Ontogenetic Aspects of Hypertension Development:
Analysis in the Rat

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I. Introduction 1228

II. Rat as a Model for Studying Developmental Aspects of Hypertension 1228

III. Late Cardiovascular Effects of Interventions in Critical Developmental Periods 1229

IV. Cardiovascular Abnormalities Linked to Genetic Hypertension 1231

V. Maturation of Cardiovascular Phenotypes in Genetic Hypertension 1233

A. Functional and structural alterations 1233

B. Abnormal neurohumoral regulation 1237

VI. Prevention of Genetic Hypertension Development by Pharmacological Treatment in Critical Periods 1241

A. Early interventions on sympathoadrenal system 1243

B. Pharmacological blockade of the RAS 1244

C. Early transient treatment with other antihypertensive drugs 1249

VII. Modification of Genetic Hypertension Development by Nutritional Interventions in Critical Periods 1249

A. Postnatal electrolyte intake 1250

B. Dietary nutrient intake 1252

VIII. Maternal Influence 1254

A. Embryonic environment 1254

B. Maternal electrolyte intake 1254

C. Maternal protein intake 1255

D. Placental glucocorticoid metabolism 1255

E. Pup nutrition and maternal care 1256

IX. Developmental Aspects of Salt-Induced Hypertension 1256

A. Age-dependent susceptibility to salt-dependent hypertension 1257

B. Pathogenesis of salt-dependent hypertension in young and adult rats 1257

X. Implications for Human Studies 1259

XI. Conclusions 1261

Zicha, Josef, and Jaroslav Kuneš. Ontogenetic Aspects of Hypertension Development: Analysis in the Rat. Physiol. Rev. 79: 1227–1282, 1999.—In this review, we attempt to outline the age-dependent interactions of principal systems controlling the structure and function of the cardiovascular system in immature rats developing hypertension. We focus our attention on the cardiovascular effects of various pharmacological, nutritional, and behavioral interventions applied at different stages of ontogeny. Several distinct critical periods (developmental windows), in which particular stimuli affect the further development of the cardiovascular phenotype, are specified in the rat. It is evident that short-term transient treatment of genetically hypertensive rats with certain antihypertensive drugs in prepuberty and puberty (at the age of 4–10 wk) has long-term beneficial effects on further development of their cardiovascular apparatus. This juvenile critical period coincides with the period of high susceptibility to the hypertensive effects of increased salt intake. If the hypertensive process develops after this critical period (due to early antihypertensive treatment or late administration of certain hypertensive stimuli, e.g., high salt intake), blood pressure elevation, cardiovascular hypertrophy, connective tissue accumulation, and end-organ damage are considerably attenuated compared with rats developing hypertension during the juvenile critical period. As far as the role of various electrolytes in blood pressure modulation is concerned, prohypertensive effects of dietary Na⁺ and antihypertensive effects of dietary Ca²⁺ are enhanced in immature animals, whereas vascular protective and antihypertensive effects of dietary K⁺ are almost independent of age. At a given level of dietary electrolyte intake, the balance between dietary carbohydrate and fat intake can modify blood pressure even in rats with established hypertension, but dietary protein intake affects the blood pressure development in immature animals only. Dietary protein restriction during gestation, as well as altered mother-offspring interactions in the suckling period, might have important long-term hypertensive consequences. The critical periods (developmental windows) should be respected in the future pharmacological or gene therapy of human hypertension.
I. INTRODUCTION

There is general agreement that the relatively high prevalence of hypertension (~15–20%) in most developed countries has a significant impact on the risk of cardiovascular, renal, and other diseases. According to World Health Organization statistics, cardiovascular diseases remain the major cause of death in all developed countries. The understanding of early changes preceding clinical manifestations of hypertension or of other cardiovascular diseases and the knowledge of how cardiovascular risk factors change in the course of time are essential for improving the prevention measures. Although the full manifestation of severe cardiovascular disturbances usually occurs in adulthood and/or senescence, the roots of several polygenic cardiovascular diseases can be traced back to early ontogeny (26, 416, 427).

This review summarizes contemporary information on early abnormalities in the structural and functional development of the cardiovascular system (including their neurohumoral control) and their impact on the subsequent development and maintenance of genetic hypertension. Alterations in neurohumoral regulatory mechanisms [sympathetic nervous system (SNS), renin-angiotensin system (RAS), and endothelium-derived vasoactive factors], cellular growth control, steroid action, lipid metabolism, cell membrane function (ion transport, signal transduction, and cell Ca\(^{2+}\) handling) belong to the most probable candidates for the “primary defects” in the pathogenesis of hypertension. Our attention is focused on the rat as a principal model for the study of ontogenetic aspects of hypertension development. The main issue is to clarify whether the defined stimuli acting in the respective critical periods (developmental windows) can alter the further fate of predisposed individuals. Cardiovascular abnormalities might thus be considered as late consequences of altered development of the cardiovascular apparatus that was affected by certain stimuli operating in earlier stages of ontogeny. There are two seemingly contrasting approaches for how to investigate this hypothesis. Hypertension development in the genetically predisposed organism can be attenuated or prevented by interventions in particular ontogenetic stages (see sects. vi and vii) or the hypertensive process can be induced in the normotensive organism by appropriate stimuli applied in corresponding developmental periods (see sect. ix). Both approaches might well represent the two sides of the same coin.

II. RAT AS A MODEL FOR STUDYING DEVELOPMENTAL ASPECTS OF HYPERTENSION

In both rats and humans, the development of hypertension and its subsequent complications take place in characteristic ontogenetic periods. An effective study of particular cardiovascular alterations emerging in the course of the developmental process (from their early onset until their late consequences) requires the use of adequate animal models. They should be genetically well-defined, accessible for techniques used in cardiovascular physiology, and economically acceptable; in addition, their life span must be short enough compared with that of the investigators. Indeed, a major part of the experimental hypertension research has been carried out in rodents, namely, in the rat (197), which is very convenient for the study of cardiovascular physiology. Although the mouse seems to be a more appropriate tool for genetic manipulations (29, 106, 668), its small size precluded physiological measurements for a long time. The information about the rat genome is still not extensive enough, but multiple inbred rat strains with genetic hypertension (192, 758) represent valuable material for modern genetic analysis ranging from gene-phenotype relationships over quantitative trait loci cosegregation to congenic or transgenic strain construction (286, 591, 611, 637).

The detailed knowledge of numerous aspects of developmental physiology in the rat together with the possibility to induce or to prevent the hypertensive process by specific interventions in particular stages of ontogeny make this animal species a useful tool for hypertension research. In the rat, a wide spectrum of experimental hypertension models is available that differ in the contribution of genetic and environmental factors to the elevation of blood pressure (BP). In some cases, e.g., salt-sensitive Dahl or Sabra rats, the developmental interaction of both the above components is of fundamental importance for hypertension development.

The worldwide availability of spontaneously hypertensive rats (SHR) has made it possible not only to identify numerous cardiovascular abnormalities in this model but also 1) to estimate the degree of their genetic determination (see sect. iv), 2) to study their ontogenetic aspects in detail (see sect. v), and 3) to evaluate their role in the pathogenesis of hypertension by means of various interventions (see sects. vi and vii). Depending on the age at which such interventions were initiated and their duration, research could not only be focused on the therapy of established hypertension but especially on its prevention. Sophisticated attempts to modify the natural course of hypertension development by various transient interventions (pharmacological, nutritional, etc.) have allowed the identification of well-defined periods of remarkable sensitivity of the developing organism to a wide spectrum of exogenous or endogenous factors that can attenuate or accelerate further development of hypertension. An example of this is the long-term attenuation of hypertension development observed in SHR subjected to early transient antihypertensive treatment (7, 70, 263, 472, 600).
The results obtained in SHR represent a major part of our knowledge on genetic hypertension (>90% of papers available in this field). Altogether, SHR studies offer such a complexity of information that could never have been attained in any other strain of genetically hypertensive rats. The frequent use of SHR can be criticized for reasons ranging from the heterogeneity of various breeding colonies (394, 516, 621) to inadequate use of normotensive control strains (588, 678) and to the inappropriateness of SHR as a model of human essential hypertension (139, 477). Nevertheless, the rational use of this hypertensive model provides a lot of valuable information. Genetically hypertensive rats, which are often used for targeting specific hypertensive mechanisms by distinct classes of antihypertensive drugs (successfully employed in the treatment of essential hypertension), might become a valuable tool in the search for specific age periods in which transient drug treatment could prevent abnormal cardiovascular development.

Particular forms of experimental hypertension in the rat represent long-term alterations in the development of the cardiovascular system and its regulations. Such alterations are highly dependent on the genotype, but they can be modified by various environmental factors (nutrition, stress, or exercise) applied during specific ontogenetic periods of increased susceptibility to their action (see sect. III). To a certain extent, this must also be true for human essential hypertension, which is a polygenic disease with an important genetic component, but its progress can also be influenced by some of the above-mentioned environmental risk factors. One of the main drawbacks of past research on essential hypertension was the fact that a considerable effort had been concentrated on the effects of particular risk factors in adults with established hypertension who had already passed the presumed critical periods (developmental windows) of high susceptibility.

III. LATE CARDIOVASCULAR EFFECTS OF INTERVENTIONS IN CRITICAL DEVELOPMENTAL PERIODS

If cardiovascular diseases were considered as late consequences of abnormal cardiovascular development (Fig. 1), the adequate research strategy should be based on 1) detailed knowledge of cardiovascular changes occurring in particular stages of ontogeny, 2) precise definition of critical age periods characterized by increased susceptibility of the developing organism to the action of defined endogenous factors and/or exogenous stimuli, and 3) recognition of differences in the mechanisms by which the organism responds to a given stimulus during or beyond the respective critical period, leading to either permanent transformation or only transient modification of the cardiovascular phenotype.
throughout its whole life span. In contrast to the relatively
precise definition of the above developmental periods, the
onset of senescence in the rat is not yet clearly defined.
Rats older than 24 mo can be regarded as aged, although
there are differences in longevity between various rat
strains (202).

Each developmental period is associated with char-
acteristic transformation of the cardiovascular system.
The development of the cardiovascular system represents
a dynamic sequence of events leading to the expression of
genomic information responsible for the structure, func-
tion, and regulation of particular regions of the cardiovas-
cular apparatus.

The morphogenesis of heart and vessels occurs
mainly during the prenatal period, but it continues par-
tially even after birth. Extensive hyperplasia of the rat
heart, which is typical for the fetal period, still persists
shortly after birth. In this period, the heart grows more
rapidly than the body so that the heart weight-to-body
weight ratio attains its maximum at 4–5 days of postnatal
life and then declines (110). Despite the diminished cel-

ular division, the number of cardiomyocytes in the heart
doubles during the first 3–4 wk of life. The diameter of rat
cardiomyocytes increases from ~5–6 μm at birth to
14–18 μm in adulthood (391, 773). The postnatal increase
of work load and metabolic demands leads to cardiac
enlargement mainly by cell hypertrophy, although the
increased growth rate of rat pups from reduced litters fed
by a single mother is accompanied by enhanced prolifer-
ation of cardiomyocytes (587).

Despite the fact that conduit arteries already achieve
their adult number of smooth muscle layers during em-
byronic development (743), the postnatal thickening of
the media is due to the intensive production of connective
tissue, cell proliferation, and hypertrophy (450, 545). In
the rat, suckling and weaning periods represent the time
of intensive maturation of vascular structure (240) and its
sympathetic innervation (639), which develops concomi-
tantly with the major ontogenetic changes of the SNS
(666, 776) and the RAS (324, 576), both of which are
involved in the regulation of growth and proliferation of
vascular smooth muscle cells.

In the suckling period, rat dams can influence the
development of the cardiovascular phenotype of their
pups not only by behavioral interactions and milk com-
position but also by eliciting the characteristic cardiovas-
cular response, i.e., BP elevation and heart rate accelera-
tion, which occurs in the pups in response to maternal
milk delivery (515).

Prepuberty and sexual maturation represent the
stage when profound readjustment of central hemody-
namics occurs in the rat (9). High cardiac output and low
systemic resistance (per unit of body mass) are charac-
teristic hemodynamic features of weanling rats (584). The
subsequent developmental rise of systemic resistance,
which is accompanied by the decrease of cardiac output,
results in the mature pattern of hemodynamic mechani-


sms maintaining BP in adults (8, 667). The structural
resetting of resistance vessels is responsible for the major
part of the increase in vascular resistance and reactivity
(1, 201), whereas the developmental changes in vascular
sensitivity to vasoactive agents are less important for the
maturational rise in systemic resistance. The intense mat-
uration of the baroreceptor reflex also occurs in these
developmental periods (10, 680).

Considerable cardiovascular changes accompany se-
nescence in both humans and animals (for reviews on
cardiovascular aging, see Refs. 150, 202, 463). The most
prominent senescent alterations concern the structure
and function of arteries that accumulate collagen and
become less elastic (450, 742). Higher vascular stiffness
leads to a further BP increase and might also affect
baroreceptor sensitivity. Furthermore, the balance in the
production of endothelium-derived relaxing and con-
stricting factors is substantially altered in senescent ani-

mals (670, 705).

Detailed knowledge of the ontogenetic changes oc-
curring in the cardiovascular system during particular
critical periods (developmental windows) cannot only
facilitate the search for primary genetic defects (respon-
sible for subsequent development of high BP) but also the
effective targeting on discrete hypertensive mechanisms
in appropriate stages of ontogeny. There are numerous
eamples that particular stimuli influencing the organism
during relatively short critical periods of its early de-
velopement might result in long-term permanent alterations
that often become apparent in adulthood only.

A low protein intake of pregnant rats is associated
with increased BP of their offspring (401). Reduced pla-
cental activity of 11β-hydroxysteroid dehydrogenase
(11β-HSD), an enzyme metabolizing maternal glucocorti-
coids, increases the access of these steroids into the fetus
of rats fed low-protein diets during pregnancy (411). This
indicates that glucocorticoids might participate in pro-
grameing maternal diet-induced hypertension in the rat
(405). Indeed, this type of hypertension can be prevented
by a pharmacological blockade of maternal glucocortic-
soid synthesis (403). Furthermore, fetal exposure to ex-
genous glucocorticoids during the last trimester of ges-
tation initiates the development of hypertension mani-
fested in adult rats (431). Similarly, the inhibition of
placental 11β-HSD activity by carbeneoxolone in pregnant
rats with normal protein intake elevates BP of their adult
offspring (404, 446).

Another example is altered cholesterol metabolism
in adult animals that is partially dependent on the nutri-
tion during early postnatal life (for reviews, see Refs. 246,
308). Maternal milk represents a high-fat nutrition for rat
pups, which is gradually exchanged for a high-carbohy-
drate diet in the course of weaning period, i.e., between
the 15th and 28th day of postnatal life (21). When this stepwise dietary change occurs abruptly as a part of premature weaning, e.g., at the age of 16–18 days after birth, plasma cholesterol falls rapidly in weanling rats (248). The persistent changes in the activity of some liver and gut enzymes involved in cholesterol metabolism (252), which are induced in the weaning period, explain why in adulthood plasma cholesterol levels of prematurely weaned rats are dependent on the diet consumed between postnatal days 15 and 30. Lowest plasma cholesterol was found in animals weaned to a high-fat diet, whereas highest values were observed in rats weaned to a high-carbohydrate diet (250, 251). Plasma cholesterol of adult rats (including the response to high-fat or atherogenic diets) could be modified not only by premature weaning to different diets (249) but also by overfeeding of pups following litter size reduction (247) as well as by an increased dietary fat intake before the natural weaning occurring at the end of the first postnatal month of life (111). This dependence of adult metabolic responsiveness on early nutritional experience might explain the variety of differences between particular breeding colonies and laboratories performing hypertension research.

The major importance of critical periods for the later BP response to this hypertensive stimulus might involve only those mechanisms that are available at a given stage of development in which the stimulus is applied. Moreover, this stimulus represents a different load for immature and adult organisms. Consequently, the activation of distinct pathogenetic mechanisms in young and adult animals exposed to such environmental stimuli could elicit completely different long-term cardiovascular effects (322, 381, 790). The greater response and severe late consequences induced in immature animals can be ascribed to the obvious plasticity and/or vulnerability of the developing organism that is prone to considerable adaptations during early ontogeny.

We assume that the same principles are also true for the development of genetic hypertension. Primary genetic defects lead to the full manifestation of cardiovascular diseases in adulthood or senescence through a cascade of intermediate phenotypes (47). The onset of particular abnormal intermediate phenotypes seems to be confined to distinct developmental periods. The expression of altered genetic information must apparently occur at a very early stage of development because a moderate BP elevation has already been detected in SHR shortly before birth (19). Gray (240) therefore suggested that some primary abnormalities should be present even in fetal SHR. Such early abnormalities (e.g., sympathetic hyperactivity or membrane defects) might serve as important stimuli for subsequent abnormal development of the cardiovascular system and/or its regulation. Their possible correction by interventions in early critical periods should normalize cardiovascular development and prevent the development of genetic hypertension (for details, see sect. vi).

It is evident that the adult cardiovascular phenotype results from the complex interactions between developmental processes and environmental factors during critical developmental periods. Both the intensity of developmental processes and the probability of their modification by environmental factors decrease with progressing age. It is therefore not surprising that cardiovascular abnormalities usually originate from alterations occurring during early stages of ontogeny.

IV. CARDIOVASCULAR ABNORMALITIES LINKED TO GENETIC HYPERTENSION

Up to now, hundreds of abnormalities have been revealed in genetically hypertensive rats by comparing hypertensive and normotensive strains. It is, however, evident that the majority of such differences plays a minor role, if any, in the pathogenesis of hypertension. In the last decade, a wide spectrum of genetic methods has been used to elucidate this problem. More than 200 papers concerned the relationship of candidate genes to particular cardiovascular abnormalities as well as the cosegregation of BP with various quantitative traits. \( F_2 \) hybrid and back-cross populations together with recombinant inbred and congenic strains represent the principal tools for such analysis (285, 637). Despite the tremendous progress in molecular genetics and genetic analysis, the dissection of particular genetic components of complex cardiovascular traits, such as hypertension, is still difficult because of multiple gene-gene and gene-environment interactions (611, 635).

Table 1 shows some quantitative traits or candidate genes that have been found to be linked to BP in segregating populations. Although this list might focus our attention on some important mechanisms, the results of genetic analysis cannot be used as the sole indicative criterion. There are several reasons why both positive and negative results should be considered with caution.

The adult phenotype (e.g., BP level) is a result of multiple interactions of genetic and environmental factors occurring during ontogeny (Fig. 1). The expression of genetic information during particular developmental windows can be substantially affected by specific environmental stimuli that may influence the organism in corresponding critical ontogenetic periods. If this concept is true for hypertension development, it must also be valid.
for the above-mentioned genetic studies. Indeed, the degree of genetic determination of BP decreases with advancing age of animals (304). Moreover, distinct chromosomal loci might have essentially different BP effects in the very same set of F2 hybrids when BP was determined in animals of various age (619).

