TÜBİTAK

Turkish Journal of Medical Sciences

http://journals.tubitak.gov.tr/medical/

Research Article

Turk J Med Sci
(2019) 49: 1748-1753

doi:10.3906/sag-1908-137

Role of aortic stiffness and inflammation in the etiology of young-onset hypertension

Serkan GÖKASLAN[1,*], Çiğdem ÖZER GÖKASLAN[2], Emin DEMİREL[2], Sefa ÇELİK[3]

[1]Department of Cardiology, Faculty of Medicine, Afyonkarahisar University of Health Sciences, Afyonkarahisar, Turkey
[2]Department of Radiology, Faculty of Medicine, Afyonkarahisar University of Health Sciences, Afyonkarahisar, Turkey
[3]Department of Biochemistry, Faculty of Medicine, Afyonkarahisar University of Health Sciences, Afyonkarahisar, Turkey

Received: 28.08.2019 • **Accepted/Published Online:** 23.10.2019 • **Final Version:** 16.12.2019

Background/aim: Young-onset hypertension is a form of condition diagnosed in patients aged below 40. Cytokines such as interleukin (IL)-6 and also MCP-1 may play a role in the development of arterial hypertension. Aortic stiffness can be detected by measuring pulse wave velocity (PWV). We aimed to explore the relationship between inflammation and aortic stiffness and investigate their roles in the etiology of young-onset hypertension.

Materials and methods: We enrolled 16 patients diagnosed with young-onset hypertension and 16 volunteers without hypertension. The plasma levels of MCP-1 and IL-6 were determined using an enzyme-linked immunosorbent assay and quantitative enzyme-linked immunoassay, respectively. Carotid-femoral PWV was measured using an arteriograph device.

Results: Compared with those in normotensive controls, the plasma levels of IL-6 and MCP-1 and the PWV values were significantly higher in patients with young-onset hypertension ($P < 0.001$). PWV values were also positively correlated with the levels of MCP-1 and IL-6. However, no statistically significant difference was noted in intima-media thickness between the two groups ($P = 0.224$).

Conclusion: In this study, increased PWVs and the levels of inflammation markers were associated with aortic stiffness and inflammation in patients with young-onset hypertension, suggesting they have a role in the etiology of hypertension.

Keywords: Aortic stiffness, young-onset hypertension, inflammation markers

1. Introduction

Hypertension is a major cause of mortality worldwide [1]. Young-onset hypertension is a form of this condition diagnosed in patients aged below 40. In an epidemiological study, the incidence of young-onset hypertension was approximately 0.1%, and sex, genetics, and obesity were the risk factors [2]. The predominant form of hypertension in individuals aged below 50 is essential hypertension, while isolated systolic hypertension is more commonly seen among elderly individuals [3,4].

Experimental evidence suggests that hypertension functions to aggravate inflammation by promoting the expression of cytokines [5,6]. A study conducted on healthy men demonstrated a correlation between increased blood pressure (hypertension) and elevated levels of circulating interleukin (IL)-6 [7]. Increase in IL-6 was also reported in response to the infusion of angiotensin II in humans, demonstrating a direct relationship between the two [8,9]. In addition to IL-6, MCP-1, a key chemokine involved in the onset of inflammation, may play a role not only in various pathophysiological processes occurring in the cardiovascular system but also in the development of arterial hypertension [10,11]. Vascular endothelial cells stimulate the expression of MCP-1 by mediating the inflammatory cytokines IL-1, IL-4, IL-6, and tumor necrosis factor-α [12].

Aortic stiffness contributes to vascular diseases by inducing vascular strain and endothelial dysfunction. Moreover, pulse wave velocity (PWV) has been widely accepted for diagnosing aortic stiffness in clinical practices [13–15]. Carotid-femoral PWV is recommended by the European Society of Hypertension, the European Society of Cardiology, and the American Heart Association as a clinical marker for the classification of cardiovascular risk in patients with hypertension [16–18]. The carotid intima-media thickness (IMT) is also widely used for detecting early atherosclerosis and is associated with increased cardiovascular risk factors, such as hypertension, diabetes mellitus, hypercholesterolemia, obesity, and coronary artery disease [19–22].

