ANIMAL MODELS OF HYPERTENSION AND EFFECT OF DRUGS

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ABSTRACT
Hypertension, the most common cardiovascular disease is the primary cause of stroke, coronary artery disease and sudden cardiac death. Many factors are believed to be involved in the pathogenesis of hypertension and its complications. Experimental models of human disease are used to study pathophysiological factors involved in hypertension and assess antihypertensive agents. Today different strains of rats with genetic hypertension are available and in most laboratories, therapeutic studies on hypertension are carried out on these models. As new insights into the pathogenesis of hypertension are revealed, new models are being developed to produce hypertension in animals. This article reviews experimental models of hypertension. Both conventional and genetic models of hypertension are discussed.

KEY WORDS  Experimental hypertension  blood pressure  cardiovascular disease

Introduction
Hypertension is the most common cardiovascular disease and is a major public health issue in developed as well as developing countries. Although it is common and readily detectable, it can often lead to lethal complications if left untreated. Because of its high incidence and morbidity, various classes of drugs and regimens have been advocated for the control of hypertension. Despite the large armamentaria of drugs being available for the treatment of hypertension, the last two decades have witnessed the introduction of a number of new antihypertensive drugs. Recent research during this period has also added considerably to our knowledge of the mechanisms involved in the pathogenesis of hypertension.

Human essential hypertension is a complex, multifactorial, quantitative trait under polygenic control. In order to understand the pathogenesis and to study the treatment and prevention of a disease, it is useful to develop animal models. Various models of experimental hypertension have been primarily developed to obtain information on the etiopathogenesis of hypertension. These models are also used in the pharmacological screening of potential antihypertensive agents. In the past, hypertensive animal models have been used infrequently for testing antihypertensive potential of drugs. As new molecules are being synthesized in a large number, the use of animal models is increasing for testing these molecules. New animal models of hypertension are being developed as new insights into the pathogenesis of hypertension are revealed.

The animal models of hypertension share many features which are common to human hypertension. Many of these models have been developed by utilizing the etiological factors that are presumed to be responsible for human hypertension such as excessive salt intake, hyperactivity of renin-angiotensin-aldosterone system (RAAS) and genetic factors. Since regulation of blood pressure (BP) is multifactorial, the effectiveness of an antihypertensive agent in one model does not necessarily mean that the mechanism of action of a given agent in a given model is related to the pathogenesis of elevated blood pressure.
An ideal animal model of hypertension should fulfill the following criteria:

- It should be feasible in small animals.
- It should be simple to perform and uniformly reproducible.
- It should be able to predict the potential antihypertensive properties of an agent.
- It should consume minimal quantities of compounds.
- It should be comparable to some form of human hypertension.

**Animals used:** Whereas in the past, most studies on experimental hypertension were carried out on dogs, currently rat is the preferred animal species. Spontaneous hypertensive rat (SHR), the genetic strain of hypertensive rat, is the animal of choice for screening antihypertensive agents. SHR is the cornerstone of medical research in experimental hypertension. Rabbits, monkeys, pigs and mice are also used to produce experimental hypertension.

The various types of animal models of hypertension being used are:

1. Renovascular hypertension
2. Dietary hypertension
3. Endocrine hypertension
4. Neurogenic hypertension
5. Psychogenic hypertension
6. Genetic hypertension
7. Other models

1. **Renovascular hypertension:**

   This is a very commonly used model of hypertension. In renovascular hypertension, the RAAS plays an important role. Experimentally, renal hypertension is produced by renal artery constriction, which activates peripheral RAAS and sympathetic nervous system. A number of factors like decreased blood volume may lead to sympathetic stimulation in this model. Renin is secreted by kidneys when sympathetic activity is increased. Renin converts angiotensinogen to angiotensin-I. Angiotensin-I is converted to angiotensin-II by angiotensin converting enzyme (ACE). Angiotensin-II is a potent vasoconstrictor and increases BP. Angiotensin-II also causes release of aldosterone leading to salt and water retention resulting in increased blood volume and hypertension (Figure 1).