Thus the age of examined F2 or back-cross animals might be of crucial importance for the outcome of genetic analysis because significant linkage of certain quantitative traits can be found in particular developmental periods only. A classical example of this is altered renal blood flow and glomerular filtration rate, which cosegregated with BP in F2 rats aged 11 wk, but this was not found at the age of 4 or 16 wk (258). This observation suggests that transient changes appearing at particular age periods might be essential for subsequent BP development. Indeed, the diameter of distal afferent arterioles found in 7-wk-old F2 rats correlated inversely with their BP at the age of 23 wk, but media thickness or media cross-sectional area of renal arterioles was no phenotypic predictor of hypertension development (534). On the contrary, the media-to-lumen ratio (M/L) of mesenteric resistance vessels or left ventricular mass of 9-wk-old F2 rats had no significant relationship to BP of 20-wk-old animals (259), although both the mesenteric M/L (due to changes in media thickness) and relative heart weight cosegregated with BP in adult F2 rats (501). Similarly, there was no relationship between relative heart weight of newborns and BP of adult animals of the Prague recombinant inbred strains (158). To our knowledge, significant cosegregation of relative heart weight with BP was never reported in animals younger than 16 wk (238, 390, 501, 777), but it was often demonstrated in older F2 rats in which heart hypertrophy could already develop after a sufficiently long exposure to elevated BP (163, 238, 255, 501, 721). It seems that cardiovascular hypertrophy develops in parallel with the hypertensive process but is not the primary event in the pathogenesis of genetic hypertension as might be concluded from cosegregations reported in adult animals.

Environmental conditions, under which the studied F2 animals are reared, often play a permissive role for the demonstration of genetic linkage. The most typical example is the high salt intake that is sometimes used to augment BP elevation (107, 445, 520). For example, the angiotensin-converting enzyme (ACE) genotype cosegregated with systolic BP only after salt loading of adult F2 rats (287, 520). The same was true for the associations of relative heart weight with diastolic BP (163) or of the angiotensinogen genotype with pulse pressure (449). On the other hand, the cosegregation of the 27-kDa heat shock protein genotype with relative heart weight (255) or of the renin genotype with adrenal renin mRNA (314) were no longer significant after salt loading. Furthermore, substantial differences may exist between animals subjected to a relatively short-term increase of salt intake in adulthood [the above-mentioned genetic studies in SHR or stroke-prone SHR (SHRSP)] and those that were raised on a high-salt diet since weaning (all studies on Dahl rat
in Table 1). Generally speaking, the age-dependent interactions of nutritional or other environmental factors with BP development have almost completely been ignored in the genetic analysis of hypertension.

Further problems, complicating the evaluation of the importance of candidate genes for the pathogenesis of cardiovascular abnormalities exclusively on the basis of genetic analysis, concern the influence of genetic background on the phenotypic expression of particular genes. The same genetic defect might have quite opposite phenotypic effects that depend on the second progenitor strain used for generation of a given \( F_2 \) population. Consequently, numerous genetic studies with salt-sensitive (SS/Jr) Dahl rats demonstrated a wide range of BP effects for SS/Jr alleles of the renin gene (590), ACE gene (140), atrial natriuretic peptide (ANP) receptor gene (140), inducible nitric oxide (NO) synthase gene (137), and the endothelin (ET) system genes (135). Thus the absence of cosegregation might also be due to the inappropriate choice of \( F_2 \) cross used for linkage analysis.

It is becoming clear that the future progress of hypertension research requires a combination of the genetic analysis with ontogenetic and intervention studies. This would facilitate 1) search for specific cardiovascular abnormalities in developing rats predisposed to genetic hypertension, 2) detailed characterization of corresponding pathogenetic mechanisms, and 3) verification of their importance for high BP development by means of the selective interference with particular system(s) in corresponding critical developmental periods.

V. MATURATION OF CARDIOVASCULAR PHENOTYPES IN GENETIC HYPERTENSION

The exact role of particular cardiovascular intermediate phenotypes genetically linked to BP (Table 1) can be elucidated according to their appearance during the hypertensive process, as well as on the basis of their response to defined antihypertensive treatment and/or targeting of specific hypertensive mechanisms. The same approach can be used for the evaluation of multiple candidate genes, the products of which seem to be associated with numerous neurohumoral dysregulations described in rats with genetic hypertension. Despite the substantial effort in the last two decades, it is still difficult to make a precise definition of the primary defect (genetic linkage to BP, early appearance before hypertension development, relevance to BP regulation, and BP modification by specific targeting of the defect) and to demonstrate all its attributes for any of the abnormalities investigated. The polygenic nature of genetic hypertension, the existence of multiple systems substituting each other in the control of particular phenotypes, and the difficult dissociation of primary events from the consequences of elevated BP often preclude the possibility to draw clear-cut conclusions concerning the actual importance of particular cardiovascular alterations. However, our selection of principal abnormalities of the developing cardiovascular system in genetically hypertensive rats has largely adhered to the above-mentioned criteria for primary defects.

A. Functional and Structural Alterations

1. **BP**

The early postnatal BP increase is very rapid even in normotensive rats, in which the mean arterial pressure of newborns is \( \sim 15–25 \text{ mmHg} \) and it progressively increases to \( 80–95 \text{ mmHg} \) within the first 3–4 postnatal week (241, 399, 447). Although the BP of SHR newborns was usually found to be significantly higher compared with that of Wistar-Kyoto rats (WKY) (67, 109, 241), characteristic acceleration of the BP rise in SHR mainly occurs between the 3rd and 10th week of age when their BP rapidly increases by \( \sim 30\% \) above that of WKY. The BP of WKY reaches adult levels by \( \sim 10 \text{ wk} \) of age, but in SHR, it continues to rise at least until the age of 20 wk (Fig. 2). One of the aims of our review is to document that BP development in SHR can be modified especially by the stimuli (including antihypertensive treatment), influencing the animals during the phase of accelerated BP rise that seems to coincide with the period of augmented pressure-dependent vascular hypertrophy (see also sect. vi).

2. **Resistance vessels**

There is no doubt that high BP in hypertensive humans or animals is caused by an elevation of systemic...
resistance, the greater part of which is based on structural changes in the wall of resistance vessels. However, the mutual interaction between structure and function of the arterial wall together with the importance of the distending pressure for both short-term regulation and long-term adaptation of the arterial wall might obscure the mechanisms by which the most important vasoactive systems (such as SNS or RAS) contribute to the rise of vascular resistance in genetically hypertensive rats. Thus the chronic blockade of such a vasoconstrictor system, which already started in the prehypertensive stage to attenuate hypertension development, prevents vascular hypertrophy not only by the elimination of the specific growth-promoting effects of the system investigated but also by the absence of elevated distending pressure. On the other hand, the acute blockade of this system lowers vascular tone, but it cannot affect the changes of vascular reactivity based on vascular wall hypertrophy induced by the above-mentioned long-term influence of the given system. It is thus evident that the exact analysis of certain pathogenetic mechanisms is rather difficult.

Vascular resistance seems to be reduced in newborn and 2-wk-old SHR compared with age-matched WKY (348). At the age of 4–6 wk, the unchanged systemic resistance in young SHR is accompanied by increased cardiac output (186, 454, 667), while the progression of hypertension is associated with elevated systemic resistance and almost unchanged cardiac output (Fig. 2). The early increase of cardiac output is not a necessary prerequisite for the later BP rise because the chronic β-blockade from conception until the age of 12 wk does not prevent hypertension development in SHR (572).

The elevation of peripheral vascular resistance in primary hypertension is partially caused by the decrease of arteriolar lumen diameter due to media thickening or remodeling. In animals with genetic hypertension, the hypertrophy of blood vessel walls parallels the developmental rise in BP and systemic resistance, which is maximal between the 3rd and 10th week of age. On the other hand, the relative reduction of arteriolar lumen size is almost age independent (Figs. 2 and 3). Arteriolar rarefaction is usually absent in young SHR and develops later (56, 583). These structural alterations of resistance vessels are reflected by increases in both minimal vascular resistance (representing diminished luminal cross-sectional area of the resistance vessels at complete relaxation) and maximal vascular contraction (representing active smooth muscle contractile contribution).

It is difficult to estimate the exact role of increased BP in the onset of structural alterations of blood vessels. Although neither BP elevation nor vascular alterations could be demonstrated in some SHR breeding colonies before the age of 4 wk (399, 418, 423), other investigators reported significant vascular changes in fetal (177) and newborn SHR (239, 348). If the BP of SHR were really increased before birth (19, 241), higher wall stress in utero might be the stimulus for altered development of the arterial wall (309). Increased M/L was repeatedly found in different vascular beds of SHR during the first postnatal month (177, 532, 546), together with a higher number of smooth muscle cell layers (418). Media hypertrophy in young SHR can be ascribed to increased smooth muscle cell size and greater amounts of connective tissue (726). The increased elastin content in the aortic wall of developing SHR becomes manifest as an increased number and/or greater thickness of elastic laminae (309, 546). Altered biochemical composition of the vessel wall (increased elastin and collagen) (141) and vascular wall hypertrophy also play an important role in hypertension-induced changes of wall distensibility. During the early phases of hypertension development, wall distensibility might even be augmented in SHR (348, 774), but long-term hypertension decreases arterial distensibility (122, 243, 774).

Functional changes of blood vessels based on structural alterations (increased slope and higher maximum of contraction response, reduced wall distensibility) were already detected in young SHR aged 4–5 wk (201, 372, 500, 601), and they develop in parallel with the progression of hypertension and associated structural resetting of the vasculature (197, 375, 502). On the other hand, the increased sensitivity of vascular contraction to extracellular Ca²⁺ and the higher vascular sensitivity to norepinephrine (NE) (unmasked by cocaine inhibition of NE uptake) are characteristic intrinsic abnormalities of SHR vasculature since prehypertensive stages, but they do not change with advancing age (503, 504).
Early structural changes of the vasculature have sometimes been attributed to primary, pressure-independent genetically determined factors, e.g., increased noradrenergic innervation seen in prehypertensive SHR (5, 277, 418). One of the main arguments for this opinion was that the treatment of SHR dams and their pups with the vasodilator hydralazine prevented BP elevation, but SHR still showed hypertrophy in the mesenteric vascular bed and became rapidly hypertensive after drug withdrawal (663). It is well known that hydralazine treatment augments the activity of both SNS and RAS. In fact, the treatment of SHR dams and pups with the ACE inhibitor captopril completely normalized not only BP but also the media cross-sectional area in the aorta and renal arteries (283). Similarly, neonatal sympathectomy prevented the development of hypertension and substantially reduced vascular alterations in SHR (200, 374, 420).

Further arguments for the contribution of elevated BP to vascular alterations were obtained in experiments with the protection of regional circulation against high BP. The partial constriction of the femoral artery in immature rats aged 3–5 wk, which had normalized local transmural pressure, restored the altered structural and functional development of femoral resistance vessels in SHR (69, 199). It remains to be ascertained whether early regional hypertension can also abolish other vascular alterations found in young SHR, such as diminished β-adrenoceptor-mediated relaxation (210) or augmented myogenic tone due to increased Ca$^{2+}$ influx through L-type channels (14) and/or low Ca$^{2+}$ sequestration in the sarcoplasmic reticulum (371).

Myogenic tone of small resistance vessels is increased in immature SHR but not in adult animals with established hypertension (320). However, myogenic tone of young SHR is not significantly enhanced within the physiological range of distending pressures. It should also be noted that the reduction of lumen diameter (clearly demonstrated in SHR arteries under passive conditions) disappears in the presence of myogenic tone (319). Under isobaric conditions, when the vessels are studied at physiological distending pressures, increased wall-to-lumen ratio is not accompanied by enhanced contractility of distal mesenteric arteries even in adult SHR with established hypertension and pronounced vascular structural alterations (319). Thus further detailed in vitro experiments, which will respect the in vivo pressure conditions, are required to clarify the exact contribution of structural and functional abnormalities of resistance vessels to BP rise occurring during the development of genetic hypertension.

It is evident that in rats with developing genetic hypertension there is a parallel increase of systemic resistance and vascular wall hypertrophy that is most intensive during the rapid phase of BP elevation, i.e., at the age of 3–10 wk. We suggest that this age period might be of fundamental importance for further hypertension development because the vicious circle between increasing distending pressure and progressing hypertrophy of the arteriolar wall is established in this particular developmental window. The attenuation of both pressure-induced and humorally mediated components of vascular smooth muscle hypertrophy by the transient antihypertensive treatment in this juvenile critical period postpones the subsequent BP rise, ameliorates the severity of hypertension, and lowers the incidence of various organ complications (for details, see sect. vi).

3. Kidney

The importance of the kidney in the development and maintenance of genetic hypertension is obvious. This can best be documented by BP effects of renal cross-transplantation between normotensive and hypertensive strains because the genetic predisposition to hypertension can be transferred with the donor kidney to the recipient. This was demonstrated not only in adult rats with established hypertension (49, 126, 350, 597) but also in young prehypertensive animals (127, 204, 373, 595). Although the influence of hypertensive renal damage should always be considered after transplantation of adult kidney (506), several structural and/or functional abnormalities in the immature kidney of genetically hypertensive rats have been proposed to play a primary role in the pathogenesis of hypertension. They range from altered glomerular hemodynamics to abnormal regulation of renal Na$^{+}$ transport due to possible membrane defects.

Lower Na$^{+}$ and water excretion were indeed reported in Milan hypertensive rats (MHS) or SHR aged 3–7 wk. This abnormality is most important in the weaning period and prepuberty because, as the BP rises during sexual maturation (8–9 wk of age), urinary Na$^{+}$ excretion reaches values found in normotensive controls (30, 46, 517). It has been proposed that abnormal properties of the Na$^{+}$-K$^{+}$-2Cl$^{-}$ cotransport system in the ascending limb of Henle’s loop might represent the underlying hypertensive mechanism transferred with the MHS kidney into a normotensive recipient (47). Analogous alterations of this transport system in MHS erythrocytes also persist after bone marrow cross-transplantation (48), indicating that basic alterations of the Na$^{+}$-K$^{+}$-2Cl$^{-}$ cotransport system in MHS are fully genetically determined. The common denominator of ion transport alterations in MHS seems to be point mutation in α-adducin, which is one of cytoskeleton membrane proteins (50). It should be noted that both Na$^{+}$-K$^{+}$-2Cl$^{-}$ cotransport activity (48) and α-adducin genotype (50) cosegregate with BP in $F_{2}$ hybrids. The presence of membrane defect could explain why a transplanted kidney retains its functional properties despite the opposite phenotypic properties of the recipient.

The dopaminergic control of renal Na$^{+}$ handling is
also altered during the development of genetic hypertension. Impaired transduction of the renal dopamine D₁ receptor signal has been proposed to be responsible for the diminished natriuretic response to acute volume expansion in SHR (86). In contrast to WKY, dopamine fails to inhibit the renal tubular Na⁺-K⁺ pump and Na⁺/H⁺ exchanger in SHR (85, 225) due to impaired stimulation of adenyl cyclase, phospholipase C, and protein kinase C (87, 341, 361). D₁ receptor-mediated natriuresis is also defective in salt-sensitive Dahl rats (256), which have a similar defect of dopaminergic control of the Na⁺-K⁺ pump (528) and D₁ receptor/adenylyl cyclase coupling (542) as SHR. A decreased ability of dopamine and/or D₁ agonists to stimulate D₁ receptors in proximal tubules of SHR was already demonstrated at the age of 3 wk (361). As the stimulation of adenylyl cyclase activity through D₁ receptors rises with age in normotensive but not in hypertensive rats, the coupling defect becomes more prominent in adult SHR (190). Recently, the importance of altered D₁ receptor coupling for the pathogenesis of genetic hypertension has been confirmed in F₂ hybrids of SHR and WKY (6).

On the other hand, at least in immature SHR, the impaired Na⁺ and water excretion results from reduced glomerular filtration rather than from enhanced proximal tubular reabsorption (148). Elevated renal vascular resistance in young SHR is predominantly due to the increased resistance of preglomerular vessels (148, 234) that can be partially normalized by early antihypertensive treatment (346). At the age of 4–6 wk, the diameter of afferent arterioles is smaller in SHR than in WKY and undergoes even further reduction in SHR aged 18–20 wk (222, 358). Antihypertensive treatment of young SHR reversed these arteriolar changes, suggesting that the narrowing of afferent arterioles might represent a mechanism protecting glomeruli from direct damage by high BP (359). Nevertheless, renal vascular wall thickening is not only a secondary effect of high BP because it also develops in SHR that had been made normotensive by hydralazine treatment since conception (663). Reduced afferent arteriole diameter might be a good predictor of hypertension development because its value determined at the age of 7 wk cosegregated with BP of 23-wk-old F₂ rats (534). This could also explain why BP of adolescent F₂ hybrids cosegregated with altered renal blood flow and glomerular filtration rate (258).

Enhanced renal sympathetic nerve activity might also contribute to the pathogenesis of genetic hypertension (142, 363, 553) by increasing renal vascular resistance (38, 113) and/or augmenting tubular Na⁺ reabsorption (284). A high renal NE content in young SHR aged 4–7 wk (617) is accompanied by low density of α₁- and α₂-adrenoceptors, the number of which increases in the kidney during the later development of spontaneous hypertension (623). Nevertheless, BP did not cosegregate with the density of either α₁- or α₂-adrenoceptor in the kidney of adult F₂ rats (486). There was also no significant linkage of BP to α₂-adrenoceptor gene polymorphism (366, 685).

It seems that BP effects of renal denervation strongly depend on the age of animals used. Chronic bilateral renal denervation of 4-wk-old SHR (repeated every 3 wk for 16 wk) blocked 30–40% of the expected progressive BP rise, whereas the BP of WKY was not affected (533). Kidney denervation in young SHR shifted the pressure-filtration curve to the left without affecting the pressure-flow relationship. These findings are compatible with the enlarged preglomerular and diminished postglomerular lumen diameters in the denervated kidney (702). Thus renal nerves might affect the structural development of renal vasculature in SHR. In contrast to young animals, renal denervation did not reduce BP of adult SHR (740), suggesting that the kidneys of adult SHR had been reset to maintain a permanently elevated BP.

Another abnormality, which is present in the kidney of immature SHR, is the exaggerated renal vascular reactivity to various vasoconstrictors such as angiotensin II or thromboxane A₂ (TxA₂) (83). This is based not only on increased glomerular density of angiotensin type I (AT₁) (271) and TxA₂ receptors (80) but especially on the altered ability of PGE₂ or PGI₂ to counterbalance the effects of the above vasoconstrictors on renal vasculature (81). The impaired ability of vasodilator prostaglandins to buffer the renal vasoconstriction in young SHR is due to the defect in Gα protein-dependent cAMP generation (82, 84). However, there is no information about its possible linkage to BP.

The resetting of kidney function occurs very early in ontogeny because the relationship between Na⁺ excretion and renal perfusion pressure is already shifted to the right in 3-wk-old SHR compared with WKY (605). The shift of pressure-natriuresis curve was eliminated when young SHR were treated with captopril (478) or hydralazine (365), although the latter treatment did not prevent the structural alterations of renal vasculature (663). Altered renal medullary hemodynamics in young SHR (607) may also contribute to the shift of pressure-natriuresis curve, i.e., to decreased fractional Na⁺ excretion at a given level of renal perfusion pressure. Reduced papillary blood flow, especially prominent in SHR aged 6–9 wk, is the principal abnormality of renal medullary hemodynamics during the development of spontaneous hypertension (607). The renal medulla exerts an important antihypertensive action because its destruction in prepubertal SHR substantially elevated BP of adult animals (40).

