Abstracts of Theses for Graduate Degrees

Medical College of Virginia, June, 1968

Studies on Sulfonamides: Physicochemical Properties in Relation to Biological Activity

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The importance of sulfonamides as antibacterial agents is well known. It is generally recognized that these agents act by inhibiting the utilization of p-aminobenzoic acid by bacteria. Currently, the most widely accepted explanation for the structure-activity relationships of these compounds is the Bell and Roblin theory. This theory is essentially a postmortem of the sulfonamides in that it leads to the conclusion that the most active compounds have been synthesized. There is evidence to suggest that this theory is not totally adequate. The purpose of this research was to reinvestigate the Bell and Roblin theory, as well as the interpretations of other workers, in order to explain more clearly the observed structure-activity relationships of sulfa drugs.

In a reevaluation and critique of the present theories, it was found that the Bell and Roblin parabolic pKa-antibacterial activity relationship actually consists of a series of straight lines, each corresponding to a single congeneric group of sulfonamides. A lack of such series in the ascending region of the Bell and Roblin "curve" was noted. A series of N^1-Benzoylsulfanilamides was prepared, tested, and found to generate a straight line in this region.

The Klotz mathematical treatment of the Bell and Roblin curve was examined and found to hold, not for sulfonamides as a whole, but rather for the individual linear relationships generated by congeneric series. This suggests that the "maximum" does not, as it has been construed to mean, constitute a true limit on activity; the Bell and Roblin maximum merely reflects the interplay between the absolute activity and number of sulfonamide ions in contributing to observed activity. On this basis, there is reason to believe that more active sulfonamides can be found. Suggestions on methods to approach this goal have been made.

The Fujita-Hansch treatment of the electronic and lipophilic components of sulfonamide activity was reexamined. Contrary to the results of these workers, it was found that the lipophilic properties of sulfonamides are, in general, not important in determining antibacterial activity. An analysis of some of the limitations of this technique for structure-activity correlation was presented. It was found that misuse of the Fujita-Hansch method can lead to meaningless correlations.

The electronic properties of sulfonamides were investigated for their bearing on biological activity. A series of N^1-phenylsulfanilamides and their N^4-acetyl derivatives was prepared. Transmission of N^1 substituent effects to the para-amino position was investigated by use of proton magnetic resonance (p.m.r.) spectroscopy and infrared spectrophotometry. No evidence could be gained to support transmission through the SO_2 group. Interesting p.m.r. properties of these compounds, including diamagnetic shielding of the sulfanilyl ring protons via a field effect mechanism, as well as a case of rotational isomerism about the phenyl-N^1 bond, were studied.

Transmission of substituent effects *to* the SO_2 group was investigated by infrared spectroscopy. Lack of variation of the sulfur-oxygen bond force constant was found in an extensive series of N^1-phenylsulfanilamides with a wide activity range. This did not support the Bell and Roblin postulate concerning the role of the SO_2 group in sulfonamide activity.

Based on the infrared and p.m.r. experiments, it was concluded that the electronic contribution of N^1 substituents to sulfonamide activity resides at the N^1 position and/or the N^1 substituent itself. Since the sulfonamide ion is considered to be the active species, the N^1 position is likely to be the most important factor. In this regard, the distinct linear relationships observed for individual congeneric series are explicable in terms of distinct mechanisms of interaction of aryl rings with the N^1 position and/or the general electronic state of the N^1 position of the series.

The Incisures of Schmidt-Lanterman

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The primary objective of this study was to establish the presence of the incisures of Schmidt-Lanterman within peripheral nerves—both spinal and cranial—of

representative animals from fish to man. A secondary objective was to study the ultrastructure of some of these nerves, especially the fine structure of the incisures, using the electron microscope.

Several animals were utilized from which a wide variety of peripheral nerves were removed and studied. Fresh-frozen, formalin-fixed, glutaraldehyde-fixed, as well as osmium-fixed sections, were made. Sections were stained by a variety of histological and histochemical techniques and observed with the ordinary light phase-contrast and polarizing microscopes. Ultra thin sections were studied with the electron microscope.