However, to date, the etiology of young-onset hypertension has been unclear. In this study, we aimed

* Correspondence: serkangokaslan6@gmail.com

to explore the relationship between inflammation and aortic stiffness and investigate their roles in the etiology of young-onset hypertension.

2. Materials and methods

2.1. Patients

Between December 2017 and May 2018, 16 patients diagnosed with young-onset hypertension (patient group) and 16 volunteers (control group) without hypertension (all aged below 40) were included in the study. Their sedimentation rates, C-reactive protein levels, and white blood cell levels were within normal limits. Patients with rheumatic disease, hyperlipidemia, and acute or chronic infections were excluded. Both groups were matched for age and sex. Informed consent was obtained from all participants, and the study was approved by the ethics committee of the Faculty of Medicine of Afyonkarahisar University of Health Sciences.

2.2. Enzyme-linked immunosorbent assay (ELISA)

The plasma levels of MCP-1 were determined using an ELISA kit (BMS281TEN; eBioscience, Vienna, Austria) as per the manufacturer's recommendations. Absorbance was measured at 450 nm using a spectrophotometer (BioTek, Epoch Microplate Spectrophotometer, Winooski, VT, USA). The limit of detection and coefficient of interassay variation were 2.31 pg/mL and 8.7%, respectively. The level of IL-6 was measured using a quantitative enzyme-linked immunoassay kit (Elabscience, Houston, TX, USA).

2.3. Determination of carotid-femoral PWV

Carotid-femoral PWV was measured using an arteriograph device (TensioMed, Budapest, Hungary). The pulse waveforms of the common carotid and femoral arteries were sequentially acquired. Subsequently, for determining PWV, the distance from the suprasternal notch to the femoral artery was divided by the time interval between the waves.

2.4. IMT measurements

The IMT of the right and left common carotid arteries was measured from the distant walls based on the Mannheim carotid IMT consensus [23]. An Aplio MX duplex Doppler ultrasonography device (Toshiba, Otawara, Tochigi, Japan) and a 7.5-MHz probe were used to obtain images of the intima media of the carotid artery. The best image was acquired when the patient was in the supine position. After obtaining the mean and maximum IMTs of the distal walls of the right and left arteries at 1–2 cm proximally to the bulb, the average of the mean values of the two measurements was calculated.

2.5. Statistical analysis

All data were analyzed using SPSS 21.0 (IBM Corp., Armonk, NY, USA). Comparison of the categorical and continuous variables was performed using the chi-square and Mann–Whitney U tests, respectively. The association between MCP-1, IL-6, and PWV was investigated with Spearman's correlation coefficient. An a priori power analysis could not be conducted owing to the unavailability of pilot data. Therefore, a post hoc power analysis was conducted to confirm that the current study had adequate power (99%).

3. Results

Participant demographics are shown in Table 1. Among 16 patients with young-onset hypertension who participated in the study, nine were males and seven were females, with a mean age of 31.6 (range: 18–40) years. The control group comprised 16 individuals, eight males and eight females, with a mean age of 29.9 (range: 18–39) years. The body mass index (BMI) and the mean levels of low-density lipoprotein (LDL) and triglycerides of the two groups are shown in Table 1. There were no significant differences in any of the variables examined between the patient and the control groups.

The mean level of IL-6 significantly differed between the patient (152.45 ± 80.93; range: 52.96–317.08 pg/mL) and the control groups (5.73 ± 5.94; range: 1.02–25.53 pg/mL; $P < 0.001$). The level of MCP-1 was significantly higher in the patient group (293.15 ± 148.76; range: 34.55–614.63 pg/mL) than that in the control group (13.49 ± 41.38; range: 0.39 ± 168.42 pg/mL; $P < 0.001$).

Mean PWV in the patient group (11.22 ± 2.54; range: 7.5–16 m/s) was significantly higher than that of the control group (7.36 ± 1.86; range: 4.6–10.2 m/s; $P < 0.001$). PWV was positively correlated with the levels of MCP-1 and IL-6. PWV level was positively correlated with IL-6 ($r = 0.656$, $P < 0.001$) and MCP-1 ($r = 0.614$, $P < 0.001$).

Mean IMT in the patient group (0.57 ± 0.14; range: 0.3–0.9 mm) was not significantly different from that in the control group (0.5 ± 0.1; range: 0.3–0.7 mm; $P = 0.224$) (Table 2).

