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INFLUENCE OF AGE AND HYPERTENSION ON ARTERIAL BARORECEPTOR REFLEX ACTIVITY IN MALE RABBITS

(With 5 Tables and One Figure)

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تأثير العمر وارتفاع ضغط الدم على نشاط المنعكس الضغطى فى ذكور الأرانب

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أجرى هذا البحث لدراسة تاثير العمر على كل من، نشاط المنعكس الضغطى ، حساسيه المنعكس الضغطى وآلية حدوث حساسية المنعكس الضغطى وكذلك دراسة حساسية المنعكس الضغطى فى الارانب ذات ضغط الدم المرتفع. أستخدم فى هذا البحث 60 أرنبا قسمت الى 10 مجموعات سته أرانب بكل مجموعه وقسمت الأرانب الى 3 أعمار : الأرانب البالغه (المجموعات4,3,2,1) من 4-6 أشهر وتزين 1,5-2 كيلو جرام والأرانب متوسطه العمر (المجموعات7,6,5) من 10-12 أشهر وتزن 2,5-3 كيلو جرام والأرانب المتقدمه فى العمر (المجموعات10,9,8) من 20-24 شهر وتزن 3-3,5 كيلو جرام. أجريت التجربتين الأولى والثانيه باستخدام المجموعات 9,8,6,5,2,1 لدراسه تأثير العمر على وظيفه المنعكس الضغطى عن طريق رفع ضغط الدم الشريانى بالحقن الوريدى لماده فينيل افرين (50ميكرو جرام/كجم من وزن الأرنب المختبر) ثم قياس معدل نبضات القلب، معدل سرعه التنفس، نشاط العصب الحائر ونشاط العصب الشوكى الوركى. وأجريت التجربه الثالثه باستخدام المجموعات 10,7,3 لدراسه تأثير العمرعلى حساسيه المنعكس الضغطى وكذلك اليه حدوث حساسيه المنعكس الضغطى. أما التجربه الرابعه فأجريت باستخدام المجموعه الرابعه لقياس المنعكس الضغطى فى الأرانب البالغه ذات الضغط المرتفع. أثبتت النتائج أن المستوى القاعدى لكل من ضغط الدم، معدل نبضات القلب، معدل التنفس ومعدل نشاط العصب الحائر لا يتغير تغيرا جوهريا مع تقدم العمر بينما يزيد معدل نشاط العصب الوركى زياده ذات دلاله احصائيه مع تقدم العمر. كما تبين أن كلا من وظيفه المنعكس الضغطى وحساسيه المنعكس الضغطى بعد رفع ضغط الدم الشريانى تظل طبيعيه مع زياده العمر. وأظهرت النتائج أيضا أن نشاط المنعكس الضغطى ينتقل عن طريق الجهازين العصبيين الجار سمبثاوى والسمبثاوى فى المراحل العمريه الثلاث، لكن الجهاز العصبى الجار سمبثاوى هو المسئول عن نقل الغالبيه العظمى لهذا النشاط. وكذلك تقل حساسيه المنعكس الضغطى فى الأرانب ذات ضغط الدم المرتفع وذات العلامات الأولى الداله على تصلب الشرايين.

