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MINERALOCORTICOID RECEPTORS. J.W. Funder, Baker Medical Research Institute, Melbourne, Australia 3181

Mineralocorticoid receptors (MR) are found not only in classical aldosterone target tissues (kidney, colon, parotid) but also non–epithelial tissues such as hippocampus. In addition, MR have equivalent affinity for cortisol (in the rat corticosterone) as for aldosterone, which circulates at very much lower levels. Aldosterone occupancy of MR in physiologic target tissues but not hippocampus reflects the activity in these tissues of the enzyme 11β–hydroxysteroid dehydrogenase (11HSD), which converts cortisol and corticosterone to receptor–inactive 11–keto congeners; aldosterone escapes equivalent metabolism as its C11–OH is stably cyclized with the very reactive aldehyde group at C18. Studies currently in progress in this area include (1) characterization of the NAD–dependent 11HSD species (11HSD2) operant in renal distal tubule distinct from the cloned enzyme (11 HSD1) expressed in proximal tubule, testis, liver and lung; (2) exploration of the physiologic roles of 'MR' in hippocampus, where they are occupied by cortisol/corticosterone, and function as high–affinity glucocorticoid receptors; (3) recognition of the basis for MR and GR specificity at the transcriptional level, by oncogene product binding and/or response element differences; (4) investigation of the evolutionary basis for non–selective MR by comparative studies on guinea pigs and hamsters, and (5) further studies of the molecular mechanisms underlying the resistance syndrome of pseudohypoaldosteronism, in which independent laboratories have failed to find sequence abnormalities in the coding region of MR cDNA in different kindred with absent receptor binding.

New Genetics and Endocrinology

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NON-TRADITIONAL FORMS OF INHERITANCE THAT ARE RELEVANT FOR PEDIATRIC ENDOCRINOLOGY

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Developments in molecular genetics have made it possible to recognize many new mechanisms of disease. These are particularly important to the pediatric endocrinologist since they explain a number of observations and disorders.

Mosaicism is seen frequently in disorders of growth. It explains many cases of intrauterine growth retardation, asymmetry, overgrowth and cancer. Patchy disorders such as McCune Albright are being recognized to have patches of abnormal cells interspersed with normal cells. Depending on the distribution of those cells, various systems may be involved to different degrees.

Imprinting (parental origin effect) has been recognized as playing a role in a number of disorders of growth including Prader Willi, Weidemann Beckwith and pseudohypoparathyroidism. Specific growth factors may have expression from only one parent.

Uniparental disomy occurs when both chromosomes of a pair come from only one parent. This situation appears to be much more common than previously suspected and may explain both the occurrence of autosomal recessive disorders and disorders having to do with growth, behaviour and survival.

These previously unrecognized mechanisms appear to be relatively common explanations for disease processes. They can account for transgenerational effects and for the non-penetrance and variability which has been observed in the past. It is important to consider them as possible explanations for rare and unusual disorders.

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RECENT DEVELOPMENTS IN THE UNDERSTANDING OF X CHROMOSOME INACTIVATION. Larry J. Shapiro. Department of Pediatrics, University of California, San Francisco, USA.

X chromosome inactivation is a developmentally regulated process, present in all mammals, which has presumably evolved to achieve dosage compensation between males and females in somatic cells. This phenomenon has been under study for more than thirty years in efforts to gain a better understanding of the control of gene expression, the pathogenesis of various X-linked diseases and sex chromosome aneuploidies, and to assess cell lineage and clonality in studies of tumor origins. X chromosome inactivation requires a mechanism for assessing the X/autosome ratio, a process for initiating the cis inactivation of genes on a single X, and a means of stably maintaining a pattern of inactivation once established.

Over the past few years, it has become apparent that not all genes on the X chromosome are subject to inactivation. We have been trying to identify as many such genes as possible as they may provide insight into the mechanisms which produce abnormal phenotypes in the sex chromosome aneuploidies. We have characterized several of these genes, studied the signals which control their expression and investigated their location and biology on the mouse X chromosome. A gene studied by several investigators, *XIST*, has been of particular interest as it is expressed *only* from an otherwise inactive X chromosome. We have found that *XIST* is also expressed from the single X chromosome in the testis during meiosis supporting the old theory that X inactivation is required for functional spermatogenesis.

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GENETICALLY ENGINEERED MICE TO STUDY ENDOCRINE DISORDERS

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Methods for introducing genes into mice (transgenic technology) and inactivating genes in mice (gene "knock out" technology) are rapidly contributing to our understanding of the role which specific DNA sequences play in the physiology of the whole organism. Transgenic mice are derived from fertilized zygotes in which micro injected foreign DNA has integrated into the genome. The introduced genetic sequences are present in all cells of the transgenic animal, and the offspring of a transgenic parent inherit the injected DNA sequences in a Mendelian manner. This genetic engineering approach has enabled researchers to create lines of mice which express genes not normally present, and to examine the effects of this expression at an organismal level. Several examples of how transgenic mice have been used to study endocrine disorders will be described.

Gene "knock out" mice are animals which have been engineered so that genes normally expressed by the organism have been inactivated. The creation of such animals is dependent upon the use of embryonic stem cells. These cells are primitive germ cells which can be grown in culture but which still maintain the capacity when introduced into a blastocyst to contribute to all cell types of the resulting animal. To make a gene "knock out" mouse, embryonic stem cells growing in culture are transfected with a cloned mouse gene designed to contain a mutation precluding its expression. The transfected gene integrates through homologous recombination, replacing the normal mouse gene in the genome of the stem cells. These cells are then introduced into mouse blastocysts, and the resulting animals are bred to derive a line of mice homozygous for the inactivated allele. Such gene "knock out" mice have been used to study the role of a variety of genes in the whole organism since they lack production of specified gene products. Several examples of how these animals have been used to study endocrine disorders will be described.