4. Discussion

To the best of our knowledge, this is the first study to demonstrate that patients with young-onset hypertension have significantly higher levels of plasma MCP-1 and IL-6 and aortic stiffness than normotensive individuals. These results support the use of IL-6 and MCP-1 as the biological markers of vascular impairment.

The earlier the onset of hypertension is, the longer the exposure time to the disease and the higher the risk of cardiovascular events will be. Therefore, appropriately identifying and treating the underlying pathogenesis of young-onset hypertension is important to prevent cardiovascular events. However, the literature on this subject is not sufficient. A study involving young adults demonstrated that the main hemodynamic abnormality

Table 1. The demographic features of patients with young-onset hypertension and normotensive controls.

	Patients (n = 16) (mean ± SD)	Controls (n = 16) (mean ± SD)	P-value
Age (mean)	31.6 ± 8.5	29.9 ± 5.9	0.642
Sex (male)	9 (56%)	8 (50%)	0.469
BMI (kg/m^2)	27.49 ± 3.51	24.61 ± 4.36	0.094
LDL (mg/dL)	95.8 ± 18.4	97.2 ± 20.6	0.128
Triglyceride (mg/dL)	183.2 ± 25.6	178.5 ± 23.2	0.076

Table 2. Mean levels of IL-6, MCP-1, PWV, and IMT of patients with young-onset hypertension and normotensive controls.

	Patients (mean ± SD)	Controls (mean ± SD)	P-value
IL-6 (pg/mL)	152.45 ± 80.93	5.73 ± 5.94	<0.001
MCP-1(pg/mL)	293.15 ± 148.76	13.49 ± 41.38	<0.001
PWV (m/s)	11.22 ± 2.54	7.36 ± 1.86	<0.001
IMT (mm)	0.57 ± 0.14	0.5 ± 0.1	0.224

underlying essential hypertension may be increased peripheral vascular resistance, which causes vascular remodeling in the arteries, involving extracellular matrix deposition and inflammation [24].

Previous studies showed that vascular inflammation caused vascular damage and played a key role in the pathogenesis and progression of hypertension [25–27]. However, the role of inflammatory cytokines in the mechanism and progression of hypertension remains unclear. The stimulation of human vascular smooth muscle cells by angiotensin II, the main regulator of blood pressure, resulted in increased expression and release of IL-6 [28–30]. IL-6 increased vascular smooth muscle cell proliferation, which is a characteristic of the early stages of hypertension [31]. Additionally, MCP-1 also contributed to the onset of inflammation by promoting the uptake of inflammatory cells into the vessel wall [32]. Furthermore, increased blood vessel tension due to high blood pressure increased the expression of MCP-1 mRNA in human vascular endothelial cells, which further potentiated the secretion of MCP-1 [33,34]. Thus, in young-onset hypertension, elevated levels of MCP-1 and IL-6 in the vascular endothelium may indicate the stimulation of cellular immunological processes that contribute to early vascular aging and the development of hypertension [35,36].

The present study provides evidence regarding high levels of IL-6 and MCP-1 and a predisposition to inflammation in patients with young-onset hypertension compared with those in normotensive individuals. These mechanisms may partly explain the relationship observed between increased levels of blood pressure, MCP-1, and IL-6, suggesting that IL-6 and MCP-1 may serve as the biological markers of vascular impairment.

Another known independent predictor of cardiovascular disease is increased aortic stiffness, which is considered to be an important cardiovascular risk factor [37,38]. Although increased aortic stiffness can be determined by measuring pulse waves, the latter is an independent predictor of poor cardiovascular outcomes in patients with essential hypertension [24,39]. A metaanalysis found that a 1-m/s increase in PWV was associated with an 11% increase in cardiovascular deaths [37].

Carotid-femoral and brachial-ankle PWVs are the two most commonly used PWV measurements. Brachial-ankle PWV calculation involves both elastic and muscular arteries, and is considered to be a predictor of aortic stiffness. Conversely, carotid-femoral PWV, which only involves measurement of the elastic artery, is accepted as a better indicator [17]. Aortic PWV increases with age and accelerates with the presence and severity of hypertension [40,41]. In a study using carotid-femoral PWVs, hypertension was associated with the progression of aortic stiffness in young patients compared with those in normotensive subjects [42]. The findings of this

study are in concordance with those in the literature. Carotid-femoral PWV increased in patients with young-onset hypertension compared with those in the control group. This suggests that in addition to predisposition to inflammation, increased aortic stiffness contributes to the etiology of young-onset hypertension. Furthermore, the positive relationship between PWVs and the levels of MCP-1 and IL-6 indicates that aortic stiffness may play a role in the etiology of young-onset hypertension.