SUMMARY

This study aims to evaluate the respective changes in arterial baroreflex function (changes in heart rate and respiratory rate in response to induced increase in blood pressure) in adult, middle aged and old male rabbits, as well as to assess the effect of aging on baroreflex sensitivity and baroreflex regulation of parasympathetic and sympathetic out flow. In addition, the baroreflex sensitivity was also studied in adult male rabbits rendered hypertensive. This study included 60 male Baladi rabbits (10 groups 6 animals each) and four experiments. Experiment I to evaluate the respective changes in heart rate (HR) and respiratory rate (RR) in response to acute changes in arterial blood pressure (BP) achieved with phenylephrine in adult, middle aged and old rabbits. Experiment II to study the parasympathetic (vagus) and sympathetic (sciatic) nerve activities (VNA, SNA) in the three different age groups before and during acute blood pressure changes achieved with phenylephrine injection. Experiment III to evaluate the effect of aging on baroreflex sensitivity as well as the extent to which the parasympathetic and sympathetic nerves were responsible for baroreceptor sensitivity that measured by using selective autonomic blockade. Experiment IV to evaluate the baroreflex sensitivity in adult rabbits rendered hypertensive. Aging in male rabbits was associated with preserved base line level of BP, HR, RR and VNA. While, SNA was significantly increased in old versus adult and middle aged groups respectively. Phenylephrine injection produced a significant increase in BP and VNA and a significant decrease in HR, RR and SNA in the three age groups studied. Old, middle aged and adult male rabbits had nearly similar magnitude of arterial BP response to phenylephrine and similar HR, RR, VNA and SNA responses to phenylephrine. Baroreflex sensitivity was significantly increased after elevating BP with phenylephrine injection in the three age groups and was significantly decreased after receptor blockade with either atropine or propranolol in the three age groups and the major portion of the decrease was due to the parasympathetic nervous system. In rabbits rendered hypertensive, baroreflex sensitivity was significantly decreased after elevating the BP by phenylephrine in comparison with normotensive control. It can be concluded that indices of baroreflex function and baroreflex sensitivity were relatively well maintained in old rabbits. Parasympathetic and sympathetic components of the ANS normally mediate the baroreflex in old age. Baroreflex

sensitivity is depressed in adult rabbits rendered hypertensive and with early signs of atherosclerosis.

Key words: *Baroreceptors, baroreflex sensitivity, aging, hypertension, rabbits.*

INTRODUCTION

In cardiovascular physiology, the baroreflex or baroreceptor reflex is one of the body's homeostatic mechanisms for maintaining blood pressure and ultimately maintaining circulation to the brain and other organs (Berne and Levy, 2001 and Boron and Boulpaep, 2005) Baroreceptors are pressure receptors that respond to the tension of the arterial wall and transmit information about cardiovascular status to the brain (Rau and Elbert, 2001). Variations in the extent of stretch applied to the baroreceptors modulate the afferent discharge transmitted to the CNS (Nasr *et al.,* 2005) and produce autonomic adjustments to maintain arterial blood pressure within a narrow range (Helke and Segard, 2004).

Aging is associated with a variety of alterations in cardiovascular function, glucose homeostasis and autonomic reflexes in rats (Draining *et al.,* 1985 Irigoyen *et al.,* 2000, and Dias da Silva *et al.,* 2006) and human (Dinenno *et al.,* 2000; Ferrari, 2002, Mattace-Raso *et al.,* 2006 and Skrapari *et al.,* 2006).

There has been increased interest in the influence of aging on cardiovascular homeostasis. This growing interest is due in part to the dramatic increase in the elderly population (Mc Veigh *et al.,* 1999). Also, it is due to increase in the morbidity and mortality rates associated with common cardiovascular abnormalities in old age such as orthostatic hypotension (Glasser *et al.,* 1997; Greengross *et al.,* 1997; Lanfranchi and Somers, 2002 and Mattace-Raso *et al.,* 2006).

Results obtained from aged animals and human related to baroreflex function have been less conclusive. No change in baroreflex function in rats (Wei *et al.,* 1986, Kurosawa *et al.,* 1987 and Ferrari *et al.,* 1991), rabbits (Katsuda *et al.,* 1990) racing greyhound (Cox *et al.,* 1981) and human (Ng *et al.,* 1995 and Shi *et al.,* 1996) has been reported. However, other studies revealed that baroreflex function is impaired with aging in rabbits (Frolkis *et al.,* 1975) in beagles (Hajduczok *et al.,* 1991), in rats (Irigoyen *et al.,* 2000 and Dias da Silva *et al.,* 2006) and even more in human (Laitinen *et al.,* 1998, Monahan *et al.,* 2001, Labrova *et al.,* 2005 and Mattace-Raso *et al.,* 2006).

The purpose of the present study is to evaluate the respective

change in arterial baroreflex function (changes in heart rate and respiratory rate in response to induced increase in blood pressure) in adult, middle-aged and old male rabbits, as well as to assess the effect of aging on baroreflex sensitivity and baroreflex modulation of parasympathetic and sympathetic outflow. In addition, to assess the baroreflex sensitivity in adult male rabbits rendered hypertensive.

