Ethyl acetate fraction of *Allium hirtifolium* improves functional parameters of isolated hearts of diabetic rats

Sara Khaleghi, Mahvash Hesari, Aliashraf Godini, Dareuosh Shacklebæi, Ali Mostafaie

Medical Biology Research Center, Kermanshah University of Medical Sciences; Kermanshah-Iran

ABSTRACT

**Objective:** *Allium hirtifolium* (Persian shallot) has a hypoglycemic effect on diabetic animals. The aim of this study was to assess the effect of the ethyl acetate fraction of *Allium hirtifolium* on the function of isolated hearts of diabetic rats.

**Methods:** The control and diabetic animals were randomly divided into four groups: saline- or extract-treated controls (n=10 and n=6, respectively) and saline- or extract-treated diabetic rats (n=8 and n=9, respectively), which received normal saline or extract for four weeks by daily gavage. The hearts were perfused according to the Langendorff method. Cardiac function parameters, including left ventricular developed pressure (LVDP), heart rate (HR), rate pressure product (RPP; LVDP×HR), and dp/dt were measured.

**Results:** The findings of this study showed that in the extract-treated diabetic rats, LVDP (94.5±9.1 mm Hg, mean±SEM), HR (249±15 beats/min), RPP (22732±1246) and +dp/dt (2598±230) at the baseline were significantly higher than those in the saline-treated diabetic animals, (71.5±4.0), (189±6), (13923±984), and (1701±124), respectively. Furthermore, RPP and HR were also significantly higher than the corresponding values obtained in the saline-treated diabetic rats after ischemia.

**Conclusion:** Besides blood glucose lowering action, oral administration of the ethyl acetate fraction of *Allium hirtifolium* significantly improved the baseline and post-ischemic cardiac function parameters in streptozotocin-induced diabetic rats. (Anatol J Cardiol 2017; 17: 452-9)

**Keywords:** diabetes, *Allium hirtifolium*, isolated heart, cardiac function

Introduction

Diabetes mellitus is a common disorder and an important risk factor for cardiovascular diseases (1). Diabetic patients are more prone to heart diseases, including myocardial infarction and severe ventricular arrhythmias (2). Owing to the effectiveness of herbal drugs and their low cost, they are widely prescribed, even when their biologically active constituents are not fully identified (3). *Allium hirtifolium* (Persian shallot) is a nutritious plant that belongs to the Alliaceae family (4). Based on available pharmaceutical investigations, the antioxidant and hepatoprotective effects of *Allium hirtifolium* have been demonstrated. In addition, *Allium hirtifolium* extract has antioxidant properties comparable to or slightly higher than garlic extract (5). The commonly known phytochemical compounds identified in *Allium hirtifolium* are saponins, sapogenins, and flavonoids, including shallomin, quercetin, and kaempferol (5).

Certain flavonoids in the diet can have significant cardiovascular benefits (6). *Allium hirtifolium* with a high content of flavonoids and phenolic components changes the postprandial serum lipid profile, endothelial markers, and thrombogenic factors and might be beneficial in patients with cardiovascular diseases (7). The Persian shallot significantly reduces blood glucose levels in diabetic rats (8), and aqueous shallot extract has a hypoglycemic effect on fructose-induced insulin resistance animals (9). No data is available regarding the cardiovascular effects of *Allium hirtifolium*. Given that the ethyl acetate fraction of shallot has the highest flavonoid content (38% of total flavonoid) compared with other fractions (10, 11), this study was conducted to assess the effect of the ethyl acetate fraction of *Allium hirtifolium* on the pre- and post-ischemic function of isolated hearts of diabetic rats.

**Methods**

This experimental animal study was designed to evaluate possible cardiac protective effects of the ethyl acetate fraction of *Allium hirtifolium* on streptozotocin (STZ)-induced diabetes in rats.
Drugs and chemicals
Ethanol, methanol, ethyl acetate, butanol, hexane and STZ were purchased from Sigma Chemicals (St. Louis, Mo., USA). All other chemicals were purchased from Merck (Darmstadt, Germany).

Animals
All experiments were approved by the Ethics Committee of the Kermanshah University of Medical Sciences. The study was supported by Medical Biology Research Center, Kermanshah, Iran. Male Wistar rats weighing 200–250 g were housed three per cage at the constant temperature of 22°C–24°C, with a 12-hour light/dark cycle and relative air humidity of 40%–60%. The rats fed on standard pellet chow and water ad libitum. All the animals used in the present study received humane care in compliance with the institutional animal care guidelines.

