incuced by cysteine, i.e. after the addition of cysteine, the net synthesis of RNA and protein stopped immediaately, whereas a certain quantity of DNA was synthesized. Theoretically, the alterations of cell metabolism before irradiation may enhance the radiation resistance in either of the following two ways: 1. by decreasing the intrinsic sensitivity of cells to damaging effects of irradiation, or 2. by increasing the recovery after irradiation.

The results presented here show that both of the abovementioned possibilities take part in the enhancement of radiation resistance induced by cysteine treatment in *E. coli* B/r.

E. coli B/r was grown on mineral salts-glucose medium[5]. The cells in mid-log phase of growth were treated with cysteine (2 m*M*) and incubated further at 37 °C. After 1 min or 30 min, the cells were centrifuged, washed 3 times, resuspended in iced phosphate buffer (pH 7.0) and irradiated with X-ray. Surviving fractions were determined by plating bacteria on nutrient agar[6], and on the same agar, containing acriflavine (2 µg/ml), a potent inhibitor of DNA repair[7] (Figure). The 1 min cysteine treatment raised the D_{99} (1% survivals) from 20.5 to 30.6 krad and from 9.3 to 13.5 krad for cells plated on nutrient agar and acriflavine containing agar, respectively (the dose response factors of 1.49 and 1.45). Thus, the radiation resistance induced by 1 min cysteine treatment was not affected by acriflavine, i.e. the intrinsic sensitivity of bacteria was decreased by this treatment. The 30 min cysteine treatment, however, resulted in greater resistance enhancement when the cells were plated on nutrient agar than they were plated on the same agar, containing acriflavine, the dose response factors being 2.1 and 1.7, respectively. If our model is a correct interpretation of the mechanism of biochemical radioprotective effect of cysteine, the results presented here suggest that the asynchronous synthesis of macromolecula induced by cysteine treatment enhances the recovery after ionizing irradiation. This conclusion is in agreement with some recent findings[8,9] that the functional *exr*A gene is required for radiation resistance enhancement induced by the abovementioned type of asynchronous synthesis of macromolecula.

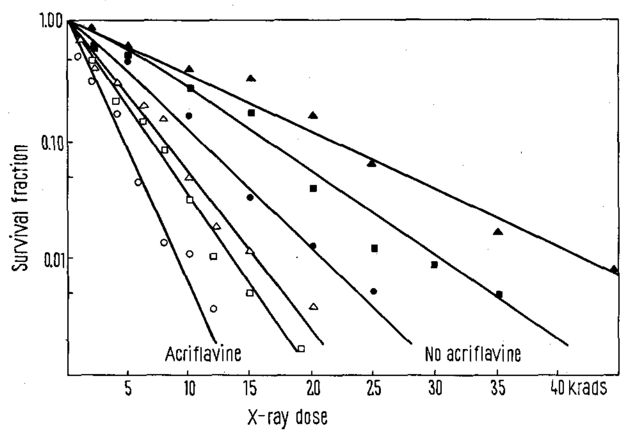

Fig. 1. Effect of acriflavine on the radiation resistance enhancement induced by cyteine treatment in *E. coli* B/r. Surviving bacterial cells were counted by making dilutions in phosphate buffer and plating 0.1 ml samples in triplicate on nutrient agar (closed symbols) or on the same agar containg 2 µg/ml acriflavine (open symbols). The plates were incubated at 37°C before counting. ○ and ●, non-treated cells; □ and ■, cells treated with cysteine for 1 min; △ and ▲, cells treated with cysteine for 30 min.

Zusammenfassung. Zur Prüfung der Strahlenresistenz von *Escherichia coli* nach bewährter Methode wird Acriflavin, ein DNA-Reparaturhemmer, zugegeben und es wird festgestellt, dass die Strahlenresistenz auch unter Cysteinschutz sinkt.

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Changes in the Degree of Orientation of Bone Materials with Age in the Human Femur

In recent years bone has increasingly been considered as a two phase composite of apatite and collagen. Of the two materials apatite has the higher elastic modulii. In a composite of this type the degree of preferred orientation of the high elastic modulii material plays a very important part in determining the physical properties of the composite[1]. A literature survey showed no systematic study of change in the degree of preferred orientation of apatite crystals with age, although the preferred orientation of the apatite crystallites in mature bone and their relationship with collagen fibres had been studied as early as 1936[2]. It was therefore decided to study this aspect of bone.

Material and method. X-ray diffraction diagrams of 20 specimens of femoral bone from 6 day to 76-year-old individuals have been studied. Of these, 6 were below 5 years of age and the rest adults of different ages. X-ray diagrams were taken for both posterior and anterior quadrants of the midshaft of the femoral diaphysis. Specimens for X-ray diagrams were obtained by cleaving wedge-shaped pieces from the middle of the quadrant.