MATERIALS and METHODS

Animals: The present study was carried out on 60 male Baladi rabbits of three different ages divided into 10 groups, 6 animals each and used in four experiments. The animals were given free access to water and commercial rabbit food and kept in the same environmental conditions. All animals were in a good state of health and appetite at the time of the study. The animals were divided into adult groups [G 1, 2, 3, and 4 (4-6 months and 1.5-2 Kg)], middle-aged groups [G 5, 6 and 7 (10-12 months and 2.5-3 Kg)] and old groups [G 8, 9 and 10 (20-24 months and 3-3.5 Kg)].

Rabbits were anesthetized with urethane (600 mg/Kg) injected intraperitonealy. The trachea was exposed by blunt dissection and intubated with a short cannula to ensure free air way during the experiments. One catheter was inserted into the right juglar vein for intravenous (iv) administration of the drugs and another one into the left common carotid artery for continuous monitoring of blood pressure. Heparin (0.25 ml) was iv injected to prevent intracatheter clotting.

Drugs and solutions: Urethane was purchased from Aldrich Chemical Company (Wis., USA) heparin (aspirigine) from Pharco Pharmaceutical (Egypt), atropine sulphate, phenylephrine and epinephrine from Sigma Chemical Company (USA) and propranolol from Macclesfeild (Cheshire UK).

The physiograph (Washington 400 MD Ossillograph Bioscience Sheerness, Kent UK) was used in this study. BP was recorded by a pressure transducer and facility coupler "FC", 137, HR by ECG transducer and FC 123 and RR by a needle electrode and FC 117.

In experiment I, three age groups of rabbits [adult group (G1), middle-aged group (G5), and old rabbits (G8)] were used to evaluate the respective changes in heart rate (HR) and respiratory rate (RR) in response to acute changes in arterial blood pressure (BP). These parameters were recorded before and during acute blood pressure

changes achieved with intravenous administration of phenylephrine (50 µg/kg BW) (Patricia *et al.,* 1999) in the three age groups.

Data analysis: Baseline level (control level) of each measured parameter (BP, HR and RR) was recorded during stable conditions for a 30 minutes control period (pre-injection level). BP, HR, and RR were recorded at 1 minute after phenylephrine injection (post-injection level). Mean blood pressure was mathematically calculated (Ganong, 2005). Heart rate was determined as beats/minute and respiratory rate was determined as frequency / minute.

In experiment II, three groups of rabbits [adult group (G2), middle-aged group (G6), and old rabbits (G9)] were used to study the parasympathetic (vagus nerve) and sympathetic nerve (sciatic nerve) activities (VNA and SNA respectively) in the three different age groups. These criteria were recorded before and during acute blood pressure changes achieved with phenylephrine injection (50 µg/kg BW).

Nerve recording: The sciatic and vagal nerve traffics were recorded after their stimulation with microelectrode connected to the stimulator cable [electronic stimulator (SEN.3201)]. The sciatic and vagal signals were magnified with Washington coupler (814-80950-0). The nerve responses were recorded on the physiograph.

Data analysis: Baseline levels (control levels) of vagal nerve activity (VNA) and sciatic nerve activity (SNA) were determined (pre-injection levels) and then repeated 1 minute after iv injection of phenylephrine (50 µg/kg) (post injection levels). VNA and SNA were determined as bursts/min.

In experiment III, three age groups of rabbits [adult (G3), middle-aged (G7), and old rabbits (G10)] were used to study the effect of aging on baroreflex sensitivity [the slope relating heart rate to acute blood pressure changes achieved with phenylephrine injection (50 µg/kg BW)], and the extent to which the parasympathetic and sympathetic nerves were responsible for baroreceptor sensitivity by using selective autonomic blockade [atropine sulphate (1 mg/kg BW) (Shirley *et al.,*1990) and propranolol (1 mg/kg BW) (Irigoyen *et al.,* 2000)].

