Results and discussion. The quantitative estimates of the inhibitory power of fragmented anti-HCG are shown in Figure 1. The fractionated antibody has a proportionally similar number of antigen binding sites to the intact antiserum except for dilution differences resulting from preparative procedures. Fragmented anti-HCG added to decreasing amounts of WA resulted in the competition curve shown. It can be seen that approximately proportional amounts of FA to WA were needed for inhibition of hemagglutination. The HI_{50} occurred at a whole antiserum protein concentration of 0.117 mg/ml (1:500 dilution). Assuming a minimum of 10% globulin in the whole antiserum, a ratio of inhibitor to antibody of 2.6:1 was obtained at this point.

The HI_{50} value obtained from Figure 1 dictated the WA concentration (0.117 mg/ml) for use in radioprecipitation inhibition. The power of fragmented anti-HCG to compete with WA for radioantigen is shown in Figure 2. The inverse of the cpm in the sediment relates the radioprecipitation inhibition throughout a spectrum of FA concentrations titrated against constant amounts of WA. Maximum RPI (69%) occurred at a FA protein concentration of 60 µg/ml (1:50 dilution). In actuality, the RPI curve represents the precipitin curve of the fragmented antibodies when plotted as the inverse function. The cpm in the sediment are representative of the intact antiserum (WA) interaction with radioantigen since goat anti-rabbit globulin does not readily precipitate the fragments. Correspondingly, the Fc fragments do not react with the antigen[1]; hence, the curve shown in Figure 2 represents the combined precipitin curves of the 2 Fab fractions.

The inhibitory potency of fragmented antibodies may be titered by hemagglutination inhibition to determine optimum FA:WA ratios and the HI_{50} index. These data are then employed to measure the immunoprecipitation inhibition capabilities of fragmented antibody. A precipitin curve can be obtained concommittantly with a curve of the percent of inhibition. Precipitin curve analysis reveals the quantities of specific FA present in the system, the zone of equivalence, areas of antibody and antigen excess, and the molecular combining ratios between antigen and antibody. Thereby, quantitative studies involving fragmented antibodies may be more readily performed.

Zusammenfassung. Das Hemmpotential der Antikörperfragmente bei Kaninchen wird mit neuer Methode demonstriert.

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Dextran Induced Changes in the Electrophoretic Mobility of Rat Bone Marrow Erythrocytes

Large macromolecules like dextran should be expected to influence cellular electrophoretic mobility and this in turn to perhaps produce changes in the cellular metabolic activity[1]. The possibility that cell charge might alter cellular activity has been previously suggested[2]. It was of interest, therefore to determine the influence that various dextran combinations had on cellular electrophoretic mobility, and to determine if electrophoretic mobility could be correlated with an earlier work on oxygen consumption[3].

Materials and methods. Male Holtzman rats, 42 days old, were used in all experiments. Animals were killed by a sharp blow to the base of the skull and cells were obtained from the bone marrow of the 2 femora and tibiae. 10 animals were used, and 10 measurements of 10 cells were made from each animal.

Dextran solutions of 1, 3, and 5% were prepared with 39,500 molecular weight dextran (lot 8, 687, Pharmacia, Sweden). One, 3, and 5% dextran solutions were prepared using 139,000 mol. wt. clinical grade H dextran (lot H 1158, Pharmachem, Pennsylvania) and 228,000 mol. wt. clinical grade HH dextran (lot HH 82262, Pharmachem). The dextrans were put into solution in Tyrode's medium. The osmolarity of the Tyrode's medium and different dextran solutions was adjusted to 310 mOsm/l with NaCl and the pH adjusted to 7.35.

Determinations of cellular electrophoretic mobility of mature erythrocytes were made using a microelectrophoresis apparatus modified from that described by BERNSTEIN et al.[4].

Results. Electrophoretic mobility of mature erythrocytes in the various dextran solutions, isologous serum, and Tyrode's medium is shown in the Figure. Cellular electrophoretic mobility is expressed in relative practical units. Determinations of cellular electrophoretic mobility was limited to mature erythrocytes because this was the only cell type that could be identified with certainty in unstained bone marrow cell preparations using phase and darkfield microscopy.

Erythrocytes of bone marrow suspended in isologous serum had the lowest relative electrophoretic mobility whereas 5% 39,500 molecular weight dextran medium produced the greatest cellular relative electrophoretic mobility.

Similar electrophoretic mobilities were displayed by cells in tyrodes and the 1% 228,000 molecular weight dextran suspending medium. It was found that as the molecular weight of the dextran suspending medium was increased, the cellular mobility decreased. However, as the concentration of a specific molecular weight dextran was increased the cellular mobility increased.

Discussion. The data reported here indicates that cells incubated in medium containing dextran have a greater cell mobility than do cells incubated in either Tyrode's or serum suspending medium. It is of interest to note that in the artificial media combinations the cell mobility is increased from 2 to 10 times as much as that found in serum. Most probably the cellular electrophoretic mobility of the cells in serum and in the artificial suspending medium is the result of different factors. The factors that need to be considered here are: Agglutination, sedimentation velocity, surface charge, and the dielectric of the suspending medium.