The incisures were present in all the myelinated nerves investigated. Their number varied from nerve fiber to nerve fiber in the same and different animals. The flaring ends of any two incisures faced each other in any one internode, or their narrow ends lay in opposition. A new type of incisure was observed with both the light and electron microscopes, especially in the heavily myelinated nerve fibers. This incisure was termed the "three lip type."

Several other observations were noted while studying the fine structure of the myelinated nerves, such as the presence of pinocytotic vesicles in the Schwann cell membrane, the presence of an axolemma, the absence of a neurilemma, and the presence of several single membrane-bound vesicles in both myelinated and unmyelinated nerve fibers. Vesicles were more numerous in the latter.

It was concluded that the incisures of Schmidt-Lanterman are a constant feature of myelinated peripheral nerve fibers, since they have been observed in all the nerves studied. The myelin lamellae are continuous across the incisures and, therefore, the incisures are not natural communication channels. They are formed, it is suggested, as a result of the linear growth of the axis cylinders and the attempt of the Schwann cells to myelinate this increase in length. The incisures do not follow any particular sequence, but vary within any one internode. The central cores of the incisures are rich in carbohydrate, whereas the peripheral layers are rich in phospholipid.

Loosely Linked Mutations Enhancing L-Arabinose Utilization in *Escherichia coli* B/r

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An unusual revertant of an L-ribulokinase structural gene mutant of *Escherichia coli* B/r has been isolated. Transduction analyses, using bacteriophage Plbt, and growth studies indicate that the revertant contains two mutations. One is in the L-ribulokinase structural gene at or near the site of the original mutation and enables partial utilization of L-arabinose. The second mutation is outside the L-arabinose operon and enhances L-arabinose utilization by the intragenic revertant. Enzyme assays show that the extragenic mutation acts by increasing the rate at which the L-arabinose operon, containing the intragenic reversion, is translated into protein. The extragenic mutation probably increases the quantity of a species of soluble ribonucleic acid that is complementary to the codon of the intragenic reversion. The net result, then, is increased translation of the polycistronic messenger ribonucleic acid of the L-arabinose operon into protein.

Induction of Fusion in Cultured Cells in the Presence of *Para-influenza 3 Virus*

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Progressive infection of HeLa cells by *para-influenza 3 virus* results in cell fusion or syncytium formation in cell culture monolayers. Results from the present study indicated that primary, diploid and heteroploid cells singly infected with *para-influenza 3 virus* fused with normal HeLa cells, demonstrating a massive fusion reaction characterized by syncytia containing 10 to 100 or more nuclei. Conversely, normal primary and diploid cells, when adsorbed with *para-influenza 3*-infected HeLa cells, fused in a minimal reaction resulting in binucleated or trinucleated syncytia. Normal heteroploid cells adsorbed with *para-influenza 3*-infected HeLa cells resulted in a massive fusion reaction. Thus, it seemed that the reaction was mediated by the fusion capacity of the normal cell. Normal diploid and primary cells demonstrated low fusion capacities, while normal heteroploid cells demonstrated high fusion capacities.

Cell susceptibility studies were performed to relate infectivity to cell fusion. *Para-influenza 3 virus* adsorbed, penetrated and eclipsed in the 24 cell types tested. Viral hemagglutinin was produced in every cell type, as indicated by positive hemadsorption studies. Infectious virus was produced by all cell types tested except rabbit kidney, L929 and primary embryonic chick tissues. Since the latter cell lines demonstrated massive fusion reactions when infected and adsorbed with normal HeLa cells, infectious virus production was ruled out as a requirement for cell fusion.

The data indicated that cell fusion induced by *para-influenza 3 virus* required the presence of viral hemagglutinin in the cell membrane of the infected cell. A normal cell had to adsorb to the infected cell through mucoprotein receptor sites for the viral hemagglutinin.