The various methods of inducing renovascular hypertension are as follows:

I. **Goldblatt method:** Goldblatt et al (1934) reported that a partial constriction of renal arteries in dogs produced hypertension. This type of hypertension has also been induced in rabbits, rats and monkeys. In rabbits and rats, a U-shaped silver ribbon clip is used to constrict the renal arteries. Rats weighing from 120 to 200 g are anaesthetized with hexobarbitone sodium (40 mg/kg body weight) and a silver clip of 0.2 mm internal diameter is placed on the left renal artery close to the aorta. The renal artery in rats can also be ligated with 4-0 silk suture. Constriction of renal artery should be more than 50%. The animal is considered hypertensive if systolic BP is more than 160 mm Hg for two consecutive days after 4 weeks of ligation.

In dogs and rabbits, retroperitoneal approach can be used and the operation is carried out through an oblique lateral abdominal incision, just below and parallel to costal margin. Muscles are split and peritoneum is stripped away from the kidney. The kidney and the renal artery are easily identifiable. The first part of renal artery is cleaned for 1 cm or so from its origin from the aorta. In dogs, renal arteries can also be constricted with a small adjustable silver clamp or with silk sutures.

In rabbits, bilateral constriction of renal arteries is achieved in two stages with an interval of 1-4 weeks between operations. Persistent hypertension, which is moderate to severe, will develop over a period of 12 months. The BP has been reported to increase from normal mean level of 106 to 160-190 mm Hg from 4 weeks to 9 months in 65% of rabbits. In dogs, renal arteries can also be constricted with a small adjustable silver clamp or with silk sutures.

Three types of hypertension are produced by Goldblatt method:
a. **Two kidney one clip (2K1C) hypertension:**
The renal artery is constricted on only one side with the other artery (or kidney) left untouched. This results in a sustained increase in BP due to increased plasma renin activity (PRA), which in turn increases circulating angiotensin-II, a potent vasoconstrictor. However, there is no salt and water retention because of the other normal kidney being intact. Thus, the resultant hypertension at this stage is renin-angiotensin dependent. After about 6 weeks, the increased angiotensin-II releases aldosterone from adrenal cortex leading to a gradual retention of salt and water. Retention of salt and water leads to decreased renin production (Figure 1). From this stage onwards, hypertension is volume dependent\(^4,6,9\). Hence, salt and water balance is critically involved in the pathogenesis of renovascular hypertension. Increased BP and increased renin activity returns to normal by unclipping or removal of the affected kidney\(^6\).

b. **One kidney one clip (1K1C) hypertension:**
Constriction of renal artery is done on one side and the contralateral kidney is removed. There is an increase in BP within a few hours. Since there is no other kidney, there is no pressure diuresis and natriuresis, so there is rapid salt and water retention (Figure 1). Plasma renin activity is usually normal. Hypertension soon becomes volume dependent\(^4,6,9\).
c. **Two kidney two clip (2K2C) hypertension:** Constriction of aorta or both renal arteries is done. There is a patchy ischaemic kidney tissue, which secretes renin leading to increased BP. The remaining kidney tissue retains salt and water. Indeed, one of the most common causes of renal hypertension in human beings is such a patchy ischaemic kidney disease\(^4\).

II. **Hypertension induced by external compression of renal parenchyma:** This type of hypertension is produced in dogs, rabbits and rats. The following methods are used to produce this type of hypertension:

a. **Page hypertension:** A sheet of cellophane is placed around the kidney and held in place by silk sutures tied loosely around the renal hilus. Both kidneys are wrapped or one kidney is wrapped and other is removed\(^14\). A fibrocollagenous shell is formed around the kidney in 3-5 days because of reaction of the tissue to the foreign material. This shell compresses renal parenchyma leading to decreased renal vascular pressure. This expands the extracellular volume leading to increased peripheral resistance and hence increased BP. In rabbits, chronic hypertension with normal or decreased renin activity is produced over a period of approximately 6 weeks\(^6,15\). However, a high proportion of animals develops severe hypertension and dies within 2 months\(^8\).

b. **Grollman hypertension:** In this method, kidney tissue is compressed by securing a ‘figure of 8’ ligature around the kidney\(^16\). The ligature around the kidney forms a figure resembling number 8. This type of hypertension can be produced in dogs, rabbits and rats. It is of two types:

1. **Two kidney one ligature (2K1L):** Ligature is applied to one kidney and contralateral kidney is left untouched.
2. **One kidney one ligature (1K1L):** Ligature is applied to one kidney and contralateral kidney is removed.