Cellular aggregation occurs with dextrans of 70,000 and greater molecular weight, whereas erythrocyte disaggrega-

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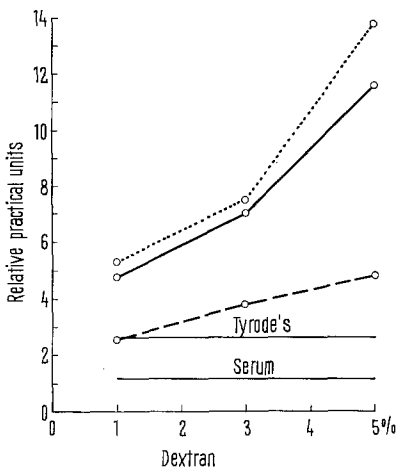
tion or reversal of sedimentation rate has been produced by dextrans of 60,000 and lesser molecular weight[1]. Sedimentation velocity studies by SKOOG and BECK[5] have shown that high molecular weight 228,000 dextrans produce a rapid sedimentation of erythrocytes.

In this study cell numbers in the cellular suspension were reduced so that the spatial distance among cells obviated agglutination problems. The short duration of time in which determinations were made likewise negated sedimentation problems.

According to POLLACK et al.[6] polymers can change the dielectric of the suspending medium without changing the surface charge of an erythrocyte. His laboratory also demonstrated that the dielectric of the suspending medium decreases as the concentration of a given polymer increases. If one assumes that similar situations might exist for all the cells in rat bone marrow; then it would appear possible to measure bone marrow oxygen consumption in order to determine some type of physiological effect of the dextran polymer on bone marrow cells. Using the data presented here, and cellular oxygen consumption data from an earlier work where similar dextrans were used[3], it is not possible at this time to correlate medium dielectric or cell charge with cellular oxygen consumption. Hence, the possible physiological effects of dextran on cellular activity remains obscure. But as pointed out in the earlier publication[3] membrane and cellular hydration may still be suspect as playing a critical role in cellular metabolism.



Electrical charge differences of mature erythrocytes resulting from variations in suspending media. —, isologous rat serum; —, standard Tyrode's balanced salt solution; O····, 39,500 molecular weight dextran in Tyrode's; O—, 139,000 molecular weight dextran in Tyrode's; O---, 228,000 molecular weight dextran in Tyrode's.

Zusammenfassung. Elektrophoretische Zellbeweglichkeit von Rattenknochenmarkzellen in Tyrodelösung mit verschiedenen Konzentrationen und Molekulargewichten von Dextran wurde mit derjenigen von Zellen in isologem Serum und in Tyrodelösung verglichen. Steigerungen in der elektrophoretischen Zellbeweglichkeit gegenüber dem Vergleichswert des isologen Serums zeigten sich in Tyrode- und Dextran-Lösungen. Die elektrophoretische Beweglichkeit des Erythrozyts schien mit der Grösse und der Konzentration des Dextranmoleküls, das in suspendierenden Medien verwendet wurde, verknüpft zu sein.

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Environmental Regulation of Oocyte Growth in the Bay Scallop *Aequipecten irradians* Lamarck

Events in the annual reproductive cycle of marine invertebrates vegetative phase, growth and gametogenesis, maturation, spawning and resting periods – coordinated with seasonal changes in the environment produce the characteristic annual cycle of gonad activity[1]. Temperature, food, and day-length affect gonad development[2–4], but the mechanisms regulating gonad growth and gametogenesis are not clear.

At Beaufort, North Carolina, the gonad growth period of *Aequipecten irradians* Lamarck occurs in summer when temperature, food abundance and day-length are maximal. In the annual cycle, the primary germ cells and oogonia (15 μ) develop before active gonad growth occurs. As the gonads begin to grow, the oocytes enter the cytoplasmic growth phase (23–45 μ). This is followed by the vitellogenesis growth phase (45–104 μ) which is completed as the oocytes reach maturity. Maturation begins with dissolution of the germinal vesicle; later the oocytes become fertilizable eggs by condensation of cytoplasm (63 μ) provided the water temperature is over 20 °C. If it is not, the oocytes disintegrate by vacuolization and rupture of surrounding membrane. Thus a minimum threshold temperature is necessary for the oocytes to become fertilizable eggs[5].

The influence of food and temperature on oocyte growth was determined by exposing scallops collected in the winter (11.8 °C) with neutral gonads to 15 °C and 20 °C and a constant photoperiod of 12 h light and 12 h darkness. One group of scallops held at each temperature was provided with phytoplankton collected from the field as food, while a second group was starved[3]. Oogonia developed at 15 °C but there was no oocyte growth. A similar response was noted for starved scallops at 20 °C. In contrast, fed scallops at 20 °C developed oocytes to completion of the vitellogenesis growth phase and dissolution of the germinal vesicle.

Scallops collected in summer (24 °C) with gonads at the beginning of the growth period and oocytes in the cytoplasmic growth phase (28 μ) were exposed to 10 °C, 15 °C and 20 °C and a constant photoperiod of 12 h light and 12 h darkness. Large quantities of phytoplankton collected from the field were provided daily as food. The

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