Most young individuals with hypertension show early vascular changes despite short-term exposure to the disease [43]. As increased carotid IMT is strongly associated with the early stages of vascular atherosclerosis in young adults, this measure can be used to evaluate early atherosclerosis and predict cardiovascular events [43]. However, in this study, we found no significant increase in carotid IMT or the evidence of early atherosclerosis. Reportedly, an increase in carotid IMT is adaptive to medial hypertrophy rather than intimal hypertrophy in young patients with hypertension [43–45]. In our study, the lack of difference in the carotid IMTs in the patient group compared with the difference observed in the control group may be attributed to the small media of the carotid artery and the inability to radiologically detect medial hypertrophy [46,47]. In addition, the patients included in this study were young; accordingly, atherosclerosis is not expected in this age group, which may be the reason why no significant difference was observed between the two groups regarding IMT. However, this was strictly a clinical study, and additional histopathological studies are warranted to confirm the findings. The other limitation of this study was that the patient group was not compared with hypertensive patients over 40 years of age. Such a comparison could increase the effectiveness of the study.

In conclusion, increased PWV and the levels of inflammatory markers were associated with aortic stiffness and inflammation in patients with young-onset hypertension, suggesting that these factors have a role in the etiology of hypertension.

References

1. Alwan A. Global Status Report on Noncommunicable Diseases 2010. Geneva, Switzerland: World Health Organization; 2011.
2. Ejima Y, Hasegawa Y, Sanada S, Miyama N, Hatano R et al. Characteristics of young-onset hypertension identified by targeted screening performed at a university health check-up. Hypertension Research 2006; 29 (4): 261-267
3. Burt VL, Whelton P, Roccella EJ, Brown C, Cutler JA et al. Prevalence of hypertension in the US adult population. Results from the Third National Health and Nutrition Examination Survey, 1988–1991. Hypertension 1995; 25 (3): 305-313.
4. Colhoun HM, Dong W, Poulter NR. Blood pressure screening, management and control in England: results from the health survey for England 1994. Journal of Hypertension 1998; 16 (6): 747-752.
5. McCarron RM, Wang L, Siren AL, Spatz M, Hallenback JM. Monocyte adhesion to cerebromicrovascular endothelial cells derived from hypertensive and normotensive rats. American Journal of Physiology 1994; 267 (6 Pt 2): H2491-H2497.
6. Liu Y, Liu T, McCarron RM, Spatz M, Feuerstein G et al. Evidence for activation of endothelium and monocytes in hypertensive rats. American Journal of Physiology 1996; 270 (6 Pt 2): H2125-31.
7. Chae CU, Lee RT, Rifai N, Ridker PM. Blood pressure and inflammation in apparently healthy men. Hypertension 2001; 38 (3): 399-403.
8. Luther JM, Gainer JV, Murphey LJ, Yu C, Vaughan DE et al. Angiotensin II induces interleukin-6 in humans through a mineralocorticoid receptor-dependent mechanism. Hypertension 2006; 48 (6): 1050-1057.
9. Chamarthi B, Williams GH, Ricchiuti V, Srikumar N, Hopkins PN et al. Inflammation and hypertension: the interplay of interleukin-6, dietary sodium, and the renin-angiotensin system in humans. American Journal of Hypertension 2011; 24 (10): 1143-1148.
10. O'Hayre M, Salanga CL, Handel TM, Allen SJ. Chemokines and cancer: migration, intracellular signalling and intercellular communication in the microenvironment. Biochemical Journal 2008; 409 (3): 635-649.
11. Stumpf C, Raaz D, Klinghammer L, Schneider M, Schmieder RE et al. Platelet CD40 contributes to enhanced monocyte chemoattractant protein 1 levels in patients with resistant hypertension. European Journal of Clinical Investigation 2016; 46 (6): 564-571.
12. Hansson GK, Libby P. The immune response in atherosclerosis: a double-edged sword. Nature Reviews Immunology 2006; 6 (7): 508-519.
13. Zhang Y, Agnoletti D, Protogerou AD, Topouchian J, Wang JG et al. Characteristics of pulse wave velocity in elastic and muscular arteries: a mismatch beyond age. Journal of Hypertension 2013; 31 (3): 554-559.
14. Yu WC, Chuang SY, Lin YP, Chen CH. Brachial-ankle vs carotid-femoral pulse wave velocity as a determinant of cardiovascular structure and function. Journal of Human Hypertension 2008; 22 (1): 24-31.
15. Kass DA. Ventricular arterial stiffening: integrating the pathophysiology. Hypertension 2005; 46 (1): 185-193.