Experimental protocols and animal grouping
Initially, the animals were randomly divided into two groups of diabetic and control rats. Diabetes was induced via a single intraperitoneal injection of STZ (60 mg kg⁻¹ body weight) dissolved in 0.1 M cold citrate buffer (pH 4.5) (12). The control rats were just injected with citrate buffer. Five days after STZ or citrate buffer injection, blood samples were taken from the retro-orbital plexus of the median canthus under ether anesthesia (13, 14). Serum glucose level was measured using a glucose oxidase method (glucose oxidase kit, Zist Chimi, Tehran, Iran). The rats with fasting serum glucose concentration higher than 200 mg dL⁻¹ were considered diabetic. Afterwards, the diabetic rats were randomly allocated into two groups: 1) the saline-treated diabetic rats (n=8) and 2) the extract-treated diabetic rats (n=9) that received normal saline or ethyl acetate fraction of shallot extract (5 mg kg⁻¹ body weight), respectively, for four weeks via daily gavage.

The control rats were also randomly allocated into two groups: 1) the saline-treated control rats (n=10) and 2) the extract-treated control rats (n=6) that received normal saline or the ethyl acetate fraction of the shallot extract (5 mg kg⁻¹ body weight), respectively, for four weeks via daily gavage.

The body weight and serum glucose levels were measured three times throughout this four-week period: 1) when the rats were allocated into the groups (the initial day of the experiment), 2) after two weeks of the treatment with the ethyl acetate fraction of shallot extract or normal saline, and 3) after four weeks of the treatment period (at the time of heart isolation).

Isolated rat heart preparation
Four weeks after treatment with the ethyl acetate fraction of shallot extract or normal saline through gavage, the animals were anesthetized by intraperitoneal administration of pentobarbital sodium (60 mg kg⁻¹, Sigma). The hearts were quickly removed and soaked in Krebs solution at 4°C. The aortic root was cannulated and retrogradely perfused in accordance with the Langendorff method using Krebs solution (pH 7.4) containing the following: NaCl (118 mM), NaHCO₃ (25 mM), KCl (4.8 mM), KH₂PO₄ (1.2 mM), MgSO₄ (1.2 mM), glucose (11 mM), and CaCl₂ (1.2 mM). The buffer was continuously bubbled with a mixture of 95% O₂ and 5% CO₂ at 37°C. Perfusion was performed under a constant hydrostatic pressure of 85 cm H₂O. A latex balloon was placed in the left ventricle through the mitral valve. This balloon was filled with distilled water and connected to a pressure transducer (MLT 844; AD Instruments, New South Wales, Australia) through a rigid polyethylene tube. For continuous monitoring of cardiac function parameters, the pressure transducer was connected to a computer via a power lab (model ML825; AD Instruments). At the beginning of the experiment, the balloon volume was adjusted to produce an end diastolic pressure of 5–10 mm Hg. This volume was then kept constant during the experiment. The indices of myocardial function included left ventricular developed pressure (LVDP), which was defined as peak systolic pressure minus end diastolic pressure, heart rate (HR; beats/minute), and rate pressure product (RPP=LVDP×HR). Coronary flow (CF) was measured through volumetric collection of the coronary effluent per minute. Cardiac parameters were measured after the 20 min stabilization period. After stabilization, global normothermic ischemia was induced by halting perfusion and immersing the heart in Krebs buffer at 37°C for 40 minutes. Following ischemia, the hearts were reperfused for 45 minutes (15).

Plant material and extraction
Allium hirtifolium (Persian Shallot) bulbs were purchased from a local vegetable market in Kermanshah, Iran. The bulbs were cleaned and shed dried at 25°C, and the dried material was ground in a blender. The powder was kept at −20°C until the time of experiments.

Preparation of ethyl acetate fraction was performed by successive fractionation (10, 11). Briefly, the homogenate of Persian shallot powder (100 g) was extracted with 50% (v/v) ethanol, stirring for 24 h. The extract was filtered, then centrifuged at 12000 ×g for 20 min at 4°C and evaporated under reduced pressure to dryness. For solvent fractionation, the extract was resuspended in distilled water and then partitioned successively with n-hexane (Hex), ethyl acetate (EA), and n-butanol (BuOH), leaving a residual aqueous fraction (Aq). Each fraction was evaporated under reduced pressure to yield Hex (0.35%), EA (0.45%), BuOH (8.7%), and Aq fractions (90.5%), respectively. Ethyl acetate fraction was used to treat the animals.

Statistics
All data are expressed as mean±SEM (standard error of mean). To test normality of distributions, the Kolmogorov–Smirnov test was applied. Regarding the normal distribution of the data, independent samples t-test was used to evaluate the significant difference between the two sets of data obtained from the independent groups. The plasma glucose concentration and body weight were analyzed using repeated measures ANOVA followed by Bonferroni as a post-test. One-way ANOVA
was applied to analyze a set of independent groups and Tukey posthoc test was used for multiple comparisons as offered by GraphPad InStat version 3.0 (GraphPad Software Inc., La Jolla, CA, USA). Differences with a p value of <0.05 were considered statistically significant.