The range of fusion reactions may depend on the number and arrangement of receptor sites, which may vary from cell type to cell type. Thus, maximum fusion would result between infected cells and normal cells with large numbers and compatible arrangements of receptor sites.

Correlations Between Acid-Base Behavior and Ion Fluxes in Frog Skin

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It has been demonstrated numerous times since the 1920's that frog skin can establish and maintain a tenfold to one hundredfold $[H^+]$ gradient from the epidermis to the corium. Investigations to determine the nature of the mechanism for developing and maintaining the $[H^+]$ gradient have not been made. The aims of the present studies were as follows: 1) to investigate the development of the $[H^+]$ gradient under various environmental conditions; 2) to investigate the effects of the glandular secretions on pH at the epidermis of the skin; 3) to determine whether the active transport of Na^+ involves a $Na^+ \rightleftharpoons H^+$ exchange; 4) to determine whether the inhibiting effect of NH_4^+ on active Na^+ transport is explained by alteration in the acid-base properties of the skin.

Experiments were carried out on isolated frog skin and on intact frogs. Frog skin contains 27.8 ± 2.0 mM CO_2 per kg of wet weight. The epidermis continuously releases CO_2 which quantitatively accounts for the H^+ produced at the epidermal surface. The alkalinity of the corium tissue results from its high bicarbonate content. The data collected in the present study show that the operation of the $[HCO_3^-]/[H_2CO_3]$ system is adequate to explain the $[H^+]$ gradient. Within narrow limits, the maximal pH difference across the skin is from pH 6 to pH 8. In individual skins the regulation of the surface pH at the epidermis is remarkably precise.

Na^+ transport diminishes when the bath is below pH 6.0. Within the limits mentioned, there is no direct linkage of Na^+ uptake by, and H^+ release from, the epidermis. At pH below 6.0, Na^+ uptake is diminished, while H^+ is absorbed by the epidermis.

Skin in solutions containing NH_4Cl (10 mM/liter) show a diminished or even reversed $[H^+]$ gradient across the skin. Na^+ outflux is increased; Na^+ influx, net Na^+ flux, and skin P.D. are decreased. Oxygen consumption is reduced slightly, if at all, compared to control skins. H^+ ions are absorbed both by the epidermis of the skin (even at pH 7) and by the corium. Since K^+ was released in nearly equivalent amounts under these conditions, a $H^+ \rightleftharpoons K^+$ exchange appears to take place. It is suggested that the inhibitory effect of NH_4^+ on active Na^+ transport results from the loss of cellular K^+, which, for unknown reasons, is a key element in maintaining the mechanism of active Na^+ transport.

A chemical analysis of glandular secretions showed that these secretions are alkaline, containing $NaHCO_3$ and $KHCO_3$. Epinephrine stimulated release of these fluids, which may have a role in regulation of skin pH for optimal Na^+ transport.

The Mating Pattern of Five Strains of *Drosophila equinoxialis*

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Speciation, i.e., the formation of new species from subspecies or races, cannot be observed directly, since it is a very slow process. Evidence of this process is available in the race-species borderline cases, in which races or subspecies have almost reached the degree of divergence of true species.

In addition to the classical case in *Drosophila paulistorum,* its sibling species *D. equinoxialis* was also found to be a borderline case, although to a lesser degree. Intraspecific crosses of this species show various results: fertile hybrids, or fertile female and sterile male hybrids, or no progeny at all.

Previous investigation had led to the assumption that a sharply delimited receptive period for females of *D. equinoxialis* might be a factor in the reproductive isolation between incipient species.