16. Mancia G, Fagard R, Narkiewicz K, Redon J, Zanchetti A et al. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the management of arterial hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Journal of Hypertension 2013; 31 (7): 1281-1357.

17. Zhang Y, Agnoletti D, Xu Y, Wang JG, Blacher J et al. Carotid-femoral pulse wave velocity in the elderly. Journal of Hypertension 2014; 32 (8): 1572-1576.

18. Townsend RR, Wilkinson IB, Schiffrin EL, Avolio AP, Chirinos JA et al. Recommendations for improving and standardizing vascular research on arterial stiffness: a scientific statement from the American Heart Association. Hypertension 2015; 66 (3): 698-722.

19. Nambi V, Chambless L, Folsom AR, He M, Hu Y et al. Carotid intima-media thickness and presence or absence of plaque improves prediction of coronary heart disease risk: The ARIC (Atherosclerosis Risk In Communities) study. Journal of the American College of Cardiology 2010; 55 (15): 1600-1607.

20. Nichols WW, Pepine CJ, O'Rourke MF. Carotid-artery intima and media thickness as a risk factor for myocardial infarction and stroke. New England Journal of Medicine 1999; 340 (22): 1762-1763.

21. Ikeda N, Kogame N, Iijima R, Nakamura M, Sugi K. Impact of carotid artery ultrasound and ankle-brachial index on prediction of severity of SYNTAX score. Circulation Journal 2013; 77 (3): 712-716.

22. Cheng KS, Mikhailidis DP, Hamilton G, Seifalian AM. A review of the carotid and femoral intima-media thickness as an indicator of the presence of peripheral vascular disease and cardiovascular risk factors. Cardiovascular Research 2002; 54 (3): 528-538.

23. Touboul PJ, Hennerici MG, Meairs S, Adams H, Amarenco P et al. Mannheim carotid intima-media thickness consensus (2004-2006). An update on behalf of the Advisory Board of the 3rd and 4th Watching the Risk Symposium, 13th and 15th European Stroke Conferences, Mannheim, Germany, 2004, and Brussels, Belgium, 2006. Cerebrovascular Diseases 2007; 23 (1): 75-80.

24. Laurent S, Boutouyrie P, Asmar R, Gautier I, Laloux B et al. Aortic stiffness is an independent predictor of all-cause and cardiovascular mortality in hypertensive patients. Hypertension 2001; 37 (5): 1236-1241.

25. Virdis A, Schiffrin EL. Vascular inflammation: a role in vascular disease in hypertension? Current Opinion in Nephrology and Hypertension 2003;12 (2): 181-187.

26. Hilgers KF. Monocytes/macrophages in hypertension. Journal of Hypertension 2002; 20 (4): 593-596.

27. Li JJ, Fang CH, Huio RT. Is hypertension an inflammatory disease? Medical Hypotheses 2005; 64 (2): 236-240.

28. Kranzhöfer R, Schmidt J, Pfeiffer CAH, Hagl S, Libby P et al. Angiotensin induces inflammatory activation of human vascular smooth muscle cells. Arteriosclerosis, Thrombosis, and Vascular Biology 1999; 19 (7): 1623-1629.

29. Schieffer B, Schieffer E, Hilfiker-Kleiner D, Hilfiker A, Kovanen PT et al. Expression of angiotensin II and interleukin 6 in human coronary atherosclerotic plaques: potential implications for inflammation and plaque instability. Circulation 2000; 101 (12): 1372-1378.