III. **Coarctation of aorta:** Renal blood flow can be decreased by compressing the aorta. Coarctation can be done just above the renal arteries, between renal arteries and superior mesenteric arteries or between two renal arteries with the right artery above and the left artery below the site of coarctation\(^17\). An increase in BP similar to 2K1C model can be produced by applying a rubber band to abdominal aorta along with constriction of right renal artery for 8 weeks\(^18\). Coarctation can be followed by unilateral nephrectomy to produce this type of hypertension\(^6,19\).

IV. **Reduced renal mass:** Reducing renal tissue to five-sixth (5/6) by renal mass ablation produces hypertension. In this method, the right kidney is removed and 2 or 3 branches of left renal artery are ligated to produce infarction of approximately 2/3rd of the left kidney\(^20\).

V. **Glomerular sclerosis secondary to microsphere embolisation** is also used to produce renal hypertension\(^21\).

The mean arterial pressure in renal hypertensive rats produced by above methods (231.64±3.1 mm Hg) is significantly higher than the mean arterial pressure in normotensive control rats (160±2.84 mm Hg)\(^22\).

2. **Dietary hypertension:**

I. **Increased salt intake:** Physiologically, normal kidney has the ability to excrete easily the daily salt load without allowing a marked rise in extracellular volume. However, general epidemiological data have shown that higher the average sodium intake in a given population, the greater will be the prevalence of hypertension. Chronic ingestion of excess salt produces hypertension in rats, which mimics human hypertension morphologically\(^23\). High salt intake hypertension has been produced in rats, rabbits and chicks by replacing drinking water with 1-2% sodium chloride for 9-12 months\(^8,24\).

High salt intake along with unilateral nephrectomy produces accelerated hypertension within 3-4 weeks in dogs\(^25\). Accelerated renal hypertension in rats is produced by...
applying a 'figure of eight' ligature to one kidney and removing the other in a single stage operation performed under strict aseptic conditions using pentobarbitone sodium (20 mg/kg, i.p.) and ether anaesthesia. The drinking water is replaced with physiological saline for three weeks and the animals are used 4 to 5 weeks after the operation.

3. Endocrine hypertension:

I. Mineralocorticoid induced hypertension:

Mineralocorticoids cause retention of sodium and water in the body until escape diuresis occurs due to increased pressure on the kidneys. No further retention of sodium and water occurs, but general level of body sodium and water is slightly raised.

Selye et al was the first to demonstrate that deoxycorticosterone acetate (DOCA) produces hypertension in rats. There is increased DOCA-induced reabsorption of salt and water leading to increased blood volume and hence increased BP. There is also increased secretion of vasopressin leading to water retention and vasoconstriction. In addition, altered activity of RAAS leads to increased sympathetic activity. Rats, especially female and young, are prone to DOCA-salt induced hypertension. Other mineralocorticoids (e.g., aldosterone) and glucocorticoids can also produce this type of hypertension.

DOCA induced hypertension is salt dependent since neither administration of DOCA nor partial removal of renal mass is effective in increasing BP when applied without salt administration. To produce hypertension, rats weighing about 100 g are kept on a diet high in sodium chloride and drinking water is replaced by 2% sodium chloride solution ad libitum. After they attain a weight of about 250 g, they are given DOCA dissolved in sesame seed oil at a dose of 10 mg/kg, twice weekly for 43 days.