In this study five strains of *D. equinoxialis* from Tefé, Trinidad, Venezuela, Honduras and Puerto Rico were used in order to determine the time and length of the receptive period. The results confirmed that this species displays much less sexual activity than the other species of the genus, and that the overall rates of insemination are relatively low. However, the five strains do not show as sharply a delimited receptive period as previously found in the Tefé strain. This strain may have changed genetically since that study; Dobzhansky and Pavlovsky have found such changes in *D. paulistorum.*

Three of the five strains studied showed similarities in the bimodality of their insemination distribution with one maximum of sexual activity occurring about three to six days after hatching and the other, approximately 10 to 12 days.

The five strains also showed several differences, namely:

a) in the time when the maxima of their sexual activity occur;

b) in the rates of insemination at the maxima and in the total percentage of insemination during the experiment;

c) in the age at which sexual maturity is reached.

Additional experiments with the Puerto Rican strain disclosed that:

a) the insemination frequency increases not only when the time of confinement increases from 24 to 48 hours, but also when males are older than females (up to the seventh day);

b) a confinement from hatching for various numbers of days gives anomalous results that are not amenable to analysis because of heterogeneity between replicates for each day.

The Intrinsic Back Musculature of *Macaca mulatta*

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Interest in the evolution of primate locomotor systems has brought into focus the functional importance of the intrinsic spinal musculature. The anatomical complexities of this region, however, are such that previous descriptions are limited and lacking in detail. The aim of the present study was to determine the morphology of the epaxial region in the rhesus monkey and to suggest a logical classification and system of nomenclature for these muscles.

The spinal muscles of three specimens of *Macaca mulatta* were dissected bilaterally and described in detail. The observations, when compared with those of previous investigations, not only revealed several morphological details which had not been previously recorded, but also clarified certain areas of confusion. The *m. splenius cervicis* was determined to be a distinct muscular element, though fused to the more medial *m. splenius capitis*. The thoracic portion of the *m. ilicostalis* was observed to have a lateral series of accessory origins arising from the tendons of its own preceding fascicles. The cervical origins of the *mm. longissimus cervicis* and *capitis* were found to arise from the posterior zygapophyses of the lower cervical vertebrae. The division of the *m. semispinalis capitis* into a medial *m. biventer cervicis* and a lateral *m. complexus* was shown to result from differences in attachment of these two parts to the occipital bone. The *m. semispinalis lumbocervicalis* (dorsi) was shown to form a continuous column from the sacrum to the axis.

Mechanical Effects of Acoustic Transients on Tobacco Mosaic Virus

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The mechanical breakage of tobacco mosaic virus (TMV) due to the action of acoustic transients has been investigated. The acoustic transients were produced by transient heating of a Prussian blue dye solution (attenuation coefficient of 1000 per cm) when a ruby laser light (20×10^6 watts) was incident on the dye surface. A quartz piezoelectric transducer was used to determine the amplitude and form of the acoustic wave. The production of acoustic waves by transient heating is discussed, and the theoretical forms of the acoustic wave determined for various boundary and initial conditions are compared to the experimentally measured values.

The electron microscope was used to compare particle length distributions of control TMV solutions and solutions exposed to the acoustic transients. Two conditions of exposure were studied by varying the boundary conditions of the TMV solutions. In one case, the TMV solution was exposed to a single acoustic transient, whereas, in the other, the solution was exposed to an acoustic transient which was reflected within the solution. Significantly greater breakage was produced in the latter case, which demonstrates the importance of boundary conditions in the biological effects of pressure transients.

Calculations were made of the magnitude of hydrodynamical forces producing tensions in the TMV particle. A laser intensity of 1.6×10^8 watts per cm^2 incident on the absorbing dye solution was found to be sufficient to cause significant breakage at the 5% level (Kolmogorov-Smirnov test). The corresponding tension on the TMV particle was calculated to be 6.3×10^{-5} dynes.

A Comparative Cytogenetic Study of Sex-Determining Mechanisms in the Acarina

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Meiosis is described in virgin females, inseminated females, and males of the acarid mite *Caloglyphus mycophagus* (Megnin). The sex-determining mechanism is an XO-type in this species, and the male is the heterogametic sex, having a diploid chromosome number of 15. Oogenesis in mated females is regular.