The mechanisms controlling the pressure-natriuresis relationship also involve the action of 20-hydroxyeicosatetraenoic acid (20-HETE) produced by renal cytochrome P-450 monooxygenases from arachidonic acid. Individual members of the cytochrome P-450 gene family (CYP4A)
are overexpressed in the kidney of young SHR (383, 638) in which increased 20-HETE production was demonstrated (307, 547, 615). 20-HETE participates in renal blood flow autoregulation and tubuloglomerular feedback control through its potent vasoconstrictor and tubular effects. The inhibition of renal cytochrome P-450 activity lowered BP in young (7-wk-old) but not in adult SHR (430, 616). Chronic inhibition of this enzyme by SnCl₂ treatment of young SHR prevented hypertension development, and BP reduction persisted even if this treatment was withdrawn at the age of 15 wk (185). Because cytochrome P-450 inhibitors are capable of decreasing high preglo\-merular resistance in deep nephrons of young SHR (307), 20-HETE may contribute to the resetting of pressure-natriuresis curve through alterations in papillary blood flow.

An attempt was made to test whether BP of adult rats may depend on renal cytochrome P-4504A genotype (675). Under the conditions of normal salt intake, CYP4A genotype did not cosegregate with BP of F₂ rats, but this genotype had a significant relation to the BP response elicited by a high salt intake in F₂ hybrids of SHR and Brown Norway rats (675). The latter finding is compatible with the role of CYP4A2 gene in the pathogenesis of salt hypertension in salt-sensitive Dahl rats (606, 674).

It is evident that the alterations of renal blood flow, glomerular filtration rate, tubular Na⁺ reabsorption, and pressure-natriuresis relationship can already be demonstrated in young genetically hypertensive rats before the rapid phase of BP increase. Most of these alterations cosegregate with BP in F₂ hybrids, and they can often be corrected by early treatment with ACE inhibitors (for details, see sect. vB). At present, it cannot be excluded that early renal abnormalities could represent primary defect(s) in the pathogenesis of genetic hypertension as it has been suggested on the basis of renal cross-transplantation studies.

B. Abnormal Neurohumoral Regulation

1. SNS

The crucial role of the SNS in the pathogenesis of hypertension is mediated not only by increased resting sympathetic tone leading to enhanced vasoconstriction but also by trophically induced cardiovascular hypertrophy which, at least in SHR, may even precede the rise of BP (1). The importance of the sympathoadrenal system for the development of high systemic resistance and cardiovascular hypertrophy in rats with genetic hypertension has convincingly been demonstrated by neonatal sympa\-thectomy of SHR (125, 200, 374, 420) (for details, see sect. vA).

Although SNS plays a dominant role in BP mainte\-nance of both normotensive and hypertensive animals, there is still sparse convincing evidence for the genetic linkage of SNS abnormalities to high BP (Table 1). However, important SNS alterations occur so early in ontogeny that they cannot be revealed by linkage studies performed in adulthood (usually at the age of 12–18 wk).

Ontogeny of the neural component of BP control as well as the maturation of the functional unit consisting of sympathetic nerve terminals and their target tissues (i.e., vascular smooth muscle) is most intensive in the first 3–5 wk of postnatal life of the rat (166, 470, 666, 710). In certain vascular beds of SHR, enhanced sympathetic innervation was observed before the onset of hypertension (for review, see Ref. 277 and impairment of the vascular wall developed in parallel with enhanced sympathetic activity (666). Thereafter, the development of SNS alterations in SHR slows down but continues until the age of ~24 wk (337).

The early increase of sympathetic innervation in SHR seems to be related to elevated levels of the nerve growth factor (NGF) that were found in the mesenteric artery and aorta of young SHR aged 3–8 wk but not in those of adult animals (165, 713, 776). Nerve growth factor mRNA in the mesenteric vessels of SHR is already increased at birth (187). In addition, NGF gene is linked to the BP in F₂ hybrids (342). As a trophic protein, NGF also takes part in vascular hypertrophy mediated by enhanced sympathetic innervation of blood vessels. Such interaction seems to be most intensive during the weaning period, i.e., in the third and fourth postnatal week (776). In fact, chronic treat\-ment of newborn normotensive rats with NGF for 2 wk had no effect on vascular morphology (421), whereas NGF treatment prolonged until puberty increased vascular sympathetic innervation and caused a hyperplastic response of vascular smooth muscle cells (similar to those seen in SHR), although it did not elevate BP (775).

In hypertension, the postsynaptic α₁-adrenergic func\-tions become dominant, whereas β-adrenergic functions are attenuated. This is reflected by a reduction in the number of β-adrenoceptors and in the production of its second messenger, cAMP, whereas the number of α₁- adrenoceptors remains unchanged or is increased, and the production of their second messengers, inositol trisphosphate and diacylglycerol, is enhanced in cardiac and vascular tissues (for review, see Ref. 131). However, ontogeny of altered adrenergic functions in genetically hypertensive rats is still not fully understood.

The increased sympathetic innervation of resistance vessels (5, 277, 418) and the enhanced BP response to ganglionic blockade (666) in young SHR suggest augmented vasoconstriction mediated by α₁-adrenoceptors. This might be not only due to a greater NE release from sympathetic nerve fibers of SHR (733) but also due to the hyperreactivity of resistance vessels (caused by well-known structural changes) or their supersensitivity to
α1-adrenoceptor agonists (unmasked by cocaine inhibition of neuronal catecholamine uptake) (372, 503).

The reduced density of β-adrenoceptors in the aorta (68) and heart (441, 755) of adult SHR contrasts with unchanged values in prehypertensive SHR (55, 58, 755). However, the unaltered β-adrenoceptor density in young SHR might involve lowered β1-adrenoceptor density counterbalanced by increased density of β2-adrenoceptors (487). The early increase of myocardial NE content precedes the downregulation of β1-adrenoceptors and the increase of Gα protein expression leading to desensitization of cardiac adenyl cyclase in SHR (58). These changes were detected in the heart and aorta of young SHR before the development of spontaneous hypertension (461).

Presynaptic modulation of NE release from sympathetic nerve endings is an important mechanism by which numerous vasoactive substances or drugs can affect vascular tone (707). Diminished feedback inhibition and increased feedback facilitation, which enhance NE release per nerve impulse, seem to be augmented in genetic hypertension. Spontaneously hypertensive rats are characterized by a reduced negative feedback of NE release mediated by presynaptic α2-adrenoceptors. This impairment is usually greater in younger (4–10 wk old) than in older SHR (218, 687, 706). Early alteration of this feedback control is compatible with elevated plasma NE levels observed only in young but not in adult SHR (561, 687). The dysfunction of presynaptic α2-adrenoceptors reveals the facilitatory effects of β2-adrenoceptors in the vasculature of SHR. Enhanced facilitation of NE release (mediated by presynaptic β2-adrenoceptors) results in augmented pressor responses to periarterial nerve stimulation that precedes hypertension development in SHR (709). In contrast to normotensive rats, which are characterized by an age-related loss of both vascular α- and β-adrenoceptor responsiveness, SHR exhibit an age-related loss of vasodilator responsiveness mediated by β-adrenoceptors despite the persisting high vasoconstrictor responsiveness to α-adrenoceptor agonists (60).

The modulation of sympathetic neurogeno vasoconstriction through presynaptic β2-adrenoceptors stimulated by epinephrine also explains how the adrenal medulla could be involved in the early development of spontaneous hypertension. Hypertension development was attenuated only in those SHR that were subjected to adrenal demedullation at 6 wk of age or younger. Furthermore, hypertension development in young demedullated SHR was restored by chronic epinephrine supplementation (59).

The enhanced sympathetic innervation due to elevated NGF levels is a characteristic finding in the vasculature of immature SHR before and during the rapid phase of BP rise. Augmented vasoconstrictor response to α1-adrenoceptor agonists, the downregulation of β1-adrenoceptors, and enhanced NE release from sympathetic nerve endings (due to high sympathetic nerve traffic together with decreased feedback inhibition and increased feedback facilitation of neurotransmitter traffic mediated by presynaptic α2- and β2-adrenoceptors) were also demonstrated in weanling and prepubertal SHR.

2. RAS

The importance of this system for the pathogenesis of genetic hypertension is underlined by the fact that the blockade of RAS by early gene therapy (318, 452, 575) or by pharmacological interventions with ACE inhibitors or AT1 receptor antagonists in the juvenile critical period (207, 262, 498, 538, 749) may attenuate or even prevent the development of hypertension.

The genetic evidence for the participation of RAS in the pathogenesis of hypertension is summarized in Table 1. Thus the renin gene (592), ACE gene (140), as well as AT1A and AT1B receptor genes (135, 136) cosegregate with BP in f2 populations derived from salt-sensitive Dahl rats. On the other hand, the ACE gene (287, 520) but not the renin gene (445) or angiotensinogen gene (298) was linked to BP in SHRSP × WKY f2 rats, whereas BP was significantly associated with the renin gene (395, 686) and ACE gene (779) but not with the angiotensinogen gene (449) in SHR × WKY f2 hybrids. However, recent experiments with the renin gene transfer do not support the importance of this candidate gene in the pathogenesis of high BP in SHR (677) or Dahl rats (329, 679). As far as the functional parameters of peripheral RAS are concerned, BP cosegregated with renal renin secretion (642) but not with plasma levels of ANG II (299). The latter observation is not surprising because local tissue RAS activity might be more important than circulating ANG II.

Recent experiments with gene therapy of immature SHR (318, 452, 575) have demonstrated that the perinatal delivery of AT1 receptor antisense cDNA gene caused a long-term attenuation of hypertension development that lasted up to 7 mo of age. The early expression of AT1 receptor antisense transcript in various tissues resulted in persistent selective attenuation of the respective cellular ANG II actions. This was documented by reduced AT1 receptor mRNA levels in the mesenteric artery and adrenal glands, decreased ANGII binding to AT1 receptors in the heart, diminished contractile response of aorta and renal resistance arterioles to ANG II, attenuated pressor response to ANG II administration, and lowered diposogenic effects of ANG II. Furthermore, it restored the endothelium-dependent vasorelaxation to ACh and corrected the contractile response to phenylephrine or KCl. Finally, this early gene therapy prevented the alterations of vascular Ca2+ homeostasis characteristic for untreated SHR. All these changes were still observed 3–7 mo after a single AT1 receptor antisense gene delivery to SHR pups.
aged 5 days (223, 452, 464). Because losartan exerted no significant effects in SHR subjected to early gene therapy (318, 452), the above antisense delivery seems to have similar cardiovascular effects as chronic treatment of young SHR with AT1 receptor antagonists (456, 498, 538). It is of interest that such early gene therapy lowered the BP just at the age of 4–12 wk, i.e., in the developmental window (critical period) in which transient antihypertensive therapy is capable of inducing long-term BP reduction that persists even after drug withdrawal (see also sect. vB and Fig. 5). The above observations should attract our attention to RAS abnormalities detected in SHR during the first three postnatal months.

Multiple abnormalities of renal and circulating RAS have been described in immature rats with genetic hypertension, the kidney of which is characterized by increased reactivity to ANG II. Preglomerular vessels of young SHR are hyperresponsive to ANG II (723). Angiotensin II-stimulated Na+ reabsorption in proximal tubules is substantially enhanced in 5-wk-old SHR (693). An elevated renal ANG II content and increased ANG II receptor binding capacity in tubular brush-border membranes were indeed found in immature SHR (465), together with a greater number of glomerular (117, 271) and proximal tubular AT1 receptors (92). Renal RAS seems to be more active in young SHR compared with age-matched WKY. Increased renin mRNA levels (620), higher kidney renin activity (417, 652), and enhanced renal renin secretion (11, 282) might contribute to the elevation of plasma renin activity occasionally reported in young SHR (641), although the last observation was not confirmed by later studies (22, 357, 649).

There are also signs of hyperactive brain RAS in immature SHR. Higher ANG II levels (574) together with increased ANG II receptor binding capacity (618) and augmented neuronal response to ANG II (524) were reported in the hypothalamus of young SHR. The levels of angiotensinogen and angiotensinogen mRNA are also elevated in the brain of immature SHR (279, 691).

Enhanced activity of renal and brain RAS together with the greater responsiveness of the kidney and hypothalamus of immature SHR to ANG II explain why the early impairment of AT1 receptors causes long-term attenuation of the development of genetic hypertension. On the other hand, the contribution of direct and indirect effects of ANG II to the elevation of vascular tone and to the hypertrophy of the resistance vessel wall in rats with developing genetic hypertension still remains to be determined.

3. Kallikrein-kinin system

The cosegregation of BP with restriction fragment length polymorphism marking the kallikrein gene family of SHR in Prague recombinant inbred strains (581) indicates the importance of this system in genetic hypertension (74, 462). Further evidence was provided by human tissue kallikrein gene delivery that lowered BP in various hypertensive models including SHR and salt-sensitive Dahl rats (77). A single injection of the respective cDNA construct to newborn SHR attenuated the development of genetic hypertension as evidenced by the BP decrease found at the age of 9–17 wk (78). In contrast, systemic kallikrein gene delivery to adult SHR lowered their BP for 6 wk only (78, 725).

The important role of the kallikrein-kinin system for BP development was further supported by the studies on the early blockade of bradykinin B2 receptors. It was demonstrated in normotensive rats that bradykinin B2 receptor blockade during prenatal and/or early postnatal periods causes BP elevation in adulthood, whereas the same treatment of adult animals has no BP effects (457).

Such a protective role of bradykinin in BP ontogeny is missing in SHR (182) in which reduced urinary kallikrein excretion has been demonstrated from the age of 4 wk, the abnormality being more pronounced in adult rats (3, 580). The association of low urinary kallikrein excretion with BP elevation has also been confirmed in inbred low-kallikrein Wistar rats that have higher BP and augmented salt-induced BP rise compared with Wistar rats with normal urinary kallikrein excretion (458). Similar findings were obtained in kininogen-deficient Brown Norway rats (349a).

The information on the changes of tissue kinins during the development of genetic hypertension is still rather contradictory. The kininogenase activity in the aorta was considerably suppressed in both prehypertensive and hypertensive SHR (560). The renal kallikrein content is also decreased in Milan hypertensive rats (23) in which this abnormality is present since birth (188). Although the tissue active kallikrein content in the renal cortex is also reduced in SHR since birth, its kininogenase activity (expressed per mg protein) is elevated in SHR from the age of 4 wk and decreases in senescence only (193, 580).

Elevated bradykinin levels were indeed found in various tissues (kidney, adrenal, lung, and heart) of young SHR aged 6 wk, but this was not seen in older animals (72). The brain kallikrein-kinin system is hyperactive especially in SHR aged 5–6 wk, although elevated kinin levels together with a higher kallikrein level and augmented kininogenase activity partially persisted in the cerebrospinal fluid of adult 18-wk-old SHR compared with age-matched WKY (355).

The available data suggest that the impairment of the kallikrein-kinin system plays an important, age-dependent role in the pathogenesis of genetic hypertension, because the early (perinatal) bradykinin “deficiency” is associated with the later BP elevation, and vice versa.
4. ANP

Atrial natriuretic peptide represents an important component of chronic BP regulation, because it can effectively decrease BP by its natriuretic and vasorelaxant effects. Indeed, chronic blockade of endogenous ANP by a specific monoclonal antibody accelerated hypertension development in SHRSP (312). On the other hand, chronic inhibition of ANP degradation, which increases plasma ANP levels, prevented salt-induced acceleration of hypertension development in SHR (330). Furthermore, salt-induced exacerbation of hypertension was prevented in SHR by long-term ANP infusion that elevated plasma ANP to levels seen in WKY fed a high-salt diet (331).

Atrial natriuretic peptide also participates in central baroreflex control of sympathetic nerve activity. Acting on forebrain structures [anterior hypothalamic area (AHA)], ANP elevates BP, whereas its action on hindbrain sites [nucleus tractus solitarii (NTS)] lowers BP (551). Consequently, a microinjection of ANP antibody into AHA caused a dose-dependent BP decrease in SHR but not in WKY (763), whereas antibody administration into NTS elevated BP and restored impaired baroreflex control of the heart rate and sympathetic nerve activity in SHR (764, 781). In young SHR, a reduced ANP content and lower number of ANP binding sites in the forebrain (25, 473) seem to be less important for hypertension development than the decreased ANP content combined with diminished ANP binding in hindbrain structures (25, 614).

The genetic analysis revealed that the ANP locus is linked to BP in SHR × WKY F2 hybrids (778, 780). Furthermore, the ANP receptor (guanylate cyclase A) gene was also found to cosegregate with BP in some F2 populations derived from New Zealand genetically hypertensive rats (265) or salt-sensitive Dahl rats (140).

The age-dependent involvement of ANP in the pathogenesis of genetic hypertension is still not clear, but the overexpression of human ANP gene decreased BP only in young (4-wk-old) but not in adult (12-wk-old) SHR (443). Although immature SHR are lacking sufficient ANP secretion, they are able to respond adequately to its vasorelaxant and natriuretic action. Impaired ANP release might be a reason why plasma ANP levels are usually not elevated in immature SHR, but they increase with advancing age and/or with the progress of hypertension (245, 306, 766). Moreover, the atrial ANP content is reduced in adult but not in immature SHR (245, 689, 766). Less effective atrial ANP release after saline infusion and/or atrial pressure elevation has already been demonstrated in young SHR, the difference between SHR and WKY being augmented with increasing age (613). An exaggerated natriuretic response to exogenous ANP was reported in 6- but not 11-wk-old SHR (578). Similarly, vascular relaxation and cGMP response to ANP were enhanced in mesenteric arteries of 4-wk-old SHR but not in vessels of animals aged 12–16 wk (629).

All available data indicate that the insufficient ANP release in young genetically hypertensive rats decreases the potency of ANP system to lower BP. The reduction of vasorelaxant and natriuretic ANP effects in adult genetically hypertensive rats further impairs the antihypertensive role of ANP during the later hypertensive stages.

5. Endothelium-derived vasoactive factors

The importance of the endothelium and its multiple vasoactive products in the regulation of vascular tone under normal and pathological conditions has intensively been investigated in the last decade (for review, see Refs. 162, 196, 531, 627). It is beyond the scope of this review to describe all complex regulatory interactions between circulating and locally produced vasoactive factors as well as their influence on vascular smooth muscle in various forms of genetic hypertension. Our attention will therefore be focused on the developmental abnormalities of principal endothelium-derived vasodilating and vasoconstricting factors including NO and endothelins.

In fact, there is only little evidence that NO or ET alterations are linked to genetic hypertension. The respective genetic analysis has never been performed in SHR in which the development of endothelial dysfunction was studied in detail. The only available information has been obtained in Dahl rats in which particular forms of NO synthase (NOS) and various components of the ET system were tested for cosegregation with BP. Under the conditions of high salt intake, ET-3 (100) as well as ET-2 and ETB receptor loci (135) were suggested to be linked to BP in F2 hybrids derived from salt-sensitive Dahl rats. Surprisingly, neither constitutive endothelial NOS (Nos3 gene) nor neuronal NOS (Nos1 gene) was found to be linked to BP, but the locus for inducible NOS (containing Nos2 gene) cosegregated with BP in at least two different F2 populations (137, 138). Nevertheless, subsequent genetic analysis of this broad region of rat chromosome 10 excluded Nos2 as a candidate gene (170).

The lack of genetic linkage between NO and high BP is rather unexpected because the stimulation of NO synthesis by l-arginine treatment prevents the development of salt hypertension in young salt-sensitive Dahl rats (90, 296), and the delivery of the human endothelial NOS gene attenuated hypertension development in adolescent SHR (444). Recently, we have demonstrated an inverse relationship between BP and vasodilator effects of endogenous NO in F2 hybrids derived from the cross of hypertensive Prague hereditary hypertriglyceridemic rats with normotensive Lewis rats (unpublished data).