In another method, unilateral nephrectomy is performed followed by DOCA administration. All operated rats receive an injection of amoxicillin (10 mg/kg, i.m.) daily for 5 days and local application of neosporin-H to prevent infection. A week later, DOCA (25 mg/kg/wk, s.c. for 5 wk) dissolved in cotton seed oil, is injected into nephrectomised rats. Alternatively, nephrectomised rats could receive DOCA from silicon rubber implants (200 mg/rat) implanted subcutaneously. NaCl (1.0%) solution is substituted for drinking water and given ad libitum.

II. Adrenal regeneration hypertension:

Hypertension is produced in rats by unilateral nephrectomy followed by removal of right adrenal gland and enucleation of left adrenal gland. Enucleation is carried out by making a small incision in the capsule of adrenal gland through which the bulk of glandular tissue is extruded by gentle application of pressure with curved forceps. Drinking water is replaced with 1% saline. Hypertension develops during regeneration of adrenal glands in about 2 weeks. Once hypertension has been established, neither removal of the regenerated adrenal nor substitution of saline with drinking water brings about a return of BP to normal.

This type of hypertension develops more readily in female and young rats. This model is used mainly to study the role of various factors e.g., steroids, in the pathophysiology of hypertension.

4. Neurogenic hypertension:

One of the most important negative feedback in the control of BP originates from baroreceptors located in the carotid sinus and aortic arch. Afferents of baroreceptors travel along 9th and 10th cranial nerves. The first synapse is present in the nucleus tractus solitarius (NTS) of the medulla oblongata. NTS is not merely a relay center, it also contains complex synaptic connections and innervation from many brain areas and receives information from periphery. Stimulation of baroreceptors causes inhibition of vasomotor center leading to vasodilatation, bradycardia and decrease in BP. Sectioning of the baroreceptor nerves leads to persistent rise in BP in dogs, cats and rabbits. Neurogenic hypertension can be produced by the following methods:
I. **Denervation of sinoaortic baroreceptors:**

This is the most often used neurogenic model of hypertension. In dogs, cardioaortic nerve is located at the junction of superior laryngeal and vagus nerve and runs in the form of several fine strands. These strands unite and may be traced back as a white band lying within the vagal sheath alongside the cervical sympathetic nerve. Following bilateral vagotomy and carotid sinus denervation, the region is painted with 5% phenol and then alcohol to ensure complete denervation of the carotid sinus. There is sudden increase in BP. The dog is allowed to equilibrate for approximately 30 min and a bolus of the test compound can be given by intravenous administration. BP returns to normal within about 2 days because the response of vasomotor center to the absent baroreceptor signals fades away, which is called “resetting of baroreceptors”. Thus, this is only an acute type of hypertension.

In rabbits, right carotid sinus can be removed together with 2 cm segments of the right cervical sympathetic and depressor nerves while the left carotid sinus can be removed later on. In rats, sinoaortic denervation leads to marked and sustained increase in BP, which is comparable to renovascular hypertension or DOCA-induced hypertension. Some investigators found no increase in BP in dogs with sinoaortic denervation as compared to sham-operated dogs since BP was very labile in sinoaortic denervated dogs. They concluded that primary function of baroreceptors is not in regulation of BP but in minimising variations in BP caused by excitement, postural changes and diurnal rhythm. Bilateral destruction of NTS, the site of termination of the baroreceptor afferents, is more specific and complete than baroreceptor denervation and leads to severe hypertension that could be fatal. Baroreceptor denervation is not recommended as a model for screening antihypertensive agents.

II. **Electrical or chemical stimulation of different areas of the brain leads to development of hypertension in rats e.g., electrical stimulation of hypothalamus, glutamate injection into the rostral ventrolateral medulla.**

5. **Psychogenic hypertension:**

It has been reported that elevation of BP resulting from repeated exposure to stressful situation may lead to a state of persistent hypertension. Borderline hypertensive rats (BHR) are useful for psychogenic hypertension. BHRs that were exposed to daily sessions of either short (20 min) or long (120 min) duration air-jet stimulation developed hypertension within 2 weeks in comparison to home cage controls. Animals exposed to 120 min stress sessions had significantly higher systolic BP relative to the 20 minute group.