Pachytene is the earliest meiotic stage observed. Nuclear extrusion is evident during the diffuse diplotene stage in oocytes while seemingly attached to a central structure of the ovary by a cone-shaped organelle. Nurse cells are not evident during the growth period of the oocyte, and it is suggested that nutrients supplied the oocyte during the growth period may be derived from the central structure. A similar structure is also observed in a testis. At metaphase I, eight bivalents are observed. Both products of the first maturation division divide at the second maturation division. After the fusion of the pronuclei, either 15 or 16 chromosomes are observed in cleaving eggs.

Virgins of this species fail to oviposit. A description is given of the aberrant morphology and behavior of bivalents in postdiakinetic oocytes which have not been penetrated by a sperm.

Meiosis is also described in both sexes of the arrhenotokous strain of the anoetid *Histiostoma feroniarum* (Dufour) as well as in a thelytokous strain of this species. Male meiosis consists of a single mitotic division without suggestion of a pseudomaturation division. Oogenesis in the arrhenotokous strain is regular with seven bivalents evident at the first maturation division. At metaphase of the first cleavage division in unfertilized eggs, seven chromosomes are observed, whereas in fertilized eggs, 14 chromosomes are present. The thelytokous strain of *H. feroniarum* is apomictic. Oogenesis in this strain is accomplished by an ameiotic mitosis with only one pseudomaturation division. Fourteen chromosomes are evident at metaphase of this division. The same number of chromosomes is also present in cleaving eggs.

The modes of reproduction in the *Acari*, the cytological and cytogenetic status of the group, as well as the possible evolution of sex determination are discussed in light of the above findings.

The Energetics of Contraction of Isolated Muscles of Frogs as Measured by Their Consumption of Oxygen

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The experiments described deal with the oxygen consumption of isolated sartorius muscles of the frog. Double nerve-muscle preparations were used, and the muscles were stimulated to contract in a moist chamber at 12 C under both isometric and isotonic conditions. In the isotonic series, shortening of the muscles was not arrested by means of a stop. Separate experiments were conducted under conditions in which the loads did and did not reextend the muscles in the relaxation phase of the contraction-relaxation cycle.

The resting rates of respiration of the muscles, nerves and pelvic bone (to which the muscles were attached) were found to be of the same order of magnitude. Changes in length of the muscles did not influence the resting rate of respiration. The resting rate of respiration of muscles in the delta state was found to consist of two phases—an initial phase, in which there was a rapid decrease in the rate of respiration with time, and a secondary phase, in which there was a steady small decrease in rate with time. In the initial phase, the rate of respiration was approximately 70% greater than the values obtained for "normal" muscles. In the secondary phase, the rate of respiration was approximately 40% greater than the normal values. Changes in the length of the muscles did not influence the rate of respiration.

Under both isometric and after-loaded isotonic conditions, the active oxygen consumption followed the behaviour already established in earlier investigations. Heavily loaded isotonic contractions resulted in the consumption of more oxygen than in the isometric situation, and lightly loaded isotonic contractions resulted in the consumption of less oxygen than in the isometric case. The same behaviour was obtained independent of whether or not the loads reextended the muscles in the relaxation phase. Hence, the potential energy dissipated by the falling load is not stored in the muscle as metabolic energy. Qualitatively similar results were obtained for muscles contracting from initial lengths corresponding to sarcomere spacings in the range 2.0μ to 2.5μ. The implications of these results, in terms of current theories of the energetics of muscular contraction, are discussed.

The active consumption of muscles in the delta state was found to be approximately constant under both isometric and isotonic conditions. The values of the active consumptions obtained were approximately 50% less than those obtained for normal muscles. There was no significant difference between the maximal mechanical efficiencies of normal muscles and muscles in the delta state. The average value obtained was about 13%.