The available data do not support the idea that NO participation in BP homeostasis is attenuated during the early stages of genetic hypertension. Both endothelial
NOS (eNOS) expression and plasma NO\textsubscript{2}/NO\textsubscript{3} levels are unchanged in 4-wk-old SHR, and their alterations appear later during the progression of hypertension (96). Production of NO in vascular smooth muscle after the activation of inducible NOS (iNOS) is augmented even in prehypertensive SHR (754). Increased urinary NO\textsubscript{2}/NO\textsubscript{3} excretion as well as higher vascular eNOS and renal iNOS proteins have also been reported in immature SHR (720). Thus the l-arginine/NO pathway is upregulated in young SHR both before and after the onset of hypertension. Consequently, the development of hypertension in SHR is not due to primary defect in NO production. The only impairment of NO synthesis up to now reported in prehypertensive SHR concerns constitutive eNOS. This enzyme, which requires higher amounts of its cofactor tetrahydrobiopterin in the hypertensive strain, produces less NO but more superoxide anions in the aorta of 4-wk-old SHR compared with WKY (118). Although increased superoxide production contributes to the enhanced inactivation of NO in SHR (634), the relative contribution of NO to BP maintenance is similar in SHR and WKY until the age of at least 24 wk (490, 757).

Vascular NO is responsible for a major part of the vasodilator action of the endothelium-derived relaxing factor (EDRF), the release of which is stimulated by ACh. The hypotensive effects of ACh are not altered in young SHR, but the impairment appears in mature SHR in parallel with aging and/or hypertension progress (436, 700, 757). The extent of NO participation in the maintenance of basal vascular tone as well as in ACh-induced vasorelaxation can partly be estimated from the in vitro production of cGMP by vascular guanylate cyclase. Basal cGMP levels are unchanged in arteries of younger SHR but reduced in SHR aged 12–20 wk. The ACh-induced formation of cGMP decreases more with age in SHR than in WKY (213, 650). Moreover, there is increasing evidence for the gradual reduction of ACh-induced endothelium-dependent vasodilation of isolated blood vessels with increasing age in both normotensive rats (670) and SHR (315, 367, 728). These age-dependent changes can be ascribed not only to a progressive reduction of EDRF but especially to a gradual increase of endothelium-derived contracting factor (EDCF).

The age of experimental animals is a major factor determining the in vitro vasoconstriction induced by higher ACh concentrations in blood vessels. This is mediated by cyclooxygenase-sensitive EDCF, which is usually considered to be PGG\textsubscript{2} (315, 349) or TxA\textsubscript{2} (130, 367). Acetylcholine-induced vasoconstriction gradually increases with age and is substantially greater in SHR compared with WKY (315, 367, 728). Thus the increased EDCF production is a major feature of endothelial dysfunction in aged SHR.

Thromboxane A\textsubscript{2} might play an important role in the pathogenesis of spontaneous hypertension not only through its vasoconstrictor action but also by promoting vascular smooth muscle cell proliferation (311). Indeed, the delayed onset and/or attenuated hypertension development was found in SHR and Lyon hypertensive rats chronically treated with some TxA\textsubscript{2} synthase inhibitors and/or TxA\textsubscript{2} receptor antagonists (61, 224, 647). In contrast to young animals, the treatment of adult SHR with TxA\textsubscript{2} synthase inhibitor had no BP-lowering effect (647).

Among the mediators responsible for the increased tone of resistance arteries in hypertension are also endothelins (for review, see Refs. 272, 522, 627). The mesenteric arteries and mesangial cells from young SHR release in vitro more ET-1 than those from WKY (305, 491), but the plasma levels of immunoreactive ET-1 in both young (3–6 wk old) and adult (12–18 wk old) SHR usually do not differ from those of age-matched WKY (301, 368). Young SHR also do not differ from age-matched WKY in the sensitivity of their mesenteric vessels or aorta to ET-1 (123, 701). The chronic blockade of circulating ET-1 by specific antibody did not influence BP development in either young or adult SHR and SHRSP animals (360, 688). Similarly, chronic treatment of both young and adult SHR with the combined ET\textsubscript{A}/ET\textsubscript{B} receptor antagonist bosentan had no effect on the BP (343, 437, 438, 659). However, chronic ET receptor blockade attenuated the acceleration of hypertension induced in SHR or SHRSP by a high-salt diet (544, 673) or DOCA-salt treatment (439), suggesting that ET-1 may be involved in the pathogenesis of salt-induced hypertension rather than of spontaneous hypertension.

The endothelial dysfunction leading to impaired vasodilation in SHR develops in parallel with the progress of hypertension. Decreased endothelial production of vasodilator factors together with increased formation of vasoconstrictor agents seem to be the consequence of endothelium damage induced by long-term BP elevation. This opinion is further supported by the beneficial effects of antihypertensive treatment on endothelial dysfunction in genetically hypertensive rats (for details, see sect. viB2). On the other hand, we cannot exclude the possibility that increased formation of oxygen free radicals can lower vasodilator effects of NO through its enhanced inactivation, but the impaired endothelium-dependent vasorelaxation was rarely observed in immature SHR. Thus the endothelial dysfunction in spontaneous hypertension is most likely the insufficient compensation of high BP in adulthood rather than a primary pathogenetic defect in youth.

VI. PREVENTION OF GENETIC HYPERTENSION DEVELOPMENT BY PHARMACOLOGICAL TREATMENT IN CRITICAL PERIODS

Numerous structural and functional alterations of the cardiovascular apparatus appear during the period of
rapid BP rise which, in the rat, takes place at the age of 3–10 wk. The appropriate antihypertensive treatment applied during this period should not only prevent the development of high BP but should also limit cardiovascular hypertrophy. The regression of cardiovascular hypertrophy is usually the greater the earlier the antihypertensive treatment had been introduced in developing animals (730, 732). The age-dependent efficacy of antihypertensive treatment has been demonstrated not only for ACE inhibitors (2, 263, 276) but also for β-blockers (33, 732) or hydralazine plus guanethidine (730); namely, the latter drug combination was relatively effective in normalizing the cardiovascular abnormalities in young SHR (731). It is evident that the older SHR animals are and the longer the duration of their hypertension is, the more difficult it is to induce the regression of hypertensive structural changes in their heart and vessels. The increasingly poor reversibility of cardiovascular alterations in long-standing hypertension indicates the presence of structural changes (e.g., collagen accumulation in the vessel wall) that develop more gradually than smooth muscle hypertrophy. Collagen regression is far less readily achieved by antihypertensive therapy than the reversal of smooth muscle hypertrophy (742). The enhanced accumulation of collagen in the arterial wall of SHR might be a nonspecific consequence of long-term BP elevation. The existence of similar changes in immature rats with induced forms of experimental hypertension (see sect. ix) suggests that an altered content of noncontractile proteins in blood vessels is a part of the response of the developing organism to high BP, irrespective of the cause of the hypertensive process.

The beneficial effects on the development of high BP and cardiovascular hypertrophy, which were achieved by long-term treatment of young (but not old) SHR with a combination of sympatholytics and vasodilators (730), have facilitated the discovery of the juvenile critical period for the development of hypertension in SHR (7). In 1974, Albrecht (7) described long-term prevention of hypertension development (and amelioration of end-organ damage) after transient antihypertensive treatment with a combination of guanethidine and dihydralazine in puberty and puberty, i.e., at the age of 3–11 wk. The BP of treated SHR remained normotensive for at least 14 wk after withdrawal of antihypertensive therapy and reached BP levels of untreated SHR 20–25 wk after the treatment had been discontinued (Fig. 4). A similar time course of delayed hypertension development was observed after antihypertensive therapy withdrawal in young SHR that were treated with either perindopril (263) or nitrendipine (600) in the same age period (Fig. 4). Albrecht's pioneer work was later extended in a remarkable series of Harrap's papers (260, 262, 263) that demonstrated the long-term benefit of short-term treatment of SHR with ACE inhibitors in the critical developmental period that can be localized between weeks 2 and 10 of age.

It is evident from Figure 4 that the attenuation of hypertension development after the withdrawal of early antihypertensive treatment does not depend solely on the functional elimination of the RAS. The long-term BP decrease seen in SHR during the first 3–4 mo after therapy withdrawal is most probably due to a reduction of vascular hypertrophy which, in untreated SHR, develops during the juvenile critical period (developmental window) un-
under the influence of a stepwise increase of transmural distending pressure. Blood pressure-dependent vascular hypertrophy can be attenuated in SHR either locally (i.e., distally to femoral artery constriction) (69, 199, 257) or in the whole animal kept under effective antihypertensive therapy. The existence of two distinctly different types of vascular hypertrophy or remodeling (BP dependent, ANG II independent vs. ANG II dependent, BP independent) (376) should be kept in mind when considering the long-term effects of transient RAS blockade (see sect. VI).

There is an important difference between drugs directly influencing RAS (ACE inhibitors or AT1 receptor antagonists) and other classes of antihypertensive agents (98, 280). After the withdrawal of transient treatment of young SHR with ACE inhibitors or AT1 receptor antagonists, BP rises to hypertensive levels but still remains significantly lower compared with untreated SHR (718). The resulting late BP reduction, which is characteristic solely for these two classes of antihypertensive drugs, can be found in SHR aged at least 25–30 wk, i.e., ~4 mo after this therapy had been discontinued. This persisting BP reduction, which represents only a minor part of the pronounced BP fall induced by active antihypertensive therapy, seems to be a late consequence of early transient elimination of the ANG II effects. On the contrary, the termination of antihypertensive therapy in young SHR with most other antihypertensive drugs is usually followed by a full recovery of high BP up to the levels seen in untreated SHR (98, 280).

A. Early Interventions on Sympathoadrenal System

There is no doubt that the development of hypertension in the rat can be prevented by impairment of the SNS at early stages of ontogeny, namely, in the suckling and weaning periods when SNS development is most intensive (639, 666, 776). The peripheral administration of 6-OH-dopamine to newborn SHR indeed attenuated hypertension development, whereas such treatment of adult animals had only transient BP-lowering effects (718). Early sympathectomy has provided convincing evidence that SNS plays a dominant role in the mechanisms responsible for the development of spontaneous hypertension in the rat. The most effective method for performing early sympathectomy in SHR is repeated injection of the antiserum against the NGF in the first postnatal week (125, 200) followed by guanethidine treatment up to the age of 4 wk (333, 424). This approach almost completely eliminates the function of SNS, but the residual catecholamine secretion from the adrenal medulla must be prevented by adrenal demedullation (420) or the effects of adrenal catecholamines must be abolished by α1-adrenoceptor blockade (374). Early sympathectomy not only elicits a BP decrease (dependent on the degree of SNS destruction) but also reduces cardiovascular hypertrophy. Hypertension and left ventricle hypertrophy were indeed absent in those sympathectomized SHR that were deprived of adrenal catecholamine effects (374, 420). Early sympathectomy had minimal effects on large conduit arteries of SHR but prevented hyperplastic changes of smooth muscle cells in the muscular arteries and normalized the diameter of small resistance vessels (424). Adrenal demedullation carried out in 4-wk-old sympathectomized SHR further increased the lumen diameter in muscular arteries but did not eliminate the strain differences in the number of smooth muscle cell layers, media cross-sectional area, or M/L (420). A combination of early sympathectomy with α1-blockade by prazosin lasting until puberty completely prevented cardiac hypertrophy and normalized hindquarter vascular resistance in SHR at least until the age of 35 wk (374).

The importance of SNS alterations in particular developmental periods for the pathogenesis of genetic hypertension can be evaluated in rats subjected to transient treatment with various drugs affecting SNS during early ontogeny. Thus a single injection of NGF antiserum administered to SHR aged 19–24 days markedly reduced their vascular sympathetic innervation and BP in adulthood without affecting vessel wall thickness (63). The chronic α-blockade by norgaline in the prenatal period was reported to lower the BP of adult SHR (494). Recently, a long-term BP reduction was found in adult SHR treated with α1-adrenoceptor antagonist terazosin during the first three postnatal weeks (472) when SNS maturation is accelerated (470). This is a typical example of the late consequence (15% BP reduction at 14 wk of age) induced by an early intervention on a particular regulatory system (SNS) that undergoes intensive maturation at a specific critical (suckling) period in which the respective intervention (α1-blockade) is applied. When this blockade has started in SHR beyond the critical period of rapid SNS maturation (at the age of 4 wk), BP development was not affected (335, 622).

In contrast to early α1-blockade, chronic β-blockade from conception until adulthood prevented neither hypertension development nor cardiovascular hypertrophy (425, 572). Moreover, lifetime β-blockade in SHR was associated with a substantial rise of systemic resistance (530, 599). On the other hand, the attenuated development of hypertension together with a reduced gradient of the resistance curve (reflecting wall-to-lumen ratio), diminished maximal pressor response, and especially lowered vascular resistance at maximal dilatation were demonstrated in SHR subjected to chronic β-blockade from youth until the age of 8–10 mo (731, 732). It seems that, in SHR, the late chronic β-blockade has much better antihypertensive effects than the same treatment applied in early ontogeny.
The severe impairment of the SNS during its most intensive phase of development (in the suckling and weaning period) substantially attenuates the development of genetic hypertension and accompanying cardiovascular hypertrophy. This is due to the absence of both the trophic influence and vasoconstrictor effects mediated by α1-adrenoceptors, whereas the elimination of β-adrenergic effects seems to be of minor importance.

B. Pharmacological Blockade of the RAS

The chronic inhibition of ANG II production by ACE inhibitors and/or chronic blockade of AT1 receptors interfere with important (perhaps primary) mechanisms controlling the structure and function of the cardiovascular system in genetically hypertensive rats. The long-term antihypertensive effects of these drugs in young SHR have been ascribed to multiple changes ranging from cardiovascular remodeling to altered kidney function. It should be kept in mind that the treatment of young animals, predisposed to genetic hypertension, with these antihypertensive drugs not only prevents the BP rise but also abolishes the development of cardiovascular hypertrophy due to eliminating both the growth-promoting effects of ANG II and the increased distending pressure. In fact, not only ANG II supplementation (52) but also a high salt intake or aldosterone infusion (261) administered during chronic treatment of immature SHR with ACE inhibitors can raise BP, enhance cardiac and vascular hypertrophy (proportional to BP elevation), and partially prevent the BP decrease persisting after drug withdrawal. Such two distinctly different mechanisms of vascular hypertrophy (BP dependent, ANG II independent vs. ANG II dependent, BP independent) have been discussed in detail by Korner and Bobik (376). The important role of pressure-dependent but ANG II-independent component of vascular hypertrophy (and systemic resistance) explains why greater BP reduction is achieved when antihypertensive therapy is introduced at an earlier age, and why the BP rise progresses at a slower rate after withdrawal of prolonged antihypertensive therapy. In contrast, the ANG II-dependent but pressure-independent component, which is gradually lost during the prolonged RAS blockade in immature SHR, might be related to the late effects of early transient RAS elimination.

1. Age-dependent response to RAS blockade and critical period for antihypertensive treatment

It is obvious that the actual and late effects of RAS blockade strongly depend on the age of genetically hypertensive rats. The treatment of immature SHR with ACE inhibitors (98, 207) or AT1 receptor antagonists (498, 717) prevents hypertension development, attenuates cardiac and aortic hypertrophy (559), and limits media thickening and/or lumen narrowing in resistance vessels (419). In addition, the BP of SHR treated with these drugs in prepuberty and/or puberty remained lower even after therapy withdrawal (2, 229, 262, 456, 538, 598, 749). The persistent reduction of mean arterial pressure resulting from the attenuated rise in peripheral vascular resistance (263, 598) is accompanied by a smaller amplitude of pulse pressure due to improved arterial compliance (207, 377, 598). The long-term decrease of systemic resistance persisting after drug withdrawal can partly be ascribed to a lower degree of wall hypertrophy in resistance vessels (98, 207, 498), decreased vascular sensitivity to α1-adrenoceptor agonists (88, 703), and sympathoinhibition (35). Some of these changes might be attributed to the improvement of ion transport alterations because short-term treatment with ACE inhibitors in prepuberty or puberty can correct abnormal cell Ca2+ regulation (704) and increased passive membrane permeability for Na+ (369), i.e., cellular alterations known to cosegregate with BP (51, 65, 66). The long-term restoration of baroreflex impairment (274, 751) and especially the leftward shift of the pressure-natriuresis curve (168) due to changes of both glomerular (359, 659) and medullary hemodynamics (40, 168) represent further possible mechanisms that are often considered responsible for the long-term BP reduction observed in SHR in which ANG II action had been impaired in early ontogeny (for details, see sect. viB2).

In contrast to the persistent cardiovascular effects seen after the withdrawal of ACE inhibitors or AT1 receptor antagonists in young SHR, none of the above effects was found after such treatment of mature SHR with fully established hypertension, i.e., in animals older than 5 mo. The antihypertensive therapy had lowered their BP, but its withdrawal was followed by a rapid BP return to the levels of untreated animals (94, 263, 539, 714, 715). Persistent BP reduction after withdrawal of long-lasting captopril treatment was confirmed by radiotelemetry in SHR treated since youth, whereas only a marginal BP decrease was observed in those treated from the age of 23 wk (377). Although chronic antihypertensive treatment of adult SHR with ACE inhibitors lowered the BP and reduced cardiac hypertrophy, it had no effect on the regression of structural abnormalities of mesenteric arteries (233).

The response of immature and adult SHR to ACE inhibitor treatment has also been directly compared in several studies. Perindopril treatment affected the hypertrophy of blood vessels more than that of the heart in young SHR, whereas the reverse was true in SHR treated in adulthood (276). Indeed, enalapril treatment ensured better normalization of the vascular amplifier in perfused hindquarters of SHR treated in youth compared with those treated only in adulthood (2). Similarly, AT1 receptor antagonists lowered both the minimal and maximal vascular resistance more in young than in adult SHR (538,
Moreover, the contractile response of mesenteric arteries to periaortimal nerve stimulation was reduced by chronic treatment of young SHR with AT1 receptor antagonists (227), whereas these changes were not found after the same treatment of adult SHR (226). All the abovementioned age-dependent differences were still evident for some time after drug withdrawal.

Only a few studies have explored the late cardiovascular consequences of ACE inhibition during intrauterine and early postnatal life. The antihypertensive treatment with an intermediate dose of captopril (50 mg·kg⁻¹·day⁻¹) before and during pregnancy did not influence the BP of SHR progeny in adulthood (76). A detailed study with a higher dose of captopril (100 mg·kg⁻¹·day⁻¹) indicated that prenatal treatment was partially effective in the prevention of spontaneous hypertension, but the effect was smaller compared with the similar treatment in prepuberty (751). Major permanent consequences (BP reduction, attenuation of cardiovascular hypertrophy, enhancement of baroreflex function, amelioration of endothelial impairment) were seen when captopril therapy started prenatally and continued at least until the eighth postnatal week (353, 593, 749, 751). On the other hand, altered renal development (often resulting in a substantial renal damage) has been reported when the treatment with ACE inhibitors (208, 244) or AT1 receptor antagonists (745) started in suckling rats.

Harrap and co-workers (260, 263) reported that transient perindopril treatment of SHR (between 2 and 10 wk of age) caused long-term normalization of renal hemodynamics, attenuation of heart hypertrophy, and a BP decrease that persisted up to the age of 25–80 wk. In contrast, similar perindopril treatment of adult SHR (aged 20–24 wk) lowered BP with a subsequent rapid BP return to high values after drug withdrawal (263). However, prolonged antihypertensive therapy (lasting at least 8–12 wk) and/or the use of very high doses of ACE inhibitors can induce moderate permanent BP reduction and partial structural normalization even in SHR aged 12–15 wk in which hypertension had already been developed (2, 95, 226, 422, 529, 724).