Other types of stress have been applied, such as emotional stimuli, psychosocial stress, immobilization stress and electric stimuli, but in all cases the results were similar. However, the degree and stability of hypertension was not comparable to other types of hypertension. The stress-induced hypertension was associated with either normal or suppressed PRA values, suggesting that the hypertension in these animals is not renin-dependent. As stress plays an important part in development of human hypertension, this model is very frequently used to study the pathophysiology of hypertension.

6. **Genetic hypertension:**

In 1963, Okamoto and Aoki introduced a new model of experimental hypertension that required no physiological, pharmacological or surgical intervention. The so called ‘spontaneous hypertensive rat (SHR)’ was developed by meticulous genetic inbreeding that uniformly resulted in 100% of the progeny having naturally occurring hypertension. Since then, several expert panels have reported that SHR is an excellent model of experimental hypertension that could serve as a counterpart for clinical essential hypertension as well as model for complications of hypertension. Estimates have ranged from a single gene to as many as six major genes that determine high blood pressure in these rats. In SHRs, BP gradually increases until it is maintained at a markedly elevated level after approximately 12 weeks of age. In unrestrained male SHRs, mean arterial pressure is approximately 190-200 mm Hg as compared to 115-130 mm Hg in normal rats. During the early stable stages and
developmental phase of hypertension, elevated BP is maintained in large part by enhanced central sympathetic outflow. In the later stages increased total peripheral resistance with a normal cardiac output and decreased permeability of the glomerular membranes form the basis for the long term maintenance of the hypertension. Comparison between spontaneous and essential hypertension reveal that both are caused by multifactorial inheritance and have in common an increased vascular resistance which is caused by neural vasomotor tone and non-neural structural alterations. Guidelines have been published for the maintenance and use of SHRs.

Spontaneous hypertension has been observed in a number of different strains of common laboratory animals, at least 6 different strains of rats, and at least one strain each of dogs and rabbits. The New Zealand strain of Smirk seems to be most similar to the Japanese SHR, though this has not been studied as widely. In contrast, the Milan strain developed by Bianchi et al, seems to be different involving primarily alterations in renal sodium and water metabolism; therefore it may not be analogous to essential hypertension. A fourth strain developed by Dahl shows a high sensitivity ("S" strain) to sodium intake in comparison to its normotensive control ("R" strain), which is sodium resistant. Furthermore, a fifth strain of hypertensive rats 'the Sabra strain' have been bred recently in Israel, and a sixth strain in France, the Lyon strain.

Among these strains, SHRs develop not only moderate to severe hypertension but also typical complications of hypertension. Stroke prone SHRs (SHRSP), which are selectively bred from among SHRs, are extreme examples that develop cerebrovascular lesions spontaneously in over 80% of rats. Because of high mortality rate of stroke in man, the similarity of stroke in SHRSP is important for further application of this model to studies of stroke in man. Other variants derived from SHRs are obese SHRs and arteriolipidosis-prone SHRs. Extreme environmental conditions such as stress and excess salt accelerate the development of hypertension and aggravate the hypertensive complications.

Complications such as cerebral hemorrhage, thrombosis, nephrosclerosis and myocardial lesions in SHRs and especially cerebral lesions in SHRSPs, are pathogenically, pathologically and epidemiologically similar to those observed in essential hypertension in man. Therefore, these models can be used to study not only the pathogenesis and therapy but also prophylaxis in essential hypertension and its complications. Because of apparent similarities of the SHR to essential hypertension, SHR models are highly recommended for screening potential drug candidates for hypertension.

Experimental models of genetic hypertension have also been developed in animals other than the rat. Though it has been produced in one strain of rabbit and dog they have not been studied extensively for practical, financial and other reasons. Rats are mainly used because of their small size, short life-span and low cost.

7. Other models:

1. **Obesity-related hypertension:** Wistar fatty rats (WFR) derived from cross between obese Zucker and Wistar Kyoto rats show persistent hyperinsulinemia and hypertension after 16 weeks of age and may be a good model to elucidate the relationship between hyperinsulinemia and hypertension.