Anaerobic Utilization of L-Lactate by a Strain of Human Somatic Cells In Vitro

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Minnesota esophageal epithelium (Minn-EE) cells, a stable heteroploid human cell line of esophageal origin, are unique in being able to anaerobically utilize L-lactate, instead of glucose, in the culture medium. Growth studies showed that Minn-EE cells could grow

under anaerobic conditions when glucose in the medium was replaced by compounds which could supply reduced pyridine nucleotides, i.e., β-hydroxybutyrate and malate. In no case could these reduced compounds elevate the growth level to that attained by cells cultured in lactate or glucose. Glucose could not be replaced by oxidized compounds, such as pyruvate, under these conditions.

Radioisotope labeling experiments revealed that lactate and pyruvate were equally permeable and were metabolized in the same manner by Minn-EE cells. It was shown that these cells contained an endogenous carbohydrate reserve which could supply the energy needed for growth. However, this endogenous reserve could not account for the growth seen.

Studies of the enzyme lactic dehydrogenase (LDH) indicated that the total enzyme activity increased during incubation under anaerobic conditions. The isozyme pattern of LDH remained constant and did not change, regardless of the type of medium or oxygen tension. The types of LDH isozymes present in Minn-EE cells are different than those of other cell culture lines and tend to facilitate lactate oxidation. Aerobic glycolysis is the predominant means by which glucose is metabolized by cells in culture. It is suggested that the lactate formed in this process represents a storage form of reduced pyridine nucleotide which can be drawn upon, when required, for biosynthetic processes.

A Study of Anaerobic Streptococci Isolated from the Subgingival Crevice Area of Man

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Thirty-six strains of obligately anaerobic streptococci were isolated from the subgingival crevice area of 18 clinic patients. The condition of the oral cavities of these patients ranged from perfect clinical health to advanced periodontal disease.

The organisms were studied by a number of classic techniques, which were modified to harmonize with the anaerobic environment necessary for these organisms. While studying the general growth characteristics of these organisms, it was found that high peptone concentrations in the growth media produced substantial increases in growth of the organism. Another finding of interest was that glucose, in concentrations approximating 0.5%, inhibited a large proportion of the organisms studied. Only one strain studied required glucose or other sugar for growth, and this same strain proved to be an obligate serophile.

Microscopic morphology proved variable and was not helpful in grouping the organisms, whereas colonial morphology made it possible to group them into five types.

All strains were tested for a number of biochemical reactions with the following results: Only one strain fermented glucose and a number of other sugars. All strains were catalase negative. Only one strain was capable of gelatin liquefaction. Two strains reduced nitrate to nitrite. All strains were indole negative. A majority of strains formed hydrogen sulfide. Only one strain produced an acid reaction in litmus milk. No grouping of the organisms could be made on the basis of biochemical test results.

Agglutination tests on eight selected strains indicated a heterologous group with only weak cross reactions.

The results of this study would indicate that the anaerobic streptococci selected by techniques used for this study constitute a highly diversified group based on classic techniques of grouping and study. These classic techniques have not proved adequate to group and classify the organisms.

Existing schemes of classification are inadequate, and the isolants obtained in this study cannot be confidently placed in any recognized species.

The study confirms other published data indicating that anaerobic streptococci are indigenous to the subgingival crevice area of man.

Immunoelectrophoretic Analysis of Egg White, Yolk and Plasma Proteins of the Differentiating House Sparrow, *Passer domesticus*

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The objectives of the research were: (1) to analyze the relationship among egg yolk, egg white and plasma proteins during ontogeny of the house sparrow, *Passer domesticus;* (2) to evaluate developmental changes by comparing immunoelectrophoretic patterns of embryo, hatchling and adult plasmas; (3) to study patterns of males and females to ascertain whether each sex had unique proteins; and (4) to compare protein moieties of house sparrow, chicken and human plasmas by observing heterologous reactions.