**Results**

**Body weight**

The weights of the animals during four weeks of the treatment with the ethyl acetate fraction of shallot extract or normal saline are illustrated in Figure 1a, b. Repeated measures ANOVA on the data in each group revealed that the body weight in the saline- and extract-treated control rats gradually increased throughout the experiment period, whereas in the saline- and extract-treated diabetic rats this parameter significantly changed in the opposite direction (Fig. 1a).

One-way ANOVA analysis on the data showed that body weights of the different groups were the same in the initial day of the experiment while at the end of the second and fourth weeks this parameter was significantly lower in the saline-treated diabetic rats compared to the saline-treated control rats ($p<0.01$ and $p<0.001$, respectively). The results also indicated that four weeks extract administration in the diabetic rats, significantly improved weight gain in comparison to the values in the saline-treated diabetic rats ($p<0.01$) although their average body weights were still lower than those of the saline- and extract-treated controls (Fig. 1b).

**Serum glucose concentration**

Serum glucose concentrations of the animals in the initial day and two and four weeks later are demonstrated in Figure 2a, b. The analysis of serum glucose concentrations with repeated measures ANOVA revealed that in the saline-treated control group the glucose concentration significantly increased in comparison to the initial day ($p<0.05$ or $p<0.01$) while in the saline-treated diabetic rats this parameter was fairly constant throughout the experiment period ($p>0.05$) and significantly lower than those of the saline- and extract-treated controls (Fig. 2b).
Figure 3. Typical traces of PowerLab-recorded left ventricular developed pressure before and after exposure to 40 minute global ischemia in the four experimental groups
treated control rats this parameter was constant throughout the experiment period. On the other hand, after four weeks of diabetes induction, serum glucose levels in the saline-treated diabetic rats significantly increased in comparison to the initial day. On the contrary, it significantly decreased throughout the experiment period in the extract-treated diabetic and control rats (Fig. 2a).

One-way ANOVA analysis of the data showed that diabetes in the saline-treated diabetic group resulted in a significant elevation of serum glucose concentration compared to the saline- and extract-treated controls. Treatment with shallot extract in the extract-treated diabetic rats decreased the serum glucose levels after two and four weeks of the experiment compared to those in the saline-treated diabetic rats (p<0.001). However, serum glucose concentrations were still significantly higher than those in the saline- and extract-treated control rats (Fig. 2b).

Cardiac function parameters
The typical traces of left ventricular developed pressure recorded in this study are illustrated in Figure 3. The values of cardiac function parameters, including RPP, LVDP, HR, dp/dt, and CF, at the 20th minute of baseline and 45th minute of the reperfusion period are shown in Table 1.

Baseline
The results of a one-way ANOVA test in the isolated heart experiments showed that the baseline values of cardiac function parameters, including RPP, HR, and CF, in the saline-treated diabetic rats were significantly lower than the corresponding values in the saline- and extract-treated controls. Furthermore, according to independent samples t-test, +dp/dt in the saline-treated diabetic group was significantly lower than that in the saline-treated control group (p<0.05). On the other hand, the baseline values of all cardiac function parameters except CF in the extract-treated diabetic rats did not differ significantly compared to the corresponding values in the extract- and saline-treated controls. In other words, there were no significant differences between the baseline values of these three groups before ischemia. As it is shown in Table 1, the values of RPP HR, and +dp/dt in extract-treated diabetic rats are significantly higher than the corresponding values in the saline-treated diabetic animals (one-way ANOVA).

Reperfusion
As illustrated in Table 1, the values of RPP and HR in the saline-treated diabetic at the 45th minute of the reperfusion period are significantly lower than the corresponding values in the saline- and extract-treated controls, whereas the values of RPP LVDP and HR in the extract-treated diabetic rats did not significantly differ from the corresponding values in the saline- and extract-treated controls. The RPP and HR values of the isolated hearts of the extract-treated diabetic rats are significantly higher than the corresponding values in the saline-treated diabetic rats. In addition, the results of independent samples t-test revealed that LVDP in these two groups was significantly different (p<0.05).

Discussion
The results of this study demonstrated that, in addition to lowering the blood glucose level, oral administration of the ethyl acetate fraction of Allium hirtifolium significantly improved the
baseline and post-ischemic cardiac function parameters deteriorated in the STZ-induced diabetic rats.

Considering the fact that diabetes mellitus can result in cardiac function disturbances, we aimed to assess the effect of the ethyl acetate fraction of *Allium hirtifolium* on the function of isolated hearts of diabetic rats in the normal and post-ischemia conditions.

Mahmoodi et al. (8) have shown that oral administration of hydroalcoholic extract of *Allium hirtifolium* markedly enhances the GCK mRNA expression in the rat liver and consequently decreases blood glucose. In our study, we used the ethyl acetate fraction of *Allium hirtifolium*, and a prominent hypoglycemic effect was reached through using much lower doses (5 mg/kg) compared to Mahmoodi’s study (100 and 200 mg/kg), indicating higher purification of the active ingredients and hence higher potency.