30. Lee DL, Sturgis LC, Labazi H, Osborne JB Jr, Fleming C et al. Angiotensin II hypertension is attenuated in interleukin-6 knock out mice. American Journal of Physiology Heart and Circulatory Physiology 2006; 290 (3): H935-940.

31. Ikeda U, Ikeda M, Oohara T, Ohuchi A, Kamitani T et al. Interleukin 6 stimulates growth of vascular smooth muscle cells in a PDGF-dependent manner. American Journal of Physiology 1991; 260 (Pt 2): H1713-1717.

32. Rossi D, Zlotnik A. The biology of chemokines and their receptors. Annual Review of Immunology 2000; 18: 217-242.

33. Okada M, Matsumori A, Ono K, Furukawa Y, Shioi T et al. Cyclic stretch upregulates production of interleukin-8 and monocyte chemotactic and activating factor/monocyte chemoattractant protein-1 in human endothelial cells. Arteriosclerosis, Thrombosis, and Vascular Biology 1998; 18 (6): 894-901.

34. Wung BS, Cheng JJ, Chao YJ, Lin J, Shyy YJ et al. Cyclical strain increases monocyte chemotactic protein-1 secretion in human endothelial cells. American Journal of Physiology 1996; 270 (4 Pt 2): H1462-1468.

35. Sheikine Y, Hansson GK. Chemokines and atherosclerosis. Annals of Medicine 2004; 36 (2): 98-118.

36. Schutte AE, Schutte R, Huisman HW, van Rooyen JM, Fourie CM et al. Are behavioural risk factors to be blamed for the conversion from optimal blood pressure to hypertensive status in Black South Africans? A 5-year prospective study. International Journal of Epidemiology 2012; 41 (4): 1114-1123.

37. Vlachopoulos C, Aznaouridis K, Stefanadis C. Prediction of cardiovascular events and all-cause mortality with arterial stiffness: a systematic review and meta-analysis. Journal of the American College of Cardiology 2010; 55 (13): 1318-1327.

38. Ben-Shlomo Y, Spears M, Boustred C, May M, Anderson SG et al. Aortic pulse wave velocity improves cardiovascular event prediction: an individual participant meta-analysis of prospective observational data from 17,635 subjects. Journal of the American College of Cardiology 2014; 63 (7): 636-646.

39. Blacher J, Asmar R, Djane S, London GM, Safar ME. Aortic pulse wave velocity as a marker of cardiovascular risk in hypertensive patients. Hypertension 1999; 33(5): 1111-1117.

40. Mitchell GF, Parise H, Benjamin EJ, Larson MG, Keyes MJ et al. Changes in arterial stiffness and wave reflection with advancing age in healthy men and women: the Framingham Heart Study. Hypertension 2004; 43 (6): 1239-1245.

41. Tomiyama H, Arai T, Koji Y, Yambe M, Motobe K et al. The age-related increase in arterial stiffness is augmented in phases according to the severity of hypertension. Hypertension Research 2004; 27 (7): 465-470.

42. Benetos A, Asamopouolos C, Bureau JM, Temmar M, Labat C et al. Determinants of accelerated progression of arterial stiffness in normotensive subjects and in treated hypertensive subjects over a 6-year period. Circulation 2002; 105 (10): 1202-1207.

43. Stein JH, Korcarz CE, Hurst RT, Lonn E, Kendall CB et al. Use of carotid ultrasound to identify subclinical vascular disease and evaluate cardiovascular disease risk: a consensus statement from the American Society of Echocardiography Carotid Intima-Media Thickness Task Force. Endorsed by the Society for Vascular Medicine. Journal of the American Society of Echocardiography 2008; 21 (2): 93-111.

44. Mancini GB, Dahlof B, Diez J. Surrogate markers for cardiovascular disease: structural markers. Circulation 2004; 109 (25 Suppl 1): IV22-30.

45. Roman MJ, Saba PS, Pini R, Spitzer M, Pickering TG et al. Parallel cardiac and vascular adaptation in hypertension. Circulation 1992; 86 (6): 1909-1918.

46. Grobbee DE, Bots ML. Carotid artery intima-media thickness as an indicator of generalized atherosclerosis. Journal of Internal Medicine 1994; 236 (5): 567-573.

47. Van Bortel LM. What does intima-media thickness tell us? Journal of Hypertension 2005; 23 (1): 37-39.