Data analysis: the sigmoid relationship between systolic BP (mm Hg) and R-R interval (m sec) was analyzed after phenylephrine injection (Symth *et al.,* 1969). The slope (msec / mmHg) of this relationship was used as index of the baroreflex sensitivity (BRS).

BRS before and after atropine and propranolol blockade was compared to determine the percentage of the response mediated by parasympathetic and sympathetic limbs of the reflex.

In experiment IV, the baroreflex sensitivity was studied in adult male rabbits rendered hypertensive (G4) by intra-peritoneal injection of epinephrine (0.6 mg/kg BW) for 15 days (Mohamed *et al.,* 1989). Transverse sections from the aortas of group 3 (adult control), group 4 (adult hypertensive) and group 10 (old normotensive) were taken, prepared and examined under light microscope.

Statistical analysis: Data were expressed as means ± SE and analyzed by using Prism program version 3. Differences between groups were tested using unpaired "t" test. Differences were considered significant at P value 0.05 or less. The sigmoid baroreflex relationship between systolic BP and R-R interval were compared and analyzed by the method of Zar (1984) using least square linear regression.

RESULTS

The first two experiments (groups 1,2,5,6,8 and 9) were performed to study the pressure induced reflex changes in four criteria assessing the baroreflex function (HR, RR, VNA and SNA) in response to elevating the blood pressure by iv injection of phenylephrine.

Table (1) shows that phenylephrine injection produced a significant increase in BP ($P<0.001$) and VNA ($P<0.001$, $P<0.01$, $P<0.01$) and a significant decrease in HR ($P<0.001$, $P<0.01$, $P<0.05$), RR ($P<0.05$) and SNA ($P<0.05$) in the three age groups studied (adult, middle aged and old rabbits).

The results of these two experiments demonstrated that aging in male rabbits was associated with nonsignficant increase in BP and nonsignificant decrease in HP, RR and VNA. While, SNA was significantly increased ($P<0.01$, $P<0.05$) in old versus adult and middle aged groups respectively (Table, 2).

Indices of baroreflex function in old rabbits including reflex responses of the HR and RR and reflex changes in parasympathetic and sympathetic nerve activities in response to increased BP with phenylephrine were similar to those documented in adult and middle-aged rabbits. The three age groups have nearly similar magnitude of arterial BP response to phenylephrine and similar HR, RR, VNA and

SNA responses to phenylephrine.

In experiment III, baroreflex sensitivity was significantly increased ($P<0.001$) after elevating the BP with phenylephrine injection in adult, middle-aged and old male rabbits. The three age groups studied showed no difference in the characters of baroreflex sensitivity, the baroreflex slope increased immediately and lasted nearly for 60 seconds. The maximum response of baroreflex sensitivity was measured at 30 seconds. The baroreflex sensitivity was nearly similar, the percentage increase in baroreflex sensitivity with respect to the immediate effect for adult was 426.47 %, for middle-aged was 401.52 and for old rabbits was 365.35 % (Table, 3). The parasympathetic and sympathetic components of the autonomic nervous system were normally responsible for mediating the baroreflex in the three age groups. Baroreflex sensitivity was significantly decreased after receptor blockade with either atropine ($P<0.001$, $P<0.001$, $P<0.01$) or propranolol ($P<0.01$) in the three age groups. However, the major portion of the decrease in baroreflex was due to the parasympathetic nervous system (62.99, 61.33, 58.54 %), despite the slight decrease in the parasympathetic component and the slight increase in the sympathetic components with the increase in age (Table 4).

From experiment IV, it is clear that the baroreflex sensitivity in rabbits rendered hypertensive was significantly ($P<0.001$) decreased after elevating BP by phenylephrine in comparison with that of normotensive adult rabbits (Table, 5). Microscopic examination of the transverse sections of the aortas from adult rabbits rendered hypertensive (Fig 1, C and D) showed different picture from that of the normotensive adult rabbits (Fig 1, A). The aorta showed thickening of the wall, eroded endothelial lining and irregular arrangement of smooth muscle fibers. Foam cells were present in the intimal and subintimal layers. Whereas, microscopic examination of the aortas of the old rabbits (Fig 1, B) showed similar finding to those of the normotensive adult rabbits. The aortas showed a smooth and intact endothelial lining and regularly arranged smooth muscle fibers. There were no foam cells in the intimal and subintimal layers.