2. **Hypertension induced by cholinomimetic agents:** Physostigmine (10-80 µg/kg, i.v.), a cholinesterase inhibitor, and oxotremorine (20-40 µg/kg, i.v.), a direct muscarinic cholinergic agonist, cause a dose-dependent increase in BP. The cholinomimetic-induced hypertension has been shown to be elicited through activation of central cholinergic mechanism and mediated peripherally through sympathetic nervous system. Pretreatment with methyl scopolamine (1 mg/kg) 5-10 minutes before giving oxotremorine prevents initial hypotension in this model.

3. **Angiotensin-II induced hypertension:** Subcutaneous infusion of angiotensin-II (0.7 mg/kg/day) using minipump elicits hypertension in 4-8 weeks.

4. **Hypertension induced by cadmium:** Hypertension is produced by the chronic administration of CdCl (1 mg/kg/day, i.p. for 2 wk). CdCl-induced hypertension might be due to the fact that the metal ion might mimic...
Ca\textsuperscript{2+} ion as a partial agonist and produce a direct contractile effect on vascular smooth muscle\textsuperscript{24}.

5. **Chronic nitric oxide inhibition-induced hypertension**: SHRs when given a non-selective nitric oxide synthase inhibitor, \( \text{N}^\text{ω}-\text{nitro-L-argininemethyl ester (L-NAME),} \) for 4 weeks develop time and dose-dependent hypertension\textsuperscript{62}.

6. **Transgenic rat (TGR) models**: Transgenic models of hypertension have revolutionized the experimental work on hypertension. Several transgenic rat lines expressing candidate genes for hypertension have been produced. Introducing an additional renin gene, the murine Ren-2 gene, into the germ line of rats results in transgenic hypertensive rat strain, TGR (mREN2)\textsuperscript{27}, with an overexpression of renin. Genetic linkage studies have shown that the components of RAAS are associated with this type of hypertension. Linkage has been described for the angiotensinogen gene in human hypertension. TGR model may be useful for studying the role of local RAAS system in hypertension\textsuperscript{63}.

7. **Uterine ischaemia**: As in preeclampsia, uterine ischaemia can induce hypertension in rats and monkeys\textsuperscript{8,64,65}. In monkeys at 116±7 days of gestation, lower aortic pressure was reduced by 24±11 mm Hg by a stricture on the aorta just below the renal arteries and animals developed sustained hypertension\textsuperscript{66}.

Comparison of animal models with human hypertension is shown in Table 1. SHRs have been compared most extensively with essential hypertension in human beings.

**Measurement of BP in animal models**: Repeated measurement of BP is needed in experiments on animal models of hypertension. In acute experiments, the animals are usually anesthetized and BP is measured for the duration of the experiment by exposing an accessible artery and inserting a cannula connected to a mercury manometer or a pressure transducer. Frequent repetition of the surgical arterial exposure under anaesthesia is undesirable. Anaesthesia is used only when immobilization of the animal is required to measure BP. Anaesthesia makes determination of BP easier and more accurate but the use of anaesthesia introduces an additional variable. Anaesthetic agents have been reported to alter BP and cardiovascular reflexes\textsuperscript{66}. BP can be recorded in unanaesthetised state. There should be minimal stress to the animal. The balance of relative disadvantages favours the use of unanaesthetised state only for those methods not requiring rigid restraint, for example, aortic intubated rats.

The BP in the following animal models are measured either directly (intravascular) or indirectly (bloodless).

### Direct methods:

**In dogs**:

a) In unanaesthetised condition a small hypodermic needle is inserted into a superficial artery (\textit{e.g.}, femoral artery) and connected to a mercury manometer or pressure transducer. The system is filled with an anticoagulant solution\textsuperscript{8,67}.

b) Carotid artery can be exteriorized in dogs. Dogs are trained to cooperate and lie quiet on table. Artery is punctured with a needle attached to direct writing multichannel recorder\textsuperscript{68}.

