the value of HbBC was determined with the guaiacole peroxid method according to Owen et al.[6] both in the native and heat-incubated samples. The original manual method was mechanized with pipettors, and flowthrough cuvette. The samples were prepared in 6-sec-intervals and measured after 10 min standing in waterbath at 25 °C in the same order with the same intervals. In this way the sera of Hp 1F-1F and Hp 1S-1S phenotypes were analyzed together in the same serial procedure to avoid possible inaccuracy and divergence of the results originating from different reagents and reactions executed in different serials.

Results. The figure summarizes the results of examination. It may be noted that, as compared to the native, the HbBC value was reduced less in sera of the subtype Hp 1F-1F than in those of subtype Hp 1S-1S. Between the mean values of the 2 groups, there is a significant difference after the heat-incubation for 1 h as evidenced by the double-t-test of Student: $t = 4.09$; $p < 0.001$. The alpha polypeptide chains of Hp molecules determined by the alleles Hp^{1F} and Hp^{1S} differ in only one amino acid. In position 54, there is lysine in the alpha-1F chain and glutamine in the alpha-1S chain[7]. This causes a difference in the electrophoretic mobility of the 2 polypeptide chains on the basis of which subtyfying is carried out. According to present knowledge, the beta chains do not differ in individual phenotypes[8]. It may be supposed that this chemical difference in the alpha-chains of Hp causes also a difference in the behaviour of the Hp molecule to heat.

The phenomenon was investigated in whole human serum and not with purified Hp. However, the decrease of the HbBC in the serum under effect of heat may support to the altered behaviour of the Hp. This is justified by the dependence of subtypes and by the earlier demonstrated dependence of usual phenotypes[1,3].

At present there is no explanation for the HbBC reduction of Hp occurring upon the effect of heat. Most probably 56 °C of heat does not cause major changes in the Hp molecule. In the course of experiments made with Laurell's rocket method, there was no degradation product to be seen in heat incubated sera[9]. It is more likely that the change is configurational. The Hp molecules have a structural similarity to Bence Jones proteins[7]. There is a similarity between the 2 proteins in presenting a special behaviour against heat, too. The heat sensibility of the Hp molecules concerns one of its functions, the most characteristic and the most important – as we regard it nowadays –, the hemoglobin binding capacity. The further investigation of the decreasing hemoglobin binding capacity of the haptoglobins exposed to heat may lead to new data concerning haptoglobin-hemoglobin interaction.

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Zinc biochemistry in normal and neoplastic growth processes[1]

B. L. Vallee

Biophysics Research Laboratory, Department of Biological Chemistry, Harvard Medical School, and the Division of Medical Biology, Peter Bent Brigham Hospital, Boston (Massachusetts 02115, USA), 12 October 1976

Summary. Zinc is essential for the growth of all species. Growth arrest results from its deficiency and presumably reflects important roles for this metal at critical points of metabolism. Studies of zinc metalloenzymes show that zinc serves as a coenzyme to more than 80 enzymes, among which are the reverse transcriptases which cause leukemia in many species. Its role in nucleic acid metabolism is emphasized.

Zinc is essential for the growth of all species. Growth arrest results from its deficiency and presumably reflects important roles for this metal at critical points of metabolism. Thus, it was found 25 years ago that normal leukocytes contain substantial quantities of zinc while leukemic cells contain less than 10% of the zinc content of normal leukocytes[2]. The consequences to cellular growth and proliferation of such intracellular decreases in zinc content have only begun to be investigated recently. We have utilized various cell lines including E. gracilis[3], and human leukemic lymphoblasts to examine the involvement of zinc in the biochemical events of cell division. The biological essentiality of zinc can be discerned at various steps of cell growth and development in both normal and neoplastic cells.

The unicellular eukaryote, E. gracilis, has proved most suitable in this regard and has served to define the approaches required to study zinc metabolism in cells cultured in vitro. Importantly, the zinc content of this organism decreases to less than 10% that of normal, zinc sufficient cells when the zinc concentration of the culture medium is less than 10^{-7} M[3]. The resultant deficiency severely limits its growth. A number of striking morphological and chemical derangements are the consequence. Both RNA and protein synthesis are markedly depressed. The DNA content and cellular volume doubles while β-1,3 glucan (paramylon) aggregates. Further, proteins and peptides of unusual composition, amino acids, nucleotides and pyrophosphate accumulate. In addition, the intracellular content of Mg, Mn and Fe increases markedly. Addition of zinc to the culture media completely restores the chemical. morphological and growth patterns to those characteristic of normal E. gracilis cells[4]. These dramatic results show that in zinc-deficient cells both biochemical and morphological processes of the cell cycle, i.e. DNA and/or RNA synthesis as well as function and cell division are markedly disturbed, perhaps as the consequence of the essentiality of zinc to the primary molecular events, responsible for those events. Consequently, we have em-

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ployed Flow Cytofluorometry to define its role in the cell cycle of eukaryotic cells, since this approach permits rapid analysis of the DNA content of intact cells.