Table 1: Mean arterial blood pressure (BP), heart rate (HR), respiratory rate (RR), vagal nerve activity (VNA), and sympathetic nerve

activity (SNA) before and after phenylephrine (PE) injection in male rabbits in relation to age.

Variable	Adult		Middle- aged		Old	
	Baseline level	PE	Baseline level	PE	Baseline level	PE
BP (mmHg)						
Mean	81.00	133.17 ***	86.83	139.33 ***	92.00	144.71 ***
± SE	± 2.88	± 3.92	± 1.35	± 5.46	± 4.10	± 5.82
% Increase		64.41		60.46		57.25
HR (beats/min)						
Mean	291.67	203.33 ***	270.00	200.00 **	255.00	196.67 *
± SE	± 13.02	± 12.56	± 10.33	± 14.61	± 10.88	± 9.89
% Decrease		30.28		25.93		22.87
RR(frequency/min)						
Mean	79.17	63.00 *	73.83	62.17 *	71.17	60.00 *
± SE	± 4.09	± 4.15	± 4.38	± 2.61	± 3.57	± 1.98
% Decrease		20.42		15.79		15.69
VNA (bursts/min)						
Mean	91.67	128.17 ***	88.17	115.00 *	80.00	100.17 **
± SE	± 4.66	± 6.60	± 4.04	± 6.04	± 4.43	± 4.81
% Increase		39.82		30.43		25.21
SNA (bursts/min)						
Mean	85.33	68.00 *	97.50	82.00 *	112.50	95.83 *
± SE	± 3.75	± 4.48	± 4.01	± 3.43	± 5.32	± 4.82
% Decrease		20.31		15.90		14.82

SE: Standard error, *: Significant at 0.05 level of probability,
: Significant at 0.01 level of probability, *: Significant at 0.001 level of probability.

Table 2: Comparison of mean baseline data of arterial blood pressure (BP), heart rate (HR), respiratory rate (RR), vagal nerve

activity (VNA), and sympathetic nerve activity (SNA) in male rabbits in relation to age.

Variable	Adult	Middle-aged	Old	Adult versus middle-aged	Middle-aged versus old	Adult versus old
BP (mmHg) % Increase	81.00 ± 2.88	86.83 ± 1.35	92.00 ± 4.10	NS 7.20	NS 5.95	NS 13.58
HR (beats/min) % Decrease	291.67 ± 13.02	270.00 ± 10.33	255.00 ± 10.88	NS 7.43	NS 5.56	NS 12.57
RR (frequency/min) % Decrease	79.17 ± 4.09	73.83 ± 4.38	71.17 ± 3.57	NS 6.74	NS 3.60	NS 10.10
VNA (bursts/min) % Decrease	91.67 ± 4.66	88.17 ± 4.04	80.00 ± 4.43	NS 3.82	NS 9.27	NS 12.73
SNA (bursts/min) % Increase	85.33 ± 3.75	97.50 ± 4.01	112.50 ± 5.32	* 14.26	* 15.38	** 31.84

Values as mean ±SE, *: Significant at 0.05 level of probability,
**: Significant at 0.01 level of probability, NS: Non significant

Table 3: Baroreflex slope in response to phenylephrine (PE) injection in male rabbits in relation to age.

	Control	30 sec after PE	60 sec after PE	120 sec after PE
Adult Mean ± SE % Increase P value	1.36 ± 0.10	7.16 ± 0.60 426.47 ***	3.16 ± 0.27 132.35 ***	1.52 ± 0.11 11.76 NS
Middle-aged Mean ± SE % Increase P value	1.32 ± 0.19	6.62 ± 0.67 401.52 ***	2.95 ± 0.25 123.48 ***	1.46 ± 0.20 10.61 NS
Old Mean ± SE % Increase P value	1.27 ± 0.13	5.91 ± 0.66 365.35 ***	2.79 ± 0.19 119.69 ***	1.40 ± 0.15 10.24 NS

SE: Standard error, ***: Significant at 0.001 level of probability, NS: Non significant

Table 4: Baroreflex slope after receptor blockade in male rabbits in relation to age.