Figure 5 shows that long-lasting beneficial cardiovascular effects can be induced by transient treatment with ACE inhibitors or AT1 receptor antagonists before the age of ~20 wk. In contrast, the transient antihypertensive treatment of SHR older than 20 wk has no permanent cardiovascular effects. Spontaneously hypertensive rats are highly susceptible to this treatment during prepuberty and puberty, i.e., between the 4th and 10th wk of age when relatively short-term treatment can have pronounced long-term effects (2, 263, 498, 603). This is in good agreement with the “critical period for the development of spontaneous hypertension” described by Albrecth (7) (Fig. 4). In both cases, the slow delayed BP rise observed after the withdrawal of antihypertensive therapy probably reflects the late development of pressure-dependent vascular hypertrophy that already occurs beyond the juvenile critical period characterized by the rapid increase of systemic resistance.

Nevertheless, Table 2 indicates that only the combination of prenatal and postnatal treatment with ACE inhibitors or AT1 receptor antagonists prevents the development of genetic hypertension. Postnatal treatments, starting between 2 and 15 wk of age, successfully lower BP of SHR, and their BP effects tend to be proportional to the duration of treatment. The withdrawal of postnatal treatment is always followed by a moderate BP rise, but BP level of untreated SHR is never reached. On the other hand, if the antihypertensive treatment begins after 20 wk of age, BP is substantially decreased during the active treatment but returns to the levels of untreated SHR when this treatment is discontinued. However, no study on the effects of long-term treatment of older SHR with ACE inhibitors or AT1 receptor antagonists has yet been published.

It is thus possible that, in the case of transient RAS blockade, we are dealing with a combination of at least two independent phenomena. One of them might involve the long-term reduction of AT1 receptor density (750) induced by early antihypertensive treatment (88, 456, 749), as has been observed after the perinatal gene therapy based on the delivery of AT1 receptor antisense cDNA gene (223, 318, 452, 464). The specific critical period, during which RAS blockade or impairment could lower
AT₁ receptor density, is still unknown, but it apparently takes place in early ontogeny. The other phenomenon could be the attenuation of pressure-dependent vascular hypertrophy resulting from the absence of increased distending pressure. Such a secondary effect of RAS blockade and associated BP reduction is most pronounced at the age of 4–10 wk but can be traced up to the age of 20 wk when the developmental BP rise in SHR is completed (see also Figs. 2 and 3). This well-defined juvenile critical period for long-term attenuation of hypertension development by transient antihypertensive therapy seems to be common for several classes of antihypertensive drugs (7, 263, 600). It would therefore be useful to separate both phenomena, but this type of investigation can be complicated by the fact that effective perinatal interventions on RAS (as in the case of SNS) yield severe long-term functional impairment of this system, which is still present during the above-mentioned juvenile critical period for hypertension development.

It would be attractive to propose that the early changes in AT₁ receptor density (e.g., in the kidney) might be responsible for the persistent late BP reduction occurring after the transient RAS blockade. Actually, it would be desirable to distinguish the strictly ANG II-dependent cardiovascular changes from the pressure-dependent alterations. Perhaps a combination of low subantihypertensive doses of ACE inhibitors or AT₁ receptor antagonists with other type of effective antihypertensive therapy could reveal ANG II-dependent phenomena more convincingly than previous studies.

The SHR kidney is characterized by a shift of the pressure-natriuresis curve to the right that is already present in 5-wk-old rats (605). This can be corrected by

### Table 2. Influence of the onset and duration of antihypertensive therapy with ACE inhibitors or AT₁ receptor antagonists on blood pressure of SHR: treatment-induced BP fall and BP recovery after treatment withdrawal

<table>
<thead>
<tr>
<th>Therapy Onset</th>
<th>Duration of Antihypertensive Therapy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prenatal (since conception)</td>
<td>Short (4–5 wk)</td>
</tr>
<tr>
<td></td>
<td>Medium (8–12 wk)</td>
</tr>
<tr>
<td></td>
<td>Long (15–20 wk)</td>
</tr>
<tr>
<td>From 2 to 6 wk of age</td>
<td>Not available</td>
</tr>
<tr>
<td></td>
<td>–20%, –22%a</td>
</tr>
<tr>
<td></td>
<td>–20%, –26%b</td>
</tr>
<tr>
<td>From 13 to 15 wk of age</td>
<td>Not available</td>
</tr>
<tr>
<td></td>
<td>–30%, –10%f</td>
</tr>
<tr>
<td></td>
<td>–31%, –18%g</td>
</tr>
<tr>
<td>In rats older than 20 wk</td>
<td>Not available</td>
</tr>
<tr>
<td></td>
<td>–30%, –5%h</td>
</tr>
</tbody>
</table>

Data are given in percentages of blood pressure (BP) values found in age-matched untreated SHR. Not available indicates absence of adequate studies. References to corresponding studies are as follows: *749, 751; 88, 91, 156; 72, 40, 70, 71, 79, 175, 263, 498, 556a, 603; 72, 168, 207, 227, 229, 262, 263, 408, 538, 598, 603; 72, 98, 376, 377; 72, 95, 539, 724; 72, 426, 452, 529; 72, 263, 377, 493, 714, 715.

2. Possible mechanisms responsible for persistent BP decrease

The delayed slow BP rise as well as the late persistent BP reduction after the withdrawal of transient antihypertensive therapy with ACE inhibitors or AT₁ receptor antagonists may be due to the amelioration of several abnormalities characteristic for immature SHR. The available evidence points to structural and functional changes in the kidney, to abnormal baroreflex operation, and to altered reactivity of hypertrophied resistance vessels.

Chronic perindopril therapy of immature SHR normalized their renal functions by means of lowering renal vascular resistance, elevating renal blood flow, and increasing the glomerular filtration rate. These improvements persisted together with lowered BP for 12 wk after drug withdrawal (262). Such therapy increased the average lumen diameter of pregglomerular vessels and reduced renal vasoconstrictor responsiveness (41). The internal diameter of afferent (but not efferent) arterioles is increased by ACE inhibitor treatment of young SHR (359, 536), and this change persists even after drug withdrawal (656). The increase of afferent arteriole diameter in SHR treated with ACE inhibitors in youth results in the reduction of afferent arteriole resistance, whereas efferent arteriole resistance is not affected (173). Reduced lumen diameter accompanied by smaller cross-sectional area of the media indicate impaired growth of afferent arterioles during the development of spontaneous hypertension that can be corrected by chronic treatment with ACE inhibitors, inducing unexpected media growth and vascular remodeling of afferent arterioles in SHR (536).

The crucial importance of early renal hemodynamic changes for subsequent hypertension development is further supported by the effects of kidney cross-transplantation in SHR subjected to transient treatment with ACE inhibitors in the juvenile critical period. The transplantation of kidneys with high vascular resistance (from untreated SHR donors) into ACE inhibitor-pretreated SHR recipients abolished the characteristic long-term decrease of BP induced in SHR by early short-term antihypertensive treatment (264, 568). On the other hand, kidney cross-transplantation from SHR donors treated with enalapril at the age of 3–14 wk into untreated SHR recipients resulted in a pronounced BP fall, the part of which persisted for at least 16 wk after the transplantation (568). Nevertheless, the transplantation of kidney from SHR donors briefly treated with perindopril (at the age of 6–10 wk) into untreated SHR recipients resulted in a rapid BP rise and early death of animals (264). Further experiments are required to find the optimal age and drug dose for the pretreatment of kidney donors.
chronic treatment of immature SHR with ACE inhibitors (478) or \( \text{AT}_1 \) receptor antagonists (364) that shift arterial pressure-natriuresis/diuresis curves to the left because of a decrease of tubular reabsorption without major glomerular filtration rate or renal blood flow changes. Chronic (but not acute) therapy with the above drugs also elicits a leftward shift of the relationship between renal artery pressure and renal interstitial hydrostatic pressure (168, 364). This is the main difference between the late consequences of early transient therapy with ACE inhibitors and the effects of acute ACE inhibition in SHR kidneys (168). Because the link between renal artery pressure and renal interstitial hydrostatic pressure involves renal medullary blood flow, which is altered in SHR (607), persisting effects of early ACE inhibition are due to the reduced resistance of vessels upstream from the medullary circulation (169). This can be due to permanent changes of vascular structure or caused by the reduced formation of potent renal vasoconstrictor 20-HETE. Indeed, ACE inhibitors lower the renal P-4504A content and \( \omega \)-oxidation of arachidonic acid in the SHR kidney (496). Because 20HETE is involved in the control of renal vascular resistance and medullary blood flow in young SHR (307), its suppression may contribute to a resetting of the pressure-natriuresis relation. In fact, renal medullectomy abolishes the beneficial effects of early treatment with ACE inhibitors on renal functions in SHR and prevents persistent BP reduction after the withdrawal of ACE inhibitors (40).

Reduced baroreflex sensitivity in genetically hypertensive rats might be another target for early transient therapy of immature SHR with ACE inhibitors or \( \text{AT}_1 \) receptor antagonists. Enhanced baroreflex sensitivity is characteristic for 4- to 9-mo-old SHR subjected to ACE inhibition during the intrauterine period and early postnatal life in which the improved baroreflex control of the heart rate persisted up to 7 mo after drug withdrawal (751). The decreased density of central ANG II receptors in SHR subjected to early captopril treatment (750) might be responsible for the improvement of baroreceptor function because enhanced baroreflex control of the heart rate and sympathetic nerve activity in SHR treated with captopril throughout life (93) is related to decreased brain RAS activity (37). This also explains the smaller BP increase and reduced drinking response after intracerebroventricular ANG II administration in these animals (749). Furthermore, chronic intracerebroventricular administration of captopril to prepubertal SHR attenuated hypertension development, increased baroreflex sensitivity, blunted vascular reactivity to vasoconstrictors, and reduced sympathetic vasoconstrictor tone due to decreased central stimulation of sympathetic outflow (35, 36). Not only central but also peripheral administration of ACE inhibitors in prepuberty is capable of correcting baroreflex abnormalities in genetic hypertension. Thus chronic treatment with ACE inhibitors minimized the differences in baroreceptor control of heart rate between SHR and WKY (275), but the effect was greater if the therapy started in adult (14-wk-old) rather than in young (4-wk-old) animals (276). However, recent reports (303, 385) have indicated that the correction of baroreceptor abnormalities by chronic antihypertensive treatment of SHR is not specific for ACE inhibitors because baroreflex sensitivity could also be enhanced in younger SHR by other antihypertensive drugs. At present, it is difficult to define the exact contribution of the restored baroreflex sensitivity to persistent BP reduction found in SHR after the withdrawal of early treatment with ACE inhibitors, because some baroreflex improvement might simply follow BP reduction.

Prevention or amelioration of structural changes of resistance vessels (reduced media hypertrophy and/or increased lumen diameter) in SHR treated with ACE inhibitors (419) or \( \text{AT}_1 \) receptor antagonists (440) in prepuberty or puberty are an important part of the mechanisms responsible for the BP decrease during antihypertensive therapy and after its termination. Indeed, the reduction of media thickening and M/L, but not the increase of lumen diameter, in mesenteric resistance vessels accompanied a lowering of BP that persisted for more than 25 wk after the termination of fosinopril treatment in young SHR (603). Although vascular hypertrophy in young SHR can partially be attenuated even by low, nonhypotensive doses of ACE inhibitors (602, 644), persistent BP reduction after drug withdrawal is only observed in animals that were treated with relatively high therapeutic doses (88, 724). It is not surprising that the incomplete prevention of structural changes in immature SHR is associated with minimal modification of further BP development (53), because the influence of \( \text{AT}_1 \) receptor antagonists or ACE inhibitors on the structure of resistance arteries is dose dependent (440, 694) and proportional to the duration of treatment (498). Nevertheless, the degree of vascular structure control by this treatment cannot be taken as the only reliable predictor of persistent BP reduction after therapy withdrawal (88, 98, 498, 694).

Structural changes were demonstrated not only in small resistance vessels of SHR but also in large conduit arteries in which ACE inhibitors or \( \text{AT}_1 \) receptor antagonists (but not hydralazine) prevented arterial collagen deposition (4, 717). The collagen content in the aorta of young SHR is also diminished by chronic treatment with low nonhypotensive doses of quinapril which, however, substantially inhibited aortic ACE activity, indicating the importance of local RAS. This is in contrast to aortic smooth muscle hypertrophy that is pressure dependent because it is reduced by treatment with hydralazine or high quinapril doses (4). Angiotensin-converting enzyme inhibitors (but not hydralazine) selectively lower the nondistensible component of the vascular wall (collagen, basement membrane), whereas the distensible compo-
ient is reduced by both antihypertensive regimens (253). Indeed, the blockade of AT₁ receptors attenuates the expression of extracellular matrix proteins and transforming growth factor-β1 in blood vessels of SHR (356, 543). It has been suggested (32) that the prevention of aortic collagen accumulation in SHR treated with ACE inhibitors or AT₁ receptor antagonists is partially mediated by aldosterone inhibition. A parallel reduction of the collagen content and smooth muscle hypertrophy in SHR treated with ACE inhibitors increases arterial compliance (207, 432), which causes a reduction of pulse pressure. This might have further beneficial effects on the hypertrophy of resistance vessels, because pulse pressure has been suggested to be a periodic stretch stimulus for vascular smooth muscle growth (97). Thus the improvement of arterial mechanical properties might also contribute to the attenuation of resistance vessel hypertrophy in young SHR during antihypertensive therapy and after its termination.

There are several functional alterations in SHR arteries that are ameliorated by early transient treatment with AT₁ receptor antagonists or ACE inhibitors. Some of them (especially those concerning vascular responsiveness to catecholamines and/or sympathetic nerve stimulation) could also contribute to BP reduction after treatment withdrawal. For example, the increased sensitivity to α₁-adrenoceptor agonists can be decreased by ramipril treatment (485), and this change persists even after drug withdrawal (703). The greater contraction of SHR arteries after periarterial nerve stimulation is also attenuated during and after the treatment of young SHR with AT₁ receptor antagonists (227). The therapy with AT₁ receptor antagonists induces permanent inhibition of the facilitating effects of ANG II on vascular contraction elicited by periarterial nerve stimulation (226, 227). This persistent change might be due to chronic impairment of presynaptic AT₁ receptors that are known to facilitate NE release (336, 495). In fact, chronic treatment of SHR with ACE inhibitors or losartan (even in low doses) can abolish some abnormalities in cardiovascular noradrenergic transmission (57, 75, 396). Angiotensin II might also participate in abnormal maturation of the interaction between sympathetic innervation and vascular smooth muscle that is delayed in SHR as compared with WKY (144). The administration of AT₁ receptor antagonists reduced NGF levels in mesenteric arteries of immature SHR to the values of WKY (321). Similarly, perindopril treatment of SHR during the juvenile critical period induced a persistent decrease of renal NGF mRNA that was still detectable 10 wk after treatment withdrawal (79).

On the other hand, the correction of endothelial dysfunction by antihypertensive therapy of young SHR probably does not play a major role in BP reduction seen after drug withdrawal. The chronic treatment of SHR with high (but not low) doses of ACE inhibitors is capable of correcting impaired endothelium-dependent vasorelaxation to ACh (231, 602). Angiotensin-converting enzyme inhibitors ameliorate endothelial dysfunction in SHR aorta especially by increasing the production of EDRF (NO) rather than by inhibiting the synthesis of EDCF (PGH₂, TxA₂) (108, 604, 609). Moreover, ACE inhibitors also lower superoxide release (736). The recovery of endothelium-dependent vasorelaxation can be achieved not only by AT₁ receptor antagonists (160, 440) but also by Ca²⁺ channel blockers (440, 600). It seems that endothelial dysfunction, which appears in developing SHR later than vascular hypertrophy (601), results from hemodynamic alterations, and its correction by antihypertensive treatment is a consequence of BP reduction. Indeed, the improvement of endothelium-dependent vasorelaxation induced by ACE inhibitors lasts only a few weeks after therapy had been discontinued (175, 603).

A fundamental question concerns the molecular mechanism(s) by which chronic therapy of immature SHR with ACE inhibitors or AT₁ receptor antagonists can correct some of the above-mentioned phenotypic alterations. The reduction of AT₁ receptor density or the accumulation of kinins has been suggested as possible mechanisms mediating the long-term effects of early treatment with ACE inhibitors. Unfortunately, changes in the number of angiotensin receptors have usually been reported in animals treated pre- or perinatally, whereas the role of elevated kinin levels has often been studied in rats treated with ACE inhibitors in pubertal and pubertal stages.

Chronic administration of ACE inhibitors to SHR during early ontogeny seems to modify the number of angiotensin receptors. Thus prenatal captopril treatment lowers the density of ANG II receptors in the kidney of newborn and suckling SHR, whereas no significant changes occur in the WKY kidney (750). A similar observation was made in neurons from newborn SHR treated with captopril in utero (39). The importance of the reduced number of ANG II receptors after early treatment of SHR with ACE inhibitors is compatible with recent findings on perinatal gene therapy in SHR. It has been reported (318, 452, 575) that the early expression of AT₁ receptor antisense transcript resulted in persistent selective attenuation of cellular ANG II actions that leads to long-term attenuation of hypertension development until at least 7 mo of age.

It has been suggested that the accumulation of bradykinin is involved in mechanisms of BP reduction found in SHR after perindopril withdrawal (556a). However, the importance of elevated kinin levels for the subsequent BP decrease seen in SHR treated with ACE inhibitors in youth is not fully convincing, since AT₁ receptor antagonists induce similar late effects in young SHR as ACE inhibitors, although the late consequences of AT₁ receptor antagonists seem to be less pronounced (228, 456, 498,
In addition, chronic bradykinin B2 receptor blockade did not modify the antihypertensive and antihypertrophic action of ramipril treatment of young SHR, but it did eliminate the cardioprotective effects of ACE inhibitors (230, 232). Thus increased kinin levels might be responsible for some beneficial effects of chronic ACE inhibition such as the increase of cardiac capillary density (230), improved functional and metabolic status of the heart (232), reduction of renal NGF expression (79), elevation of aortic cGMP levels (231), and increased expression of vascular ANP type A receptors (768).

Altogether, the complex attenuation of multiple hypertensive mechanisms, which is achieved by transient antihypertensive treatment during the juvenile critical period, seems to be responsible for persistent BP reduction seen after drug withdrawal. The decrease of media hyper trophy and/or collagen accumulation as well as the diminished responsiveness of resistance vessels to sympathetic nerve activity may explain the delayed slow BP rise occurring after drug withdrawal in SHR treated with ACE inhibitors or AT1 receptor antagonists in the juvenile critical period. The long-term resetting of kidney function (namely glomerular and tubular circulation) together with changes in baroreflex operation might be important for the BP reduction persisting after drug withdrawal. The reduced density of AT1 receptors in kidney, brain, and/or blood vessels could be the probable molecular mechanism of the changes described in this chapter.

