmaceutical advance, the Government has so far been unable to furnish any effective substitute to support innovation in this field.

On September 24, 1883, Oscar Wilde, the famous author, told a distinguished British audience about his extensive travels in America the preceding year. He commented:

"A remarkable characteristic of the American is the manner in which they have applied science to modern life.

"This is apparent in the most cursory stroll through New York. In England an inventor is regarded almost as a crazy man and in too many instances invention ends in disappointment and poverty. In America an inventor is honored, help is forthcoming, and the exercise of ingenuity, the application of science to the work of man, is there the shortest road to wealth."

This happy situation, which made our country great, has been reversed. That work on aging, which if successful, could bring a major breakthrough, has had insufficient support in the USA. Research on the control of aging must be funded and pursued vigorously if those who live now are to benefit.

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