	Adult	Middle-aged	Old
Phenylephrine			
Mean ± SE	7.16 ± 0.60	6.62 ± 0.67	5.91 ± 0.66
PE after atropine			
Mean ± SE	2.65 ± 0.04	2.56 ± 0.31	2.45 ± 0.16
P value	***	***	**
% Decrease	62.99	61.33	58.54
PE after Propranolol			
Mean ± SE	4.48 ± 0.36	4.09 ± 0.39	3.47 ± 0.36
P value	**	**	**
% Decrease	37.46	38.22	41.29

PE: Phenylephrine, SE: Standard error, **: Significant at 0.01 level of probability, ***: Significant at 0.001 level of probability

Tabel 5: Baroreflex slope (msec/mmHg) of adult male rabbits rendered hypertensive after elevation of blood pressure by phenylephrine (PE) injection

	Control	Hypertensive
Slope ±SE	7.16 ± 0.60	3.08 ± 0.21
P value		***
R	0.97	0.98
% Decrease		56.98

SE: Standard error
R: correlation coefficient
***: Significant at 0.001 level of probability

DISCUSSION

The results of this study demonstrate that aging in male rabbits is associated with preserved base line level of BP, HR, RR and VNA. While, the base line level of SNA is significantly increased.

The finding of maintained baseline levels of BP,HR and RR with advancing age is in agreement with the finding of previous studies in both animals (Wei *et al.,* 1986; Alberto *et al.,* 1991, Michael *et al.,* 1998 and Irigoyen *et al.,* 2000) and human (Lars, 1977, Ng *et al.,* 1995 and Davy *et al.,* 1998).

However, other studies in both animals and human found that the blood pressure and heart rate tend to be changed with advancing age. These studies reported a higher resting mean arterial blood pressure and heart rate in old beagles compared with the young (Hajduczok *et al.,* 1991), a higher mean ABP in the older than in the middle aged and young subjects (Ebert *et al.,* 1992), a higher baseline level of arterial blood pressure and a lower heart rate in older compared with young subjects (Kevin *et al.,* 1998) and a lower heart rate in old compared with young subjects (Vaz *et al.,* 2005).

The finding of maintained VNA in rabbits with increasing age in this study is consistent with that of Wei *et al.* (1986) in rats and Farrel *et al.* (1992) and La Rovere *et al.* (1995) in humans. However, the results of Ferrari *et al.* (1991) suggested that aging not only failed to impair but also actually enhanced cardiac muscarinic receptor responsiveness in rat. While in human, Ebert *et al.* (1992) found that VNA was progressively impaired with aging. The impaired cardiac response to carotid baroreceptor activation, mediated through the vagal nerve, in the study of Klaw *et al.* (2004) means the impaired vagal control of the heart function in the elderly and thus the greater probability of the occurrence of cardiac arrhythmias.

Increased basal SNA in this study with age is in agreement with the previous observations of Iwase *et al.* (1991) who found a significant positive correlation between age and resting muscle sympathetic nerve activity (MSNA) in human. In addition, Ebert *et al.* (1992), Davy *et al.* (1998), Kevin *et al.* (1998) and Jones *et al.* (2003) found that the baseline level of MSNA were higher in old compared with young men.

The mechanisms responsible for the increase in baseline level of SNA in aged rabbits have not been explored in this study. However, data obtained from normotensive animal and human studies showed that aging is accompanied by a modestly elevated plasma norepinephrine