c) Intraaortic cannulation: A cannula or catheter filled with heparin is advanced from femoral artery to abdominal aorta in anaesthetised animals. Cannula can also be passed from proximal stump of a cut left cranial thyroid branch through

| Table 1. Comparable forms of experimental and clinical hypertension. |
|-------------------------------------------------|------------------|
| **Experimental Hypertension**                   | **Clinical Hypertension**  |
| Spontaneous hypertension                        | Essential hypertension |
| Renal artery stenosis hypertension               | Renal artery stenosis |
| Overdosage of glucocorticoids                    | Cushing syndrome     |
| Overdosage of mineralocorticoids                 | Primary aldosteronism |
| Overdosage of salt                               | Chronic high salt intake |
| Obesity related hypertension                     | Obese hypertension   |
| Uterine ischaemia                               | Preeclampsia         |
| Reduced renal mass                               | Renal diseases       |
common carotid artery down to descending thoracic aorta and BP is measured.

**In rabbits:** a) In an unanaesthetised animal, central artery of ear is cannulated. The disadvantages of this method include development of hematoma and arterial spasm until total denervation of ear is carried out and animal needs to be closely restrained. Alternatively, carotid artery can be cannulated after infusion with 1% lignocaine. Cannulas made from vinyl tubing (outside diameter 1.52 mm and internal diameter 0.86 mm) flushed with heparinised (50 u/ml) 0.9% saline is used. An exteriorised arterial loop can be used as in dogs.

**In rats:** a) A week before the experiment, each rat is anaesthetised with 40 mg/kg pentobarbital. Left or right carotid artery or femoral artery (for recording BP) is cannulated under aseptic conditions with polyethylene cannula filled with 1% heparin in normal saline. Free end of the cannula is passed under the skin and allowed to protrude 3-4 cm from the skin behind the ears of the rat. The skin incisions are sutured and a plastic skin dressing is applied. After recovery from anaesthesia (2-2.5 h) each rat is placed in an individual cage for 24 h habituation period.

On the day of the experiment, a pressure tube filled with 200 U/ml heparin in saline is tied to the implanted catheter and connected to a pressure transducer and then to the pre-amplifier and recorded on the polygraph or physiograph. Alternatively, Condon’s mercury manometer can also be used for recording BP in rats. Popovic et al. (1960) used animals for 40 days before cannula was blocked.

b) Abdominal aorta is cannulated with a small polyethylene tube and led beneath the skin to an exit at the back of the neck for recording arterial BP. But a large operation like laparotomy is required. In other methods, catheters are inserted into the inferior vena cava via the left femoral vein for intravenous injections and the abdominal aorta via the left femoral artery for the continuous recording of mean BP and heart rate on a polygraph via a pressure transducer.

**Indirect methods:** These following methods are used in unanaesthetised animals.

**In dogs:** a) Tibial artery is occluded with a cuff on the anteromedial surface of hindlimb or radial artery on forelimb and BP is recorded by auscultation or condenser microphone. The problem in this method is inherent elasticity of tissue surrounding the artery.

b) Exteriorised artery in a skin flap can be used conveniently for long periods. The problem in this model is carotid sinus reflex. Femoral artery loop has been used in dogs and cats.

**In rabbits:** a) The central artery of ear is compressed by a transparent membrane applied to ear, which permits visualisation of the central artery. The pneumatic pressure required just to break the column of blood in this portion of central artery is taken as the systolic pressure. This is a cheap and simple method. Disadvantage is that BP in rabbits is highly labile, BP in central artery is lower than carotid artery and pressure varies with the tone of arterial wall, which is affected by heat.

b) BP is recorded by a cuff around radial artery in forelimb.

**In rats:** a) Tail cuff is a common and convenient method to measure systolic pressure in rats. Tail cuff is inflated and then deflated. Pulsations disappear when cuff is inflated. When cuff is deflated pulsations start appearing when pressure in the cuff equals systolic pressure. Various devices are used. The cuff is attached to a tail cuff sphygmomanometer or more commonly to pressure transducer and BP.
is recorded on a chart. Training the animal and warming the tail are required for this method. Computerised tail cuff instrument can also be used.

b) Tail swelling: Pressure is applied to the tail with a cuff. This results in swelling of the tail. When the pressure is lowered swelling starts decreasing indicating systolic pressure.

c) Foot swelling: Pressure is applied on the proximal part of the foot. Swelling appears on the foot. Photocell is used to measure the light obstructed by the foot. Swelling decreases the light reaching the photocell.