C. Early Transient Treatment With Other Antihypertensive Drugs

It also seemed of interest to ascertain whether transient treatment of SHR with other antihypertensive drugs (not lowering RAS efficiency) during the juvenile critical period could affect BP development after therapy withdrawal. In fact, there is surprisingly little information about BP changes after the termination of chronic therapy of genetically hypertensive rats with Ca2+ channel blockers. However, the onset of hypertension development was postponed in SHR treated with nitrendipine from the fourth to the eighth week of life compared with rats receiving this drug later, i.e., at the age of 8–12 wk (600). On the other hand, a persistent BP decrease has never been demonstrated after transient treatment of young SHR or Prague hypertensive rats (PHR) with Ca2+ channel blockers (98, 280, 600). Moreover, the antihypertensive treatment of young PHR with dihydropyridine Ca2+ antagonists did not prevent hypertension development in normotensive recipients of the cross-transplanted PHR kidney (281).

The short-term blockade of pressor V1A receptors with nonpeptide vasopressin antagonist in SHR aged 6–10 wk had almost the same influence on subsequent BP development as ramipril treatment in the same critical period (70), whereas the transient blockade of antidiuretic V2 receptors augmented the subsequent BP rise (518). Unfortunately, these experiments did not last long enough to demonstrate that BP reduction seen after the withdrawal of vasopressin V1A receptor antagonists persists at least until the age of 25–30 wk. The above findings (70, 518) are rather surprising because chronic administration of the combined V1/V2 peptide antagonist of vasopressin to SHR aged 4–14 wk attenuated hypertension development but did not ensure persistent BP reduction after treatment withdrawal (661), whereas the comparable treatment of SHR with selective V1 antagonist did not affect the BP rise (660).

On the contrary, chronic therapy of young SHR with vasodilators (e.g., hydralazine) partially ameliorated the alterations of resistance vessels (98, 328, 662), but their withdrawal was always followed by a rapid BP rise to levels of untreated SHR (98, 207, 229, 663). This might be related to the effects of the RAS and SNS that are enhanced by vasodilator therapy (653). The trophic effects of both stimulated systems might overcome the moderate benefit of hydralazine treatment. This drawback of vasodilators can be eliminated by their combination with drugs reducing SNS activity, e.g., guanethidine or reserpine (206, 730). Such combinations (e.g., hydralazine–reserpine–chlorothiazide) were capable of inducing certain regression of cardiovascular hypertrophy even in adult SHR with fully established hypertension (558, 727), although the beneficial effects of this antihypertensive treatment were much greater in young than in adult SHR (442). The combined antihypertensive therapy also delayed further hypertension development in SHR after drug withdrawal (7, 206).

The long-term attenuation of the development of genetic hypertension can also be achieved by transient treatment with antihypertensive drugs not influencing RAS, when such therapy is applied during the juvenile critical period. This means that the delayed slow BP rise can also be seen after the withdrawal of drugs other than ACE inhibitors or AT1 receptor antagonists. Certain indications have been obtained for Ca2+ channel blockers, vasopressin antagonists, and vasodilators combined with sympatholytics. Nevertheless, there is still no proof that these drugs could elicit late BP reduction persisting for more than 15 wk after therapy withdrawal.

VII. MODIFICATION OF GENETIC HYPERTENSION DEVELOPMENT BY NUTRITIONAL INTERVENTIONS IN CRITICAL PERIODS

The qualitative and quantitative changes in the nutrition may influence BP of genetically hypertensive rats.
The alterations of electrolyte intake (especially those concerning Na⁺, K⁺, and Ca²⁺ intake) have a major impact on BP development when given to immature SHR. On the other hand, the changes in nutrient intake affect the BP in both young and adult animals.

A. Postnatal Electrolyte Intake

Mutual interactions of dietary electrolytes in the modulation of BP development can best be illustrated on the example of salt sensitivity of young SHR, which is attenuated by elevated dietary intake of K⁺ (219, 625), Mg²⁺ (484), and Ca²⁺ (466, 548, 550), whereas a salt-induced BP rise is accelerated by Ca²⁺ restriction (519).

1. Na⁺ intake

A high sodium chloride intake accelerates hypertension development and aggravates its complications in many strains of genetically hypertensive rats including SHR. Although Oparil et al. (552) described multiple differences between salt-sensitive (Taconic Farm) and salt-resistant SHR (Charles River), most of the currently available SHR colonies are salt sensitive (54). The mechanisms of the salt-induced BP rise in young SHR have still not been entirely elucidated, although it is generally accepted that a high salt intake further enhances sympathetic hyperactivity in SHR (147, 198, 741).

Chronic salt loading of SHR inhibits NE release in the AHA that raises BP through diminished stimulation of depressor neurons (555). Indeed, baroreflex activation of NE release in AHA is blunted by a high-salt diet in SHR but not in WKY (565). An abnormal reciprocal relationship between BP elevation and reduction of NE release in AHA is found in young SHR (586). The pathogenetic mechanism of salt-sensitive hypertension in SHR also involves the abundance of the ANP in AHA of salt-loaded SHR (763) where it presynaptically inhibits NE release from nerve terminals. This is also associated with the upregulation of neuronal AT₁ receptors (554). However, treatment of SHR with captopril throughout life, which abolished the spontaneous BP rise (749), did not prevent salt-induced BP elevation in young SHR (753).

The evaluation of age-dependent susceptibility of SHR to the hypertensive effects of high salt intake is rather difficult because further elevation of their already high BP is often limited by counterregulating mechanisms. On the other hand, valuable information on age-dependent susceptibility of SHR to changes in salt intake was obtained in studies concerning dietary Na⁺ restriction. It is evident that the development of spontaneous hypertension can be attenuated if weanling or prepubertal SHR (aged 3–7 wk) are subjected to severe Na⁺ restriction (<20 mmol Na⁺/kg diet) (179, 236, 451). In contrast, less severe Na⁺ restriction (22–50 mmol Na⁺/kg diet) did not alter hypertension development in young SHR (12, 741).

The early onset of Na⁺ restriction is critical for its late BP effects in SHR (737, 738). Thus long-term severe Na⁺ restriction, which had already started in the first postnatal month, completely prevented hypertension development and eliminated the BP difference between adult WKY and SHR aged 16 wk. The same dietary regimen applied since the end of the weaning period (4th wk of age) or in prepuberty (7th wk of age) substantially attenuated hypertension development, but the BP of adult SHR remained elevated compared with similarly treated WKY. The attenuation of hypertension in immature SHR subjected to Na⁺ restriction was associated with a reduced BP response to ganglionic blockade. In contrast, severe Na⁺ restriction, which started from the age of 10 wk or later, had no significant effects on hypertension development or the BP response to hexamethonium (737, 738).

In all the above-mentioned studies describing attenuated development of hypertension, Na⁺ intake was below the level required for normal growth and development of rats (15–20 mmol Na⁺/kg diet) (220). As Na⁺ requirements decrease with age, the observed growth deficit was most pronounced in young rats and almost absent in mature animals in which the antihypertensive effect of Na⁺ restriction was no longer evident. The important interrelationship between body growth and hypertension development is underlined by the fact that BP in the rat appears to rise in response to growth acceleration (636). The augmentation of postnatal growth of rat pups by litter-size reduction during the late suckling period increased BP of adult male borderline hypertensive rats (513).

The BP-lowering effect of Na⁺ restriction was ascribed to reduced pressor effectiveness of the SNS (696, 738). Sodium restriction not only inhibited α₁-mediated pressor effects but also prevented the development of sympathetic hyperactivity in young SHR, whereas the same dietary regimen applied to adult SHR did not affect the above properties of SNS (426).

This is in agreement with the findings of Folkow and Ely (198) who postulated that the attenuation of the sympathetic response to stress is the main effect of the long-term reduction of salt intake (5–20 mmol Na⁺/kg diet) in young 5-wk-old SHR. One of the most critical elements concerned the reduction of NE release per nerve impulse. Smooth muscle sensitivity and dose-response curves to exogenous NE remained unchanged by the Na⁺ restriction, but the response of resistance vessels to nerve stimulation was depressed (527). Reduced neuronal NE release in young Na⁺-restricted SHR was also confirmed by others (236, 481, 482). Chronically decreased SNS efficacy was reflected by lowering of BP by ~15%, although other vasoconstrictor systems were activated (180). Dietary
Na\(^+\) restriction lowered NE release from electrically stimulated portal vein or the anterior hypothalamus irrespective of the animal’s age (482). However, it was the anterior hypothalamus of SHR where low salt intake abolished the facilitation of NE release by ANG II exclusively in younger age groups (481). Unfortunately, more detailed studies on SNS effects of severe Na\(^+\) restriction are still missing in adult SHR, the BP of which appears to be insensitive to such a dietary regimen.

In summary, salt sensitivity is decreasing with progressing age. Modifications of salt intake in immature rats can modulate further BP development mainly by influencing the SNS. The possible effects of perinatal salt loading or restriction on the BP of adult normotensive or genetically hypertensive rats represent another interesting aspect that is considered in section VIII.B.

2. K\(^+\) intake

Elevated K\(^+\) intake is known to counteract the hypertensive effects of excess sodium chloride intake in both rat and human (129, 211, 483). This is especially true for young SHR or SHRSP in which the high K\(^+\) intake attenuates salt-induced acceleration of hypertension development (146, 451, 625). The normalization of central sympathoinhibitory mechanisms, peripheral sympathetic nerve activity, and altered NE metabolism is involved in the antihypertensive effects of dietary K\(^+\) in salt-loaded animals (28, 146, 212, 235). Because a high-K\(^+\) diet dose-dependently protects young SHR against the salt-induced BP rise, the level of dietary K\(^+\) intake actually determines the degree of salt sensitivity in this rat strain (219).

A high K\(^+\) intake also lowers the BP of young (681) and adult SHR (451, 746) that were not subjected to concomitant salt loading. Furthermore, chronic dietary K\(^+\) supplementation improves the survival of hypertensive rats (129, 219, 483), although the protection against organ damage and prolongation of the survival can be achieved even without a major BP lowering (178, 697, 722). A marked reduction in stroke incidence, in arteriolar hypertrophy, and in the occurrence of hypertensive vascular lesions was found in young salt-loaded SHRSP fed a high-K\(^+\) diet from the age of 5 wk (697). The high K\(^+\) intake prevented the thickening of aortic intima and media in hypertensive rats (682) and diminished cholesterol ester deposition in the aorta of SHRSP fed a hypercholesterolemic, high-salt diet (699). Furthermore, endothelium-dependent ACh-induced vasorelaxation was normalized in young SHRSP by a high K\(^+\) intake (683), which also protects the endothelium of hypertensive animals against lipid peroxidation resulting from oxidative stress (310).

The antihypertensive and vascular protective effects of high K\(^+\) intake in hypertensive rats are obvious. Although their age dependence can be expected on the basis of the reciprocal role of Na\(^+\) and K\(^+\) in the pathogenesis of hypertension, most studies have been carried out in animals supplemented with K\(^+\) since weaning, whereas there are few data from adult rats.

3. Ca\(^{2+}\) intake

It is evident that the lowering of BP induced by dietary Ca\(^{2+}\) supplementation in genetically hypertensive rats depends closely on their age. High-Ca\(^{2+}\) diet feeding from the age of 4–6 wk attenuates hypertension development in SHR (17, 467, 632). With normal levels of sodium chloride in the diet, 3-wk-old SHR showed BP lowering within 4–5 days of dietary Ca\(^{2+}\) supplementation (268). When a high-Ca\(^{2+}\) diet was offered at 4 wk of age, at least 2 wk were required before a BP decrease was observed. More than 2 mo may pass without a clear-cut BP change when the same dietary intervention began at 6 wk of age (467). In contrast, dietary Ca\(^{2+}\) manipulation in SHR aged more than 10 wk did not influence BP (414, 676).

The experiments with cross-fostering (266) suggested that Ca\(^{2+}\) intake of SHR mothers during lactation (but not during gestation) is important for setting the BP of young SHR. Using SHR × WKY F\(_2\) hybrids, McCarty (468) demonstrated an inverse relationship between the Ca\(^{2+}\) content in maternal milk and BP levels of F\(_2\) mothers. The Ca\(^{2+}\) content in solid food consumed around weaning is also a decisive factor (505). High Ca\(^{2+}\) intake at this stage of development is associated not only with decreased blood pressure but also with reduced pressor responsiveness to \(\alpha_1\)-adrenoceptor agonists (269) and ANG II (415). No information is available about whether a transient short-term increase of Ca\(^{2+}\) intake in young SHR (aged 3–5 wk) can affect further BP development.

The age-dependent BP effects of high Ca\(^{2+}\) intake were also demonstrated in salt-sensitive Dahl rats fed a high-salt diet from either 4 or 12 wk of age (393). Blood pressure was reduced in young rats but elevated in adult animals fed a high-Ca\(^{2+}\), high-salt diet. This is in accordance with other studies on Dahl rats, in which a BP decrease was found in 4-wk-old salt-sensitive animals fed a 4% Ca\(^{2+}\) diet for 6 wk (571), whereas a BP rise was reported in 6-wk-old rats fed a 2% Ca\(^{2+}\) diet for the same period (570).

Augmented in vivo BP reactivity to exogenous NE belongs to the possible pathogenetic mechanisms responsible for BP elevation in SHR subjected to a low Ca\(^{2+}\) intake from the age of 3–6 wk (270, 339). In contrast, an increased Ca\(^{2+}\) intake was reported to induce greater pressor responses to NE and ANG II in SHR fed a high-Ca\(^{2+}\) diet in adulthood only (676). Thus the age-dependent difference in vascular reactivity to pressor agents might explain different BP effects of dietary Ca\(^{2+}\) manipulation in young and adult animals.

The BP-lowering effects of dietary Ca\(^{2+}\) become more prominent under conditions of high salt intake. The
high Ca^{2+} intake is then capable of preventing the BP rise occurring in 8-wk-old SHR within 2 wk of a high-salt diet (550). The increased hypothalamic NE turnover, which prevents secondary upregulation of α_{2}-adrenoceptors (762), corrects the noradrenergic activity of sympathoinhibitory neurons in the AHA. This sympatholytic effect, together with augmented natriuresis (550) and enhanced baroreflex sensitivity (549), has been proposed to mediate the antihypertensive action of a high-Ca^{2+} diet in young salt-loaded SHR.

Multiple mechanisms have been considered to be responsible for the antihypertensive effects of increased dietary Ca^{2+} intake in experimental hypertension (for review, see Ref. 267). One of them involves cell membrane stabilization and the correction of abnormal cell Ca^{2+} handling (161). Lymphocytes of SHRSP are characterized by an elevated intracellular free Ca^{2+} concentration ([Ca^{2+}]_{i}) and Ca^{2+}-activated K^{+} efflux through Ca^{2+}-dependent K^{+} channels (214). This intermediate phenotype cosegregates with BP in F_2 SHRSP × WKY hybrids (215). A high Ca^{2+} intake lowered [Ca^{2+}]_{i} and Ca^{2+}-stimulated K^{+} efflux in lymphocytes of adult SHRSP (216). Reduced [Ca^{2+}]_{i} levels were also found in platelets, lymphocytes, and vascular smooth muscle cells of SHR fed a high-Ca^{2+} diet from the age of 8 wk (13, 579, 752). A high Ca^{2+} intake also decreased platelet and lymphocyte [Ca^{2+}]_{i} in young salt-loaded SHRSP (556).

The mechanisms, by which high Ca^{2+} intake affects cell Ca^{2+} handling in young SHR, might be based on the changes in circulating levels of particular humoral factors and/or altered vascular smooth muscle responsiveness to them. A humoral factor influencing cell Ca^{2+} handling in cultured vascular smooth muscle cells was disclosed in the plasma of 8-wk-old SHR. The increased responsiveness to this factor was present even in smooth muscle cell cultures derived from 4-wk-old SHR in which no detectable circulating levels of this factor were found (684). High Ca^{2+} intake in young SHR lowered the augmented aortic responsiveness (measured as lanthanum-resistant Ca^{2+} uptake) to the hypertensive factor (459) that was isolated from erythrocytes and other tissues (747). This hypertensive factor stimulates vascular Ca^{2+} uptake (476), exhibits BAY K 8644-like contractile properties (297), and increases the voltage-dependent Ca^{2+} current (651).

The feeding of young SHR with a high-Ca^{2+} diet also decreased plasma levels of parathyroid hypertensive factor (PHF) (433). Parathyroid hypertensive factor, which opens L-type Ca^{2+} channels in vascular smooth muscle cells, elevates BP of normotensive rats, potentiates the action of other pressor agents, and augments vascular Ca^{2+} uptake. All these responses are relatively slow and culminate 45–60 min after PHF administration (for review, see Ref. 563). Plasma PHF levels are increased in rats with various forms of experimental hypertension (SHR, Dahl salt-sensitive and DOCA-salt hypertensive rats) as well as in essential hypertensive patients, namely, in those with low-renin, salt-sensitive hypertension (434). The role of PHF in the pathogenesis of spontaneous hypertension is further supported by a BP decrease seen in SHR after the blockade of circulating PHF with a specific antibody (34). Moreover, PHF secretion was entirely suppressed by feeding 4-wk-old SHR a high-Ca^{2+} diet for 8 wk, whereas low Ca^{2+} intake elevated plasma PHF levels in these animals (433).

The removal of parathyroid glands in weanling SHR or SHRSP indeed attenuated the expected age-related BP rise (217, 460, 631, 711). This was accompanied by a lowered aortic Ca^{2+} content and reduced BP response to NE administration (633). In contrast, no BP changes were observed if parathyroidectomy (PTx) had been carried out in adult or old animals, although PTx strongly diminished the incidence of hypertensive vascular lesions (217, 352, 428, 631). Early PTx also lowered BP and decreased cardiovascular reactivity in vivo to NE in Lyon hypertensive rats (567). The cross-transplantation of the parathyroid gland from genetically hypertensive animals into normotensive recipients caused a moderate BP elevation, irrespective of the age of donors and/or recipients. This was seen in SHRSP (525), SHR (562), as well as in Lyon and Milan hypertensive rats (566).

Parathyroid hormone (PTH) might also be involved in the participation of the parathyroid gland in the pathogenesis of genetic hypertension (630). Chronic PTH administration restored the development of spontaneous hypertension in parathyroidectomized rats (711) and prevented the BP-lowering effects of a high-Ca^{2+} diet in young SHR with intact parathyroid glands (362).

It can be concluded that dietary Ca^{2+} supplementation lowers BP and vascular reactivity especially in suckling, weanling, and prepubertal animals. Its antihypertensive effect seems to be related to the correction of cell Ca^{2+} handling that might reflect the changes in circulating humoral factors, namely, those originating in the parathyroid gland.

B. Dietary Nutrient Intake

The age-dependent influence of particular nutrients on the development of genetic hypertension is less understood than that of the above-mentioned changes in dietary electrolyte intake. At a given level of dietary electrolyte intake, the balance between dietary carbohydrate and fat intake can modify the BP even in rats with established hypertension, but a dietary protein intake preferentially affects the BP development in immature animals.

1. Caloric restriction and carbohydrate intake

The hypotensive effects of acute fasting or caloric restriction, which are mediated by the suppression of SNS
activity, are greater in SHR than in WKY irrespective of animal’s age (771). The results concerning BP effects of chronic caloric restriction in young SHR are conflicting. Severe caloric restriction, which had been started in weaning SHR aged 4 wk, increased their life span without affecting hypertension development (448). Moderate caloric restriction did not influence the BP rise in young 7-wk-old SHR, although their growth was retarded (237). Nevertheless, other studies revealed decreased BP of SHR subjected to caloric restriction since weaning (191, 748). A decrease of sympathetic tone participates in the mechanisms of BP reduction in young food-restricted SHR (557). Chronic caloric restriction is also capable of reducing BP of adult SHR (535, 594).