concentration at rest, which may reflect: 1- an increase in tonic sympathetic nerve discharge as observed in this study, 2- an increased quantal transmitter release per impulse, or 3- reduced transmitter uptake at the nerve junction. Any of these possibilities will enhance the adrenergic effect on heart and blood vessels. This increase in neurotransmitter concentration is not necessarily translated into greater adrenergic responses. Factors such as adrenergic receptor subsensitivity or uncoupling could blunt end organ responsiveness in the elderly (Guraneri *et al.,* 1980; and Doecherty, 1990). Decreased number of adrenergic receptor could also contribute to this effect (Larkin *et al.,* 1996; and Xiao *et al.,* 1998). These mechanisms could, perhaps, be the basis for our observation that HR is nearly similar in both adult and old rabbits, and thus the normal expected enhancement effect of high baseline level of SNA on the HR is absent. However Klawe *et al.* (2004) suggested that the age-associated increased muscle SNA in their human study may result from impairment of arterial baroreceptors or from reduced arterial wall compliance. As a consequence, the decrease in afferent baroreflex neural activity leads to gradually increased baroreflex sympathetic outflow, manifested as increased muscle sympathetic nerve activity at rest.

To assess the relationship between aging and the changes in BP after phenylephrine injection and reflex changes in baroreceptor control, the data of the three age groups of rabbits were compared.

Phenylephrine injection produced an increase in BP and VNA responses and a decrease in HR, RR, and SNA responses in adult, middle aged and old rabbits. These changes were nearly similar in the three age groups studied. The data of the present study strength and extend the observations of two earlier investigations (Hosomi *et al.,* 1984; and Katsuda *et al.,* 1990) on aging and baroreflex function in rabbits. They suggested that the responsiveness of the baroreflex system was not affected by aging from 6 to 30 months.

In the Fischer-334 rat model of aging, which differ from human in that it is not complicated by hypertension or athrosclerosis, carotid sinus baroreflex function was well maintained in senescence (Wie *et al.,* 1986) and the reflex depression of adrenal sympathetic nerve activity to baroreceptor stimulation in anaesthetized rat was quite well preserved during aging (Kurosawa *et al.,* 1987). In addition, Ferrari *et al.* (1991) found that the baroreceptor control of heart rate and blood pressure in unanaesthetized, normotensive rat was preserved with aging, and Ng *et al.* (1995) found that increase in muscle sympathetic nerve activity in

response to upright sitting were not different in young and old subjects.

However, other studies revealed that baroreflex function is impaired with aging in rabbits (Frolkis *et al.,* 1975), in beagles (Hajduczok *et al.,* 1991), in rats (Irigoyen *et al.,* 2000 and Sakima *et al.,* 2005) and even more in human (Laitinen *et al.,* 1998, Labrova *et al.,* 2005, Mattace-Raso *et al.,* 2006 and Monahan, 2007). The studies of Jones *et al.* (2003) and Monahan (2007) reported that aging in human is associated with a marked reduction in baroreflex buffering which is related to an increase in basal sympathetic nerve activity and a reduction in systemic α_1-adrenergic vascular responsiveness. This discrepancy may be partially explained by species difference, age and strain difference in the animals used, as well as other methodological differences that may directly interfere with baroreflex function.

The result of this study concerning maintained baroreflex sensitivity with aging is in agreement with previous studies. Ebert *et al.* (1992) found that the sensitivity of baroreflex control of muscle sympathetic nerve activity (MSNA) was the same in old, middle aged and young subjects. In addition, Dawson *et al.* (1999) found that older subjects (more than 40 years) showed no age related decrease in BRS. However, Parati *et al.* (1995) and Piccirillo *et al.* (2001) demonstrated an age related decline in baroreflex sensitivity. Similarly, Dawson *et al.* (1999) found a nonlinear decline in cardiac baroreceptor sensitivity with advancing age in the third and fourth decades in human, but little further decline after the fourth decade. Age related physiological spectrally determined BRS decrease in a mid age healthy men (18-50 years) was found by Fauvel *et al.* (2007).