Zinc is essential to specific steps of the cell cycle, i.e. G_1, S and the $G_2 \rightarrow M$ transition[5], and its deficiency can block each of them, followed by growth arrest. Such studies with E. gracilis have provided basic information which allows investigation of the role of this and other metals in the metabolism of malignant cells in general, and leukemic cells, in particular. In this manner metal dependent steps pertinent to the $G_1 \rightarrow S$ transition, and S itself have been defined in a human leukemic cell line.

The involvement of the element in the premitotic events of the cell cycle is consistent with earlier observations of its importance to nucleic acid metabolism. Zinc, among other metals, stabilizes the structures of RNA and DNA and presumably affects their metabolism[6]. Moreover, it plays an important role in nucleic acid synthesis and is indispensable to the function of DNA-dependent DNA and RNA polymerases of E. coli[7,8]. The discovery of the RNA dependent DNA polymerase–reverse transcriptase–in Type C oncogenic RNA viruses[9] has stimulated study of the initiation, biochemical basis and maintenance of malignant transformations and of the manner by which a DNA copy is transcribed from viral RNA. The above indications of the importance of zinc in normal and leukemic leukocyte metabolism have prompted us to examine the RNA dependent DNA polymerases from avian myeloblastosis virus and mammalian Type C RNA tumor viruses.

1,1-O-Phenanthroline (OP) reversibly inhibits all of the RNA dependent DNA polymerases from avian myeloblastosis[10], murine, simian, feline and RD-114 RNA tumor viruses[11]. The pK_{OP} varies only from 4.7 to 4.9 and the number of moles of inhibitor bound to the metal, $\bar{n}$, from 1.3 to 2.2, values very close to those previously reported for known zinc enzymes. Combined use of microwave-induced emission spectrometry and micro gel exclusion chromatography has demonstrated stoichiometric amounts of zinc essential to the function of the enzymes from avian myeloblastosis, murine leukemia and woolly monkey Type C viruses[10,11]. Most recently we have shown further that the DNA dependent RNA polymerase II from E. gracilis also is a zinc metalloenzyme. The implications of these findings to normal and abnormal growth will be examined.

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Comparison of transfer ribonucleic acid aminoacylation by various regions of chicken brain

D. S. H. Liu and A. Richardson

Departments of Biological Sciences and Chemistry, Illinois State University, Normal (Illinois, USA),
21 September 1976

Summary. Aminoacylation of tRNA with valine was greater in the cerebrum of chicken than the cerebellum or optic lobes. This greater aminoacylation was due to both the activity of the cerebral valyl-tRNA synthetases and the accepting activity of the cerebral $tRNA^{val}$.

Studies comparing protein synthesis by various regions of brain have shown regional differences[1–3]. Recently Liu et al.[4] reported that the greater cell-free protein synthetic activity of the post-mitochondrial supernatant of the cerebrum of chicken brain compared to the cerebellum or optic lobes was partially due to soluble factors in the post-microsomal supernatant (cell sap), possibly factors responsible for the aminoacylation of tRNA. Takahashi and Abe[5] found that the aminoacylation activity of cell sap from the cerebral cortex of rabbit brain was higher, as determined by ^{32}P-pyrophosphate exchange, than the activity from the cerebellum, medulla, pons, thalamus or hypothalamus. However, Liu et al.[4] found in chicken brain that the cell sap from the cerebellum had a greater ability to attach radioactively labeled amino acids onto tRNA than the cerebrum or optic lobes. Because the cell sap contains both components required for aminoacylation, aminoacyl-tRNA synthetases and tRNAs, it is necessary to study the aminoacylation activity of specific aminoacyl-tRNA synthetases and tRNA when comparing the aminoacylation activities of various brain regions. This study is the first report comparing the aminoacylation activity of tRNA synthetases and the charging capacity of tRNA isolated from different brain regions.

Materials and methods. The post-mitochondrial supernatant and cell sap of the cerebrum (cbr), cerebellum (cbl), and optic lobes (opl) were obtained from 12-18-month-old leghorn hens according to the methods published by Liu et al[4]. Aminoacyl-tRNA synthetases were obtained from the cell sap by adding ammonium sulfate to the cell sap to 51% saturation. The resulting suspension was centrifuged at low speed and the precipitate collected and dissolved in a phosphate buffer pH 7.5 (15% glycerol, 10 mM KH_2PO_4, 5 mM KCl, 1 mM $MgCl_2$, and 20 mM 2-mercaptoethanol). The solutions were dialyzed against the phosphate buffer overnight, passed through a 1.8×20 cm Sephadex G-100 column, eluted with phosphate buffer, and fractions containing protein were pooled and used as the source of aminoacyl-tRNA synthetases.

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