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No.	Age and quadrant.	[a] Ratio of I_{max}/I_{min} of 002 ring	Remarks
1	7 weeks	1	Completely random in both quadrants.
2	1 year (anterior)	2	Small degree of orientation.
3	1 year (posterior)	4	High degree of orientation. Same femur as in No. 2.
4	56 year (anterior)	4	High degree of orientation.
5	56 year (posterior)	5,5	High degree of orientation Same femur as in No. 4.

[a] I_{max} denotes the intensity maximum in the 002 reflection of apatite. I_{min} denotes the intensity minimum in the 002 reflection of apatite.

The diagrams were taken with MoKα radiation on a flat-plate camera, using 4 cm specimen to film distance. The long axis of the bone was approximately vertical and normal to the X-ray beam. The majority of the diagrams showed that 002 relfections of apatite had intensity distribution along the circle. This intensity distribution was measured by a rotating stage photometer. The ratio of maximum to minimum intensity was taken as a measure of the degree of preferred orientation of apatite. In some cases the bone samples were decalcified either with EDTA solution or formic acid solution and the X-ray diagrams retaken to find the orientation of the collagen fibres.

A subsidiary investigation showed that the preferred orientation was cylindrically symmetrical about the long axis of the bone within the limits of observation. It was also observed that any attempt to smooth the specimen surfaces, disturbs the orientation of apatite crystals at the surface.

Results and Discussion. The Table lists a representative set of the results so far obtained. It is obvious from the table that at birth the apatite crystals were randomly orientated, whereas the matured bone showed a high degree of preferred orientation. A comparison of No. 2 and 3 shows that the degree of preferred orientation varies from anterior to posterior quadrant in one and the same bone. This difference is less in older individuals. In some of the older bones the relative degrees of orientations were reversed.

The X-ray diagrams of decalcified bones showed that the bone collagen followed the same pattern as apatite i.e. random at birth and becoming oriented with maturity, and the degree of collagen orientation is of the same order as the apatite orientation. Both the fibre direction of collagen and the c-direction of apatite crystals have preferred orientations approximately parallel to the long axis of the bone.

It has generally been assumed that the oriented formation of apatite in mature bone is due to the epitaxial crystallization of apatite on the oriented collagen fibres. In the light of the present investigation it is difficult to visualize a process which may cause randomly oriented collagen fibres in bone at birth to change over to oriented ones at a later age while the proportion of organic matter in bone is decreasing[3]. It becomes more difficult to explain the variation in the degree of preferred orientation of collagen in different areas of one and the same bone. The following tentative explanation may overcome the above difficulties.

The lack of orientation in bone at birth suggests that the formation of apatite and collagen are random and continuing processes without any preferred direction. During the use of the limbs the bones are alternatively stressed and relaxed. During the stressed condition of bone the apatite crystallites which are unfavourably oriented with respect to the direction of stress will tend to dissolve into the body fluid more than those favourably oriented. During the relaxed state both crystallization and crystal growth will occur; but more material will accrue to the favourably oriented, bigger crystals than to the smaller crystals. This differential crystal growth will occur due to the greater thermodynamic stability of larger crystals. As a result of the above two processes the proportion of favourably oriented crystals will increase with the use of the bone thereby making it more capable of withstanding the stress. In the case of the femur the main stress is compressive. A similar stress-induced recrystallization in a preferred direction has recently been reported in the case of ice movement[4].

Collagen, which occupies the intervening space between the apatite crystals, will get oriented either due to packing considerations or due to its epitaxial formation on apatite crystals. This would explain the observed increase in both preferred orientations with age. As it is known that the direction of stress, during normal use, varies with respect to the long axis of the femur from infancy to adulthood. Thus it is not surprising that the degree of preferred orientation varies at different parts of the femur.

It follows from the above hypothesis that a bone subjected to a higher level of stress will have a higher degree of preferred orientation than a bone subjected to a lower stress. To test this deduction X-ray diagrams were taken of the cylindrical part of pubic and rib bones of a crocodile. Of these pubic bone is subjected to a higher stress (being a part of its locomotion system) than the rib. The corresponding intensity ratios were 4.5 and 2 i.e. pubic bone is more oriented than the rib. This is what was expected from the proposed hypothesis. Further work to verify other deductions from this hypothesis is in progress.

Résumé. On a étudié les photographies de diffraction des rayons X de 20 spécimens d'os fémoral d'individus âgés de 6 jours à 76 ans. On a pris des photographies des quadrants postérieurs et antérieurs. Les résultats indiquent que les crystaux d'apatite de l'os d'un enfant nouveau-né étaient orientés au hasard, à l'inverse de ceux d'un adulte.

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