Various aged birds were bled by cardiac puncture. Total protein content was quantitated by the biuret method. A quantity similar to human plasma protein content was determined, and that quantity was used to elicit the antibody response in white New Zealand rabbits. Plasmas, egg yolk and egg white were reacted

with rabbit antisera by means of the highly sensitive immunoelectrophoretic analysis.

Quantitative and qualitative changes occurred in the plasma proteins during morphogenesis. Neither sex exhibited any unique precipitate. Albumin and one $alpha_1$- and two $beta_2$-globulins characteristically occurred after the eighth day of incubation. Prealbumin was present in embryos, diminished soon after hatching and increased by 14 days after hatching. Gamma globulin formed by five days after hatching; most alpha globulins, by nine days; and beta globulins, by 14 days. $Beta_2$-globulins completed the complement by 23 days. Total protein increased more than two-fold within the period between hatching and fledging.

Both egg white and yolk formed the same $beta_2$-globulin when reacted with antisera against sparrow plasma. Yolk formed the albumin precipitate. Both egg white and yolk seemed to lack gamma globulin.

The protein moieties of house sparrow and human plasmas were immunologically distinct. Albumin and certain $alpha_1$- and $beta_2$-globulins were not species-specific for either house sparrow or chicken plasmas.

The developmental pattern of proteins synthesized in the house sparrow and the chicken was comparable with changes reported in plasma proteins for both developing rat and human. The processes of differentiation were fundamentally similar for all these species.

The Influence of Alpha- and Beta-Adrenergic Stimulation on Sodium Transport Across Frog Skin

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These studies were undertaken to clarify the nature of the alterations in sodium transport produced by catecholamines in frog skin, particularly in relation to the concept of alpha- and beta-adrenergic receptors. The skin model used to interpret the results is as follows: Sodium transport inward is thought to be by a two-step process across the epidermis, i.e., passive movement across the epidermal permeability barrier into the active transport site followed by active transport inward into the internal medium. Outflux, which is small relative to influx (accounting for net inward transport of sodium), may be primarily a passive process. The mucous glands offer a possible pathway for sodium movement, although they are not felt to be important in the unstimulated isolated skin.

The effect of catecholamines and adrenergic blocking agents on sodium flux, short circuit current, and skin resistance was evaluated in isolated frog skin. Alpha-adrenergic stimulation (epinephrine following administration of the beta blocking agent pronethalol) decreased net sodium flux and short circuit current to an equivalent degree. Kinetic studies during alpha-adrenergic stimulation demonstrated a decrease in rate coefficient for entry into the skin-transporting compartment but no change in the rate coefficient presumed to be related to active transport. Beta-adrenergic stimulation (isoproterenol or epinephrine following administration of the alpha blocking agent Dibenzyline) produced an equivalent increase in sodium influx and outflux, with no change in net flux, and development of non-sodium current. The results suggest opposing effects of alpha- and beta-adrenergic stimulation on sodium permeability, although on different pathways for sodium movement; i.e., alpha stimulation decreases sodium permeability of the epidermal pathways for active transport, and beta stimulation increases permeability to sodium via another pathway. The beta effects may be related to mucous gland stimulation. Other evidence suggests that these opposing changes in Na^+ permeability may be mediated by opposing alteration in cyclic 3′, 5′ AMP production in the cells.

The influence of the intravenous administration of catecholamines and a beta-adrenergic blocking agent on short circuit current, skin resistance and sodium flux was studied in the living anesthetized frog. Epinephrine (alpha- plus beta-adrenergic stimulation) and isoproterenol (beta stimulation) produced mucous release and an increase in short circuit current which was not explained by sodium transport change. In contrast to epinephrine, isoproterenol decreased skin resistance and increased sodium outflux. Pronethalol alone decreased short circuit current net sodium flux and increased skin resistance. The results indicate that ion transport across the skin, in vivo, is influenced by alpha- and beta-adrenergic stimulation. They support the concept that the two stimuli produce opposing effects on sodium transport, i.e., alpha stimulation decreases and beta stimulation increases sodium permeability of the skin.