As clearly shown in this study by the results of baroreflex response to phenylephrine injection in adult male rabbits rendered hypertensive by intraperitoneal injection of epinephrine, the baroreflex sensitivity was reduced compared with normotensive adult male rabbits. Our results go in line with the impaired sensitivity of the cardiac baroreflex in beta adrenoceptor agonist-induced cardiac hypertrophy in mice which could be due to diminished vagal activity of the heart (Gava *et al.,* 2004). The impaired spontaneous BRS in Lyon hypertensive rats probably occurs not because of high BP levels but more likely because of either cardiac hypertrophy or a direct effect of angiotensin Π on the baroreflex loop as shown in the study of Lantelme *et al.* (1998). From a clinical perspective, they suggested that the BP-lowering effect of a drug is not sufficient to normalize the baroreflex function. Conversely, the magnitude of the regression of left ventricular hypertrophy might be

decisive. The results of this work are also consistent with that of the study of Souza *et al.* (2001) which showed that hypertension induced in rats by chronic administration of NO synthase inhibitor L-nitroarginine methyl ester (L-NAME) is assiocated with, decreased BRS, cardiac sympathetic overactivity and decreased HR variability. NO synthase inhibition in the central nervous system may increase the sympathetic activity which would be responsible, at least in part, for the increase in ABP occurring in (L-NAME) hypertensive model. Weinstock *et al.* (1984) tested the hypothesis that increased baroreflex sensitivity confers resistance to hypertension and concluded that a decrease in baroreflex control of the heart by aortic baroreceptor deafferentation can render salt-resistant Sebra strain rats sensitive to deoxycorticosterone acetate-salt-induced systolic hypertension. So, a derangement of the baroreflex arc or development of left ventricular hypertrophy are among the mechanisms that might explain the deficite in BRS observed in hypertensive rabbits in this work.

Microscopic examination, the aortas of adult male rabbits rendered hypertensive in this work showed thickened wall and eroded endothelial lining and irregularly arranged smooth muscle fibers and presence of foam cells in the intimal and subintimal layers. Fatty infiltration represents the initial lesion of athrosclerosis and could be a key role in the formation of atherosclerotic plaque in vivo (Zhang and Faber, 2001). Foam cells are cells filled with lipid droplets. Originally, the fat containing cells were thought to be macrophages but it has been established that many of them are modified smooth muscle cells (Rubin and Faber, 1994). Endothelial release of nitric oxide (NO) is impaired with aging, atherosclerosis, and diabetes (Barnett, 1991, Gerhard *et al.*, 1996, and Kinlay *et al.*, 2001). Reduced NO would not only produce vasoconstriction, which would reduce compliance, but would also facilitate vascular smooth muscle growth that would add a structural component to the increase in arterial stiffness. This may be an explanation of the observed thickening in the aortic wall of hypertensive rabbits in our study.

The results of this study demonstrated that the baroreflex gain was attenuated in the rabbit model of aging and rabbits rendered hypertensive or atherosclerotic.

Advanced age is known to be associated with attenuation of the baroreptor reflex function in human (MeGarry *et al.*, 1983, Wei, 1984, Laitinen *et al.*, 1998, Labrova *et al.*, 2005, Mattace-Raso *et al.*, 2006 and

Monahan, 2007). Arterial stiffness (mattace-Raso *et al.,* 2006) or thickening of the carotid wall (Labrova *et al.,* 2005) may explain at least in part, the reduced baroreflex observed in older adult men. It is possible that the age-related baroreflex attenuation that observed in human may not be due to aging perse but many actually reflect changes due to concomitant pathophysioloyical processes of athrosclerosis and / or hypertension. James *et al.* (1996) found that age contributed only to 7% of the variance in cardiac baroreflex sensitivity compared with 27.45% for systolic blood pressure in subjects over 60 years. It can be concluded that indices of baroreflex function and baroreflex sensitivity were relatively well maintained in old rabbits. Parasympathetic and sympathetic components of the ANS normally mediate the baroreflex in old age. Baroreflex sensitivity is depressed in adult rabbits rendered hypertensive and with early signs of atherosclerosis.

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Fig. 1: Transverse sections of the aortic wall of normotensive adult (A) and old (B) male rabbits show smooth and intact endothelial lining and regular arrangement of smooth muscle fibers. Aortas of adult rabbits rendered hypertensive (C) show thickened vessel wall. Magnified picture (D) shows eroded endothelial lining (short arrow). Foam cells in the intimal (long arrow) and subintimal layers (dashed arrow) and irregular arrangement of smooth muscle fibers (A, B and C: H and E X 40, D: H and E X 200).