High carbohydrate intake, which increases SNS activity in the rat (769), elevates BP more in SHR than in WKY (293, 770). Sugar-induced hypertension in SHR seems to be related to augmented ANG II levels and/or insulin resistance (646). The prohypertensive effect of high sucrose intake in SHR was potentiated by a low protein intake (181) as well as by moderate elevation of salt intake (582). Unfortunately, there is still no direct comparison of BP effects induced by caloric restriction or high carbohydrate intake in immature and adult SHR.

2. Protein intake

Hypertension development and stroke incidence were attenuated in SHRSP fed a high-protein diet from the age of 6 wk, whereas a low-protein diet accelerated the BP rise and shortened the life span of these animals (760). The effects of dietary protein intake on BP levels were confirmed in young (189, 340) but not in adult SHR (719). The protein composition is very important because fish proteins exerted more potent BP effects than proteins from other sources (735, 760). Diets containing fish proteins are rich in several amino acids, such as methionine or lysine (293). One of the hypotheses ascribed the beneficial effects of high protein intake in young SHRSP to methionine deficiency in this strain. Indeed, the supplementation of weaning SHRSP with taurine, a putative metabolite of methionine, reduced the severity of hypertension without modifying their body growth (521). The prohypertensive and stroke-promoting effects of low protein intake in young SHR are rather surprising because it not only restricts body growth (332) but also limits cardiac hypertrophy (767) and renal injury (172, 189), i.e., it causes changes that usually accompany the attenuation of hypertension.

Actually, attention is focused on BP elevation observed in offspring of normotensive rat mothers that were subjected to a low protein intake during pregnancy (401, 405) (see sect. VIII). (401)

3. Cholesterol intake

The feeding of pubertal SHR or SHRSP (aged 60 days) with a hypercholesterolemic diet (containing 20% suet, 5% cholesterol, and 2% cholic acid) under the conditions of high salt intake resulted in hypercholesterolemia and a high incidence of arterial fat deposits (759, 761). Surprisingly, feeding these rats with the same diet from the age of 30 days almost completely prevented the BP rise and arterial fat deposition, although hypercholesterolemia was not diminished (734, 761). The antihypertensive effect of such a hypercholesterolemic diet in young hypertensive rats is still not clear, although an attempt was made to explain it, at least partially, by the limitation of body growth (734).

No other type of hypercholesterolemic diet has been reported to modify BP of genetically hypertensive rats, irrespective of whether such dietary regimens started at weaning (610, 612, 712), in puberty (73, 497), or in adulthood (540, 772). The lack of BP effects of hypercholesterolemic diets in adolescent SHR (73, 497, 610, 612) is rather unexpected because hypercholesterolemia in these animals was associated with further impairment of endothelium-dependent vasodilatation (73) and increased plasma aldosterone levels (612).

The developmental approach to the research of altered cholesterol metabolism in genetic hypertension would be highly desirable because cholesterol metabolism of adult animals strongly depends on the nutrition during early postnatal life (246, 308) (see also sect. III). In addition, cholesterol and triglyceride metabolism of adult rats might also be modulated by protein restriction of their mothers during pregnancy and/or lactation (453).

4. Fat intake

Numerous attempts have been made to modify hypertension development by changes in dietary fat intake and/or the proportion between saturated and polyunsaturated fatty acids (PUFA). Polyunsaturated fatty acids are known to influence SNS and prostanoid metabolism (288), and these changes might participate in their anti-hypertensive effects.

The deficiency of essential fatty acids was reported to elevate BP in the rat, especially when subjected to simultaneous salt loading (119, 171, 292). On the contrary, diets supplemented with various vegetable oils (291, 672) or fish oil (294, 295) were found to lower BP in SHR. Linoleic acid (18:2 n-6), a-linolenic acid (18:3 n-3), and particularly γ-linolenic acid (18:3 n-6) were identified as being responsible for the antihypertensive effects of diets supplemented with vegetable oils in SHR (145, 183, 648). Linoleic acid supplementation also protected rats against salt-dependent hypertension (351, 698).

Diets enriched with PUFA-containing vegetable oils mitigated the BP rise in SHR, especially when their moth-
ers had been fed such diets in the last week of pregnancy and during the first postnatal month (290–292). Later dietary interventions are less effective, but long-term feeding of these diets to weanling SHR usually attenuated hypertension development (289, 655). The age-dependent effect of PUFA on BP might be related to the early alterations in fatty acid pattern that can be found in the serum, liver, and kidney lipids of SHR before or during the manifestation of hypertension (656, 657). Some of these alterations are, at least partially, corrected by feeding SHR with PUFA-enriched diets (654, 658).

Diets rich in n-6 PUFA (vegetable oils) had greater antihypertensive effects in young salt-loaded borderline hypertensive rats than diets containing n-3 PUFA (fish oil) (489), whereas the reverse was true for the lipid-lowering effects of these diets in young SHR (89). The diets supplemented with fish oil (rich in eicosapentaenoic acid 20:5 n-3 and docosahexaenoic acid 22:6 n-3) attenuated hypertension development in SHR but had no BP effects in adult rats with established hypertension (479). On the other hand, this diet slightly suppressed hypertension development in young SHRSP (284) and moderately reduced BP in adult animals of this strain (295). The mechanisms of its antihypertensive action involve the decreased production of vasoconstrictor thromboxane A2 (765), attenuated vascular contractile responses to NE or sympathetic nerve stimulation (99, 278, 664), improvement of endothelium-dependent vasorelaxation (506), and reduced Ca2+–stimulated K+ efflux in vascular smooth muscle (665). The antihypertensive effects of dietary fish oil were augmented by concomitant Na+ restriction in both young and adult SHRSP rats (294, 295), whereas the feeding of adult salt-loaded SHR with a fish oil-supplemented diet even increased BP (112).

Further studies are necessary to elucidate the possible age-dependent differences in the antihypertensive action and/or mechanisms of particular PUFA. Moreover, the feeding of pregnant rats with diets high in saturated fatty acids and/or low in linoleic acid may also cause certain intrauterine “programming” of hypertension development in the rat (402), whereas the restriction of maternal protein intake prevents the BP effects of dietary fat intake in weanling rats (406).

A. Embryonic Environment

Barker (26, 27) proposed that reduced human fetal growth due to altered transplacental nutrition (evidenced by placental enlargement) is associated with an increased risk of adult cardiovascular diseases including hypertension, ischemic heart disease, and syndrome X. Disproportionate fetal and placental growth was also found in SHR in which fetal growth is impaired (177, 184, 334, 391, 435) and placental weight is increased at term (184, 334, 435). However, decreased weight of the placenta was detected in SHR during earlier stages of gestation, and this change precedes the reduction of fetal weight (184). An enlarged volume of amniotic fluid together with the decreased amniotic K+ concentration are further abnormalities of SHR fetuses (184). Prenatal hypertension demonstrated in SHR fetuses at 20 days (but not at 18 days) of gestation is associated with a substantial decrease of uteroplacental blood flow (19). Low placental perfusion in pregnant SHR at term is preceded by the reduction of uterine blood flow at mid-gestation (354). Reduction of uterine and placental blood flow in pregnant SHR indicates that vascular resistance in the uteroplacental unit of hypertensive mothers is elevated (209). Low placental perfusion, which leads to altered oxygen and/or nutrient supply, might interfere with body growth and the development of the cardiovascular apparatus in SHR fetuses.

With the exception of a single study (20), cross-transplantation of SHR embryos between hypertensive and normotensive mothers does not influence future BP development (242, 492). The same was found in salt-sensitive Dahl rats (133, 384). The main problem of these studies is that the cross-transplanted “hypertensive fetus” still has an important part of its original intrauterine environment because the development of both placenta and amnion is determined by the genotype of the fetus. It would therefore be interesting to know whether the above-mentioned placental and amniotic abnormalities of SHR fetuses would be ameliorated after their transfer into the uterine environment of WKY mothers. The possible persistence of such changes would explain the failure to affect BP development in cross-transplanted SHR. This would indicate whether fetal abnormalities are indeed more important for the development of genetic hypertension than maternal alterations.

B. Maternal Electrolyte Intake

Perinatal salt loading or restriction exerts considerable effects on BP development in normotensive and genetically hypertensive rats. With the exception of two studies (134, 510), high salt intake during pregnancy and lactation in normotensive (114, 115, 273), Dahl salt-sensitive (475), borderline hypertensive (302), or spontane-
ously hypertensive mothers (116, 150, 153, 347) is always associated with elevated BP of their progeny later in life. Elevated salt intake in the postweaning period usually facilitates the manifestation of BP increase induced by perinatal salt loading (114, 115, 273, 302, 475). On the other hand, severe Na\(^+\) restriction (17 mmol Na\(^+\)/kg diet) during intrauterine life and suckling period impairs body growth of SHR and lowers their BP in adulthood (150), but less severe Na\(^+\) restriction (20–22 mmol Na\(^+\)/kg diet) fails to modify body weight and BP in the offspring of normotensive Sprague-Dawley (114) or SHR mothers (690).

It is still not clear how the changes in Na\(^+\) intake of the rat mother can affect the fetus or the pup. There were no changes of Na\(^+\) concentration in the plasma of rat fetuses or newborns, but Na\(^+\) concentration was increased in the amniotic fluid when mothers were fed a salt-enriched diet (114, 273). Higher Na\(^+\) and lower K\(^+\) concentrations were reported in the milk of SHR (18, 151, 474) or Dahl salt-sensitive dams (134), but dietary electrolyte manipulations did not alter milk electrolyte intake by suckling rats (18, 134, 273). Perinatal NaCl exposure might alter the sympathetic control of the circulation (164). Increased pressor response to ANG II and reduced depressor action of isoproterenol were demonstrated in adult rats exposed to high salt intake in early ontogeny (114). Further studies of both immature and mature animals influenced by variations of salt intake in the early developmental stages should clarify the pathophysiological mechanisms mediating the late effects of perinatal NaCl exposure.

### C. Maternal Protein Intake

Chronic undernutrition of normotensive rat mothers during gestation delays postnatal growth and elevates BP of their offspring (744). Prenatal protein restriction impairs glucose tolerance (400, 569), alters lipid metabolism (453, 569), and elevates the BP of adult rats (401, 569). The level of protein intake in pregnant rats is inversely proportional to BP of their adult offspring (401). Long-term BP elevation resulting from low maternal protein intake is already observed at weaning, i.e., at the age of 4 wk (412). Blood pressure increase induced by prenatal protein malnutrition is not enhanced by a high salt intake (410) or by diets supplemented with saturated fatty acids (406). There is still no information about the effects of maternal protein intake restriction on the BP of SHR offspring.

Hypertension elicited by the restriction of maternal protein intake is sensitive to captopril treatment in both the weaning period and adulthood (409, 645). Its maintenance in postnatal life is dependent on adrenal corticosteroid secretion (221). Adrenalectomy carried out at the age of 6 wk lowered high BP in rats subjected to prenatal protein intake restriction, while corticosterone replacement restored their hypertensive state. No effect of adrenalectomy on BP was observed in the offspring of dams fed a diet with a normal protein content (221).

Restriction of maternal protein intake induces placental enlargement and disproportionate retardation of fetal growth (408). The enlarged placenta is characterized by decreased 11\(\beta\)-HSD activity (411). Reduced placental protection of the fetus against maternal corticosteroids (31) seems to play a key role in the pathogenesis of hypertension induced by maternal protein intake restriction. Indeed, maternal adrenalectomy or the blockade of maternal glucocorticoid synthesis by metyrapone (403, 405) prevent the development of this type of hypertension. Altered function of hypothalamo-pituitary-adrenal axis and/or increased number of glucocorticoid type II receptors (e.g., in hippocampus or aorta) (407) suggest that these mechanisms are involved in intrauterine programming of adult BP that becomes elevated as a consequence of reduced maternal protein intake.

A moderate BP increase can also be induced when restriction of maternal protein intake is introduced in discrete periods of gestation only (413). The importance of early abnormal placental development in the pathogenesis of this type of hypertension is supported by the fact that low maternal protein intake in the first or second trimester also influences BP of the offspring (413). Consequently, persistent placental 11\(\beta\)-HSD dysfunction permits deleterious effects of maternal corticosteroids in the last trimester of gestation that seems to be the critical period for intrauterine hypertension programming (431).

### D. Placental Glucocorticoid Metabolism

Placental type II isoform of 11\(\beta\)-HSD, which catalyzes the conversion of maternal corticosterone and cortisol to inert 11-keto products, normally protects the fetus from excess maternal glucocorticoids. Insufficient function of this placental enzyme is associated with altered fetoplacental growth (low birth weight and increased placental size) and determines the programming of later hypertension development that appears as a delayed consequence of a deficient placental barrier to maternal glucocorticoids (640). In the rat, placental 11\(\beta\)-HSD activity is directly proportional to fetal weight but inversely related to placental weight (31). Fetal exposure of rats to exogenous glucocorticoids (dexamethasone), which are poorly metabolized by 11\(\beta\)-HSD, indeed lowered the birth weight and initiated hypertension development (31). In addition, the inhibition of placental 11\(\beta\)-HSD activity by carbenoxolone treatment of pregnant rats, which increased fetal exposure to maternal glucocorticoids, reduced birth weight and elevated BP in their adult offspring (404, 446). Both dexamethasone and carbenox-
alone treatments were highly effective when introduced in the last trimester of gestation (404, 431). The exposure of fetal rats in utero to glucocorticoid excess may initiate hypertension development by alterations of glucocorticoid-regulated gene expression occurring during ontogeny of the SNS, adrenergic receptor expression, or intracellular signaling processes (44, 45, 300).

E. Pup Nutrition and Maternal Care

Maternal environment during the suckling and weaning periods plays an important role in the development of the cardiovascular phenotype of rats with genetic hypertension. Significant BP reduction was observed in adult SHR or salt-sensitive Dahl (SS/Jr) rats if they were reared by normotensive foster mothers from birth to weaning (102, 103, 149, 471, 508). On the contrary, if SHR and SS/Jr litters were reciprocally cross-fostered, the high BP of adult animals was not affected (507). Nevertheless, the original finding of elevated BP in Sprague-Dawley rats reared by SHR foster mothers (480) was never confirmed in the later studies (20, 102, 103, 149). Thus the full development of high BP in the two strains of genetically hypertensive rats requires both an inherited susceptibility to hypertension and a hypertensive maternal environment. This is also true for the exaggerated BP response of borderline hypertensive rats to acute stress, which can also be attenuated by cross-fostering with WKY dams (624).

This was demonstrated by cross-fostering of SHR pups to WKY mothers at various stages of early postnatal development (469). The maternal influence on the development of high BP in SHR is limited to the first 2 wk of postnatal life, i.e., to the suckling period. Later contact of prehypertensive pups with the normotensive mother had no significant influence on the BP of adult SHR. The BP-lowering effect of cross-fostering in SHR is partly mediated by a dampening of the sympathetic-adrenal medullary contribution to BP maintenance (102). This is in accordance with the rapid development of the SNS just in the first two postnatal weeks (470), which represent a critical developmental period when particular stimuli related to mother-pup interaction can modify the developing SNS.

During the suckling period, rat mothers can affect hypertension development in their pups either by nutritional factors (milk electrolytes, quality and/or quantity of particular nutrients, biologically active agents) or through mother-pup behavioral interactions (grooming, nest attendance, nursing frequency). The latter possibility is further supported by an earlier observation that daily handling of SHR pups during the preweaning period results in attenuated hypertension development (692), indicating that adult BP can be altered by a modification of early environmental stimuli.

The milk of SHR mothers contains more Na⁺ and Cl⁻ but less K⁺ and Ca²⁺ than the milk of WKY mothers (18, 152, 474). Milk from SHR has a lower protein content, a similar fat content, but an altered fatty acid composition (474, 488). Mother-to-pup milk transfer is more effective in WKY than in SHR dams, irrespective of whether they rear their own pups or pups from a different strain. This difference was most evident 7 days after birth and disappeared later (608). It seems that WKY mothers transfer greater volumes of milk over shorter nursing intervals than SHR mothers because SHR mothers assume nursing postures more often than WKY mothers (104, 105, 512). Altered nutrition associated with changes in body growth during the suckling period might be a possible explanation for the observed differences in adult BP, although this issue is not yet clear (469, 513, 515).

Another mechanism, whereby milk delivery may influence cardiovascular development, seems to be related to the BP increase occurring in pups in response to maternal milk ingestion (514). The above hemodynamic differences, which represent exaggerated vasoconstriction mediated by SNS activation in suckling SHR, can be abolished by ganglionic blockade (626). This means that quantitatively different SHR maternal behavior (more frequent nursing, longer mother-pup contact) might affect further BP development in SHR pups by their repeated exposures to transient periods of BP elevation, whereas this deleterious factor is minimized when SHR pups are cross-fostered by WKY dams (515). It seems interesting that the maternal behavior of normotensive Sprague-Dawley dams can be altered by a high salt intake, and the resulting more intensive maternal care might be responsible (through the above pathogenetic mechanisms) for BP elevation observed in their offspring in adulthood (115).

It is evident that adult BP is a classical example of a trait that is a product of gene-environment interaction. Genetic factors are partially responsible for the differences in maternal behavioral pattern, which also partially depends on the behavioral phenotype of pups (104, 511). Thus genetically predetermined characteristics of normotensive or hypertensive pups can modify the specific types of mother-infant interactions that affect the rate of BP rise during postnatal development (515).

IX. DEVELOPMENTAL ASPECTS OF SALT-INDUCED HYPERTENSION

The induction of salt hypertension, which is an age-dependent process, represents another tool for the study of the role of critical periods (developmental windows) in the pathogenesis of hypertension in the rat. The remark-
able analogy of numerous aspects in the development of salt-induced and spontaneous genetic hypertension seems to indicate that similar ontogenetic principles govern the progress of hypertensive process in immature rats with both forms of experimental hypertension. The critical period for inducing salt-dependent hypertension in the rat (790) coincides not only with the phase of rapid body growth and associated BP rise (636) but also with the developmental window in which long-term attenuation of spontaneous hypertension development can be achieved by transient antihypertensive treatment (7, 263, 600). This age period is characterized by intensive development of pressure-induced vascular hypertrophy, which may be the common denominator for the rapid development of both forms of experimental hypertension at the age of 4–10 wk. Furthermore, a comparison of salt-induced and genetic hypertension in young animals could also reveal some aspects of the adaptation of the immature organism to long-term BP elevation.

**A. Age-Dependent Susceptibility to Salt-Dependent Hypertension**

The BP response to many hypertensive stimuli depends on the developmental stage at which they influence the organism. Weanling and prepubertal rats are more susceptible to various forms of experimental salt hypertension than adult animals (for review, see Ref. 790). Under the conditions of a high salt intake, this has been demonstrated in rats with adrenal regeneration (64), rats treated with DOCA (509) or triiodothyronine (739), Dahl salt-sensitive rats (128), and rats with renal damage due to renal mass reduction (388) or transient renal ischemia (389). The magnitude of the hypertensive response progressively diminishes with advancing age from which hypertensive stimuli begin to influence the organism (64, 154). Sodium seems to be the ion responsible for the age-dependent hypertensive effects of excess sodium chloride intake, because chronic dietary NaHCO₃ loading elicits hypertension only in young but not in adult deoxycorticosterone-treated rats (789).