Effects of antihypertensive agents: Today, each new antihypertensive drug will be studied in more than one hypertension model before it undergoes clinical trials. In the assessment of antihypertensive activity, it is essential to make sure that the drug is administered according to a fixed schedule, which corresponds to its duration of action. The BP should be measured at various intervals after administration of the drug. It is necessary to prepare a protocol for such a study, which should be strictly adhered to and monitored. Usually the drug is given after establishment of hypertension to obtain a therapeutic effect. In SHRs drugs are administered at an early stage to prevent an increase in BP. Complications of hypertension can be abolished or delayed by effective control of hypertension.

Antihypertensive drugs, according to their mode of action, may affect the blood pressure in certain types of experimental hypertension, and not in others. Diuretics and β-adrenergic blockers have no or little effect in renal hypertension. In contrast, pharmacological agents that interrupt the functioning of the RAAS to prevent angiotensin II production or binding to target receptors are highly effective in these models. These drugs e.g., ACE inhibitors, angiotensin-II type-I (AT-1) receptor antagonists and renin inhibitors are effective in initial renin or angiotensin dependant phase of 2K1C hypertension model and other renovascular models. ACE inhibitors and AT-1 antagonists decrease BP in initial phase of 2K1C hypertension. In 1K1C model, ACE inhibitors and AT-1 antagonists are not effective except in very early phase. All known AT-1 receptor antagonists and ACE inhibitors induce a marked and sustained hypotension in high renin-dependent hypertensive models such as 2K1C renal hypertensive rats and renal artery-ligated hypertensive rats. They are more effective in renal hypertensive rats than in SHRs. Both ACE inhibitor and AT-1 antagonists have negligible effect in DOCA-salt hypertensive rats, a low-renin model of hypertension. This indicates that normal levels of plasma renin activity are necessary to demonstrate the antihypertensive action of these drugs. However, ACE inhibitors and AT-1 antagonists are effective in salt induced hypertension in reduced renal mass rats and in transgenic rats, TGR (mREN2) 27. Furthermore, these drugs have been reported to decrease complications and mortality of the salt-loaded SHRSP.

Vasodilators like minoxidil, hydralazine and diazoxide are effective in renal hypertensive rats. Calcium channel blockers (CCB), ACE inhibitors and AT-1 antagonists decrease BP in 5/6 nephrectomised SHR and are also effective at reducing renal injury in these rats. Diuretics, are active in mineralocorticoid or salt induced hypertension.

Endocrine hypertension is the preferred model for screening antihypertensive activity of diuretics e.g. chlorthiazide and hydrochlorthiazide. Both endothelin receptor antagonists and CCBs are effective in endocrine hypertension. Drugs targeting sympathetic nervous system like phenoxybenzamine, guanethidine, α-methyl dopa and clonidine decrease BP in both endocrine and neurogenic hypertension. β-adrenergic blockers decrease BP neither in renal nor in endocrine hypertension, but show some effect in SHR at a later stage when sympathetic activity is increased. SHR shows inconsistent BP responses to β-blockers, diuretics and sodium restriction, which are common and effective antihypertensive drug classes in essential hypertension.

In conclusion, it may be stated that the various animal models of experimental hypertension are tools in the study of the pathophysiology of sustained hypertension and its complications. These animal models are increasingly being used for testing new chemical entities. Similarity of SHR to essential hypertension and its complications and its easy availability has made SHR, the main animal model of hypertension.
SHR is widely used because of its reliable spontaneous development not only of hypertension but also of hypertensive complications. Thus, until a better experimental model is available, we feel justified in taking the affirmative position that the SHR is indeed an excellent laboratory counterpart of essential hypertension. Development of new models incorporating recent advances in pathophysiology of hypertension can accelerate the research in understanding the pathophysiology and development of new therapeutic agents for hypertension.

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