Salt intake level in youth and adolescence is a decisive factor for the susceptibility of adult rats to salt hypertension (381). Blood pressure response to renal mass reduction was enhanced in those adult rats that had been drinking saline since prepuberty (155). On the other hand, moderate salt deprivation (0.4% NaCl diet plus thiazide diuretics) of young Dahl salt-sensitive rats did not abolish the susceptibility of adult animals to the hypertensive effects of a high-salt diet (313). However, these rats were not protected against the early effects of sodium chloride intake in the most sensitive period because salt deprivation had not been introduced until 3 wk after weaning. In fact, the effects of early severe Na⁺ restriction (<20 nmol Na⁺/kg diet) on the subsequent salt susceptibility of Dahl rats have still not been evaluated.

Irrespective of the degree of genetic predisposition to salt hypertension, the important BP effects of elevated salt intake in the postweaning period contrast with a much lesser influence of high salt intake of rat mothers during pregnancy or lactation on later BP development in their offspring (134, 273). This supports the assumption that prepuberty might be a critical period for the induction of experimental salt-dependent hypertension in the rat (322). This is particularly true for severe, self-sustaining forms of hypertension, because mild to moderate salt hypertension can be induced even in adult animals (for review, see Ref. 790). Nevertheless, salt susceptibility decreases with progressing age of animals so that the BP response of 6-mo-old salt-sensitive rats to a high salt intake was substantially attenuated in the outbred Dahl-Brookhaven strain (128) and almost absent in the inbred Dahl-Rapp strain (786).

It is evident that variations of salt intake in the developing organism can induce numerous long-term alterations of circulatory homeostasis including hypertension (16, 62, 156, 381, 499). Such permanent modifications result from the interaction between this environmental factor and active developmental processes. This is the reason why the sensitivity to various factors (including hypertensive stimuli) is often modified when the organism proceeds to the next developmental period and its relationship to the environment undergoes characteristic maturation changes (379, 380).

**B. Pathogenesis of Salt-Dependent Hypertension in Young and Adult Rats**

Several mechanisms have been proposed to contribute to the enhanced hypertensive response of young salt-loaded animals (325, 381, 790). There are multiple differences in body fluid regulation, renal functions, ion transport, hemodynamics, participation of particular vascular systems, baroreflex efficiency, arterial wall properties, and organ damage between rats in which salt hypertension was elicited in youth or in adulthood.

The expansion of plasma and blood volume as well as of total body water was greater in prepubertal DOCA salt-treated rats than in those subjected to this treatment in adulthood (323). Plasma volume was also increased in young salt hypertensive rats with reduced renal mass but not in adult animals, although extracellular fluid volume was expanded in both age groups (388). A similar age-dependent pattern of extracellular fluid distribution was also observed in Brattleboro rats with DOCA salt hypertension (792) and in Dahl salt-sensitive rats (157).

The augmented volume expansion in young salt hypertensive rats can be related to a greater degree of
damage to the immature kidney (326, 344). The inactivation of renal RAS in DOCA salt hypertensive rats is accompanied by decreased resistance in afferent arterioles that leads to increased glomerular capillary pressure causing glomerular damage (174). These alterations are more severe in immature than in adult kidneys (325) because renal renin activity is suppressed by the high salt intake to a greater extent in young than in adult rats (324). The same age difference was also seen in animals with DOCA salt or adrenal regeneration hypertension (345, 577).

The intravascular expansion in young salt-loaded rats might enhance the release of humoral inhibitor(s) of the Na\(^{+}\)-K\(^{+}\) pump in immature animals (392, 790) (see below). Suppressed Na\(^{+}\)-K\(^{+}\) pump activity and increased cell Na\(^{+}\) content were reported in erythrocytes of volume-expanded rats in which hypertension was induced by renal mass reduction and saline drinking (132). However, red cell Na\(^{+}\) content and ouabain-sensitive (OS) Na\(^{+}\) extrusion were unaltered in young and adult subtotally nephrectomized rats fed a high-salt diet, although the young group became hypertensive (788). “Sodium pump inhibition” was suggested by a moderate decrease in OS Rb\(^{+}\) uptake. This was, however, due to a reduction in the exchange of external Rb\(^{+}\) for internal K\(^{+}\) (OS 1Rb\(^{+}\)/1K\(^{+}\) exchange), which has no net transport effect, because it represents one of the reversible partial reactions exerted by the Na\(^{+}\)-K\(^{+}\) pump. Such an alteration has no relationship to hypertension and was also present in erythrocytes of adult subtotally nephrectomized rats that remained normotensive under the conditions of a high salt intake (788).

A detailed analysis of the kinetics of ouabain-sensitive red cell ion transport (786) indicated that the reduced maximal transport rate of the Na\(^{+}\)-K\(^{+}\) pump was combined with an increased affinity for internal Na\(^{+}\) in young salt hypertensive Dahl rats, whereas the reverse was true in erythrocytes of 6-mo-old animals (783). Consequently, red cell ion transport, which is measured at physiological cell Na\(^{+}\) concentrations, was indeed accelerated in young salt-sensitive Dahl rats with severe salt hypertension, whereas their microsomal Na\(^{+}\)-K\(^{+}\)-ATPase activity in renal homogenates (determined at saturating Na\(^{+}\) concentrations) was reduced (784). In contrast, elevated renal Na\(^{+}\)-K\(^{+}\)-ATPase activity and unaltered red cell transport were observed in adult salt-sensitive Dahl rats with a less pronounced BP response to high salt intake (783). Nevertheless, the above age-dependent alterations of Na\(^{+}\)-K\(^{+}\) pump activity do not seem to be directly related to the salt hypertension elicited in immature rats.

On the contrary, age-dependent salt hypertension might be associated with the alterations of passive membrane permeability for monovalent cations, namely, with enhanced Na\(^{+}\) and Rb\(^{+}\) (K\(^{+}\)) leaks that are defined as residual fluxes resistant to the inhibition by ouabain plus loop diuretics (furosemide or bumetanide). The high Na\(^{+}\) leak is the principal red cell ion transport abnormality not only in genetically hypertensive rats (for review, see Ref. 782) but also in salt-sensitive Dahl rats (786). Increased Na\(^{+}\) leak in erythrocytes of salt-sensitive Dahl rats is responsible for the elevated red cell Na\(^{+}\) content. Compared with adult animals, erythrocytes of young Dahl rats are characterized by augmented Na\(^{+}\) leak, and its magnitude is further increased by a high salt intake in salt-sensitive but not in salt-resistant rats (783, 786). On the other hand, high Rb\(^{+}\) leak was suggested to be a potential marker of salt sensitivity in the rat (785). It is increased in erythrocytes of salt-sensitive strains (Dahl, Sabra, or SHR) compared with corresponding salt-resistant controls (785, 786), whereas it is unaltered in Prague hereditary hypertriglyceridemic rats (386), the hypertension of which is not affected by high salt intake (unpublished data). In contrast to Na\(^{+}\) leak, enhanced Rb\(^{+}\) leak in erythrocytes of salt-sensitive Dahl rats is completely independent of their age or salt intake (786).

Increased salt intake also exerts age-dependent hemodynamic effects. The pronounced hypertensive response of young rats to high salt intake or DOCA salt treatment is due to a greater rise in systemic resistance compared with adult animals subjected to these hypertensive stimuli (787, 791). Both structural changes of the vascular bed and abnormal vascular tone due to different activity of various vasoactive systems and/or altered vascular responsiveness should be considered. Minimal vascular resistance is elevated in young but not in adult salt hypertensive Dahl rats (J. Nedvı́dek and J. Zicha, unpublished data). As far as the participation of individual pressor systems in the maintenance of elevated BP in salt-dependent hypertension is concerned, SNS and digoxin-like factor(s) prevail in young rats, whereas the contribution of ANG II and vasopressin is enhanced in adult animals with these forms of experimental hypertension.

Acute ACE blockade with captopril moderately reduced BP in adult DOCA salt hypertensive rats, but no significant BP changes were found in young hypertensive animals (791). Similarly, the captopril-induced BP decrease was more pronounced in adult than in young Dahl salt-sensitive rats (Nedvı́dek and Zicha, unpublished data). This conforms with the greater suppression of RAS activity in young than in adult rats subjected to chronic salt loading (324, 345, 577). Thepressor effects of vasopressin are also more important for BP maintenance in adult rats than in young DOCA salt hypertensive rats (791, 792). Moreover, vasopressin seems to be essential for the induction of DOCA salt hypertension in adult but not in young rats (792).

On the contrary, \(\alpha_1\)-adrenergic blockade by prazosin indicated greater involvement of the SNS in young than in adult DOCA salt hypertensive rats (791), and the same
was true for salt hypertensive Dahl rats (Nedvidek and Zicha, unpublished data). Furthermore, neonatal sympathectomy (induced by repeated subcutaneous 6-hydroxydopamine injections in the first two postnatal weeks) attenuated the development of DOCA salt hypertension in young rats (194) but enhanced it in adult animals (585). However, neonatal guanethidine-induced peripheral sympathectomy lowered the BP in both young and adult salt hypertensive Brattleboro rats, but it did not abolish the age-dependent BP difference (382). The prevention of age-dependent salt hypertension by treating young rats with the selective noradrenergic neurotoxin DSP-4 (671) indicated the importance of central noradrenergic mechanisms in prepuberty, which is the critical developmental period for the expression of enhanced susceptibility of young rats to salt hypertension.

The participation of endogenous “Na$^+$ pump inhibitors” (digoxin-like factors) in the maintenance of salt hypertension is also age dependent. These factors might influence sympathetic neurotransmission and/or vascular smooth muscle responsiveness to sympathetic nerve activity (24, 708). The administration of antidigoxin serum (ADS) lowered BP in young DOCA salt hypertensive rats, but it did not influence BP in those rats in which DOCA salt hypertension was elicited in adulthood (370, 794). This qualitative age-dependent difference in the BP response to ADS injection was seen even if BP-matched subgroups of young and adult DOCA salt hypertensive rats were compared (392). The acute blockade of digoxin-like factors decreased the systemic resistance in young but not in adult DOCA salt hypertensive rats (793).

Another age-dependent abnormality of BP regulation in salt hypertension is the more pronounced impairment of baroreflex efficiency that was disclosed in young salt hypertensive Dahl rats compared with the adult ones (523). The extent to which reduced arterial compliance might contribute to a greater baroreflex dysfunction in young salt hypertensive animals is still open for discussion. Decreased arterial compliance was found in rats subjected to a high salt intake or DOCA salt treatment since prepuberty, whereas no changes of arterial compliance were observed if the same hypertensive stimuli were applied in adulthood (787, 791). These findings explain why arterial compliance was reported to be decreased in younger and increased in older rats with DOCA salt hypertension (120, 121). The changes of arterial compliance in young salt-loaded rats might be elicited by a Cl$^-$ rather than a Na$^+$ overload, because they were absent in young DOCA hypertensive rats that were fed a diet containing equimolar amounts of sodium bicarbonate instead of sodium chloride (780). Reduced arterial compliance augments pulse pressure and thus further increases the systolic pressure of young salt hypertensive rats (787).

We suggest that the rapid maturation of the arterial wall properties at prepuberty and puberty might be responsible for the enhanced reactivity of young rats to long-term BP manipulations. Both the pronounced pressure-dependent hypertrophy of resistance vessels and the major reduction of arterial compliance are associated with hypertension development in this age period. It should be kept in mind that the maturation of the structure and biochemical composition of rat arteries is achieved relatively late in ontogeny (43, 564). Aortic collagen biosynthesis reaches the adult level at the age of 7–9 wk (526), whereas the morphological and mechanical properties of conduit arteries become stabilized in rats aged 10–12 wk (42). Similarly, minimal vascular resistance of the rat hindquarter matures until the age of 9–11 wk (500).

It should be noted that the chronic administration of β-aminopropionitrile prevented the development of DOCA salt hypertension (205, 316). Similarly, chronic treatment of young SHR with the collagen cross-linking inhibitor β-aminopropionitrile (541) or with elastase (317) not only reduced arterial collagen deposition but also attenuated hypertension development. In both SHR and DOCA salt-treated rats, the BP lowering induced by chronic β-aminopropionitrile treatment was accompanied by a reduction of media cross-sectional area, increased distensibility of vascular wall, and higher baroreflex sensitivity (195). Thus the major alterations of arterial mechanical properties appearing during the development of hypertension in young rats might be part of a vicious circle sustaining the severity of the hypertensive process.

At present, there is sufficient evidence for considering the postweaning period (prepuberty and puberty) as the critical period for the development of severe self-sustaining forms of salt-dependent hypertension in the rat (322). This “critical period” for the hypertensive effects of high salt intake coincides with the “developmental window” in which long-term attenuation of hypertension development together with the suppression of enhanced vascular amplifier properties (375) can be achieved by transient short-term antihypertensive treatment of young SHR (2, 7, 70, 263, 600). To a certain extent, the changes in arterial wall properties might be the common denominator of the similarities in the progression of the hypertensive process in developing rats with salt or genetic hypertension.

X. IMPLICATIONS FOR HUMAN STUDIES

Both human essential hypertension and genetic hypertension of the rat are examples of a chronic, slowly developing cardiovascular disease that results from the interactions of polygenic predisposition with environmental influences taking place at particular stages of early ontogeny. The adult BP level seems to be predetermined in the critical developmental periods in which profound
modifications of major BP regulating systems can be induced. Because such early changes may lead to important late alterations of the cardiovascular phenotype, the existence of these relatively short critical periods (developmental windows) should be taken into account when considering future prevention of human cardiovascular diseases. There is no doubt that the comparison of human essential hypertension with genetic hypertension of the rat is difficult because both species differ substantially in genetic defects, gene-environment interactions, cardiovascular regulations, as well as in the relative length of particular developmental periods. Nevertheless, basic ontogenetic principles governing hypertension development in the rat and human are often surprisingly similar.

A typical example is the disproportionate prenatal growth. The fetal period has been recognized as an important developmental window in which the programming of adult BP levels occurs (26, 27, 405, 416). Low birth weight and enlarged placenta are associated with higher BP and/or increased incidence of coronary heart disease and syndrome X in adult humans. Similar impairment of fetoplacental growth was also observed in SHR (see sect. VIII A). Greater placenta, smaller birth weight, and elevated BP in adulthood are also characteristic findings in the progeny of rat mothers fed low-protein diets (see sect. VIII C) or treated with dexamethasone during pregnancy (see sect. VIII D). It has been suggested that the exaggerated access of maternal or exogenous glucocorticoids to the respective fetal receptors may permanently affect the maturation of important BP regulating systems of the fetus (including the SNS). Thus the increased fetal exposure to glucocorticoids toward the end of gestation may link low birth weight with hypertension in adulthood.

In both rat and human, the retardation of fetal growth is followed by accelerated postnatal growth (429, 636). The disproportionality between kidney and body growth, which results in an unduly small kidney relative to body mass, has been proposed to be an important factor in the pathogenesis of essential hypertension (429, 455, 729). In fact, enhanced postnatal body growth of the rat due to various nutritional interventions is usually accompanied by a BP elevation, and vice versa (see sect. VIII A and B). The important point is that the “catch-up” growth occurs during the most rapid phases of postnatal BP rise. It should be kept in mind that the BP of normotensive rats increases three to four times during the first postnatal month. The further BP elevation, which occurs between weeks 4 and 12, is mild in normotensive rats but accelerated in hypertensive rats in which the BP rise runs in parallel with the postweaning body growth spurt (636). It has recently been suggested that the hypertension development should be considered as a consequence of “abnormal telomere control of cellular growth and aging” (15). This might help to explain some links between high BP and the known abnormalities in cell proliferation and/or apoptosis in hypertensive subjects or animals.

There are many scientific, economic, and ethical reasons why the critical periods for hypertension development are more open to study in the rat than in the human. Figure 6 shows some of these periods in which distinct interventions can modify further development of the cardiovascular phenotype in the rat (usually by influencing the activity of the SNS and/or the RAS). Resulting structural and functional cardiovascular alterations are responsible for BP changes observed in adulthood. Most of the factors listed in Figure 6 also affect BP in humans, although their impact is often considered in adult subjects only. We hope that the findings on developmental aspects of genetic hypertension in the rat might be helpful for the future research on human hypertension. At least, they could indicate the relative importance of the particular ontogenetic stages for BP effects of pharmacological or nutritional interventions, which might be completely ineffective if applied at an inappropriate age.
Detailed information about the age-dependent influence of particular factors on later structural and functional development of the cardiovascular apparatus may not only help to make treatment more effective but also to introduce measures for prevention of human hypertension and/or other cardiovascular diseases. The use of rational age-specific pharmacological interventions, which would attenuate or even prevent further development of cardiovascular abnormalities in genetically predisposed individuals, is still complicated by several factors. First of all, there is a lack of detailed knowledge about particular developmental windows (critical periods) in which the maturation of the human cardiovascular system can be influenced. Furthermore, research on the ontogenetic aspects of human cardiovascular diseases is considerably complicated by their relatively late occurrence in life. There is a long lag period between presumed early developmental windows and the appearance of the first alarming symptoms of hypertension or atherosclerosis.

The possibility to intervene on the RAS during the development of human essential hypertension can be used as a classical example. There is no doubt about the involvement of this system in the pathogenesis of various forms of human and experimental hypertension (669). Chronic treatment with ACE inhibitors for 12 mo lowers BP and normalizes the altered structure of resistance vessels in patients with established essential hypertension (628, 695). Based on the classical study of Harrap and co-workers (263) concerning a juvenile critical period for perindopril treatment of SHR, Unger and Rettig (716) have suggested that brief transient administration of ACE inhibitors to adolescents with an increased family risk of hypertension should be considered. This tentative approach is, however, associated with some major problems. We still do not know exactly when, for how long, and how intensively we should treat such young subjects to achieve the desired cardiovascular effects in their adulthood. This difficulty could perhaps be solved by large-scale studies utilizing various dosages of ACE inhibitors or AT1 receptor antagonists administered during the period(s) in which subtle hemodynamic changes and/or alterations of resistance vessels begin to appear. The goal of such trials should not be to normalize the established cardiovascular abnormalities but to prevent their development. The contemporary research potential is certainly large enough to accomplish this task successfully. However, it is to be expected that the answer will not be obtained sooner than 15–20 years after the beginning of such studies.

Actually, we are approaching the era of gene therapy of cardiovascular diseases (176, 573). The overexpression of human genes for ANP (443), kallikrein (78), or eNOS (444) in young SHR effectively attenuates hypertension development, whereas BP-lowering effects are substan-

In this review we have tried to summarize the present state of knowledge about the age-dependent regulation of BP homeostasis with special focus on hypertension development. Our attention has mainly been paid to the age-dependent effects of various pharmacological and nutritional interventions on the development and maintenance of high BP. Furthermore, we have tried to specify some critical periods (developmental windows) in which particular stimuli can substantially affect the development of the cardiovascular phenotype. The existence of the cascade of critical developmental periods, through which each organism must pass during ontogeny, helps to explain the considerable variability of the cardiovascular phenotype in adult animals. In fact, the expression of genetic information during maturation is modified by numerous environmental factors acting in specific critical periods. This environmental impact can be seen even in individuals with a practically identical genome, i.e., in highly inbred rat strains with genetic hypertension, because certain important factors (such as mother-pup interactions or the proportion between maternal milk, solid food, and water intake during the weaning period) cannot be standardized even within the same litter.

There is ample evidence indicating that short-term treatment of young genetically hypertensive rats with some antihypertensive drugs (e.g., ACE inhibitors or AT1 receptor antagonists) in prepuberty and puberty (at the
Nevertheless, critical periods (developmental windows) should certainly be respected in the future prevention or gene therapy of human hypertension.

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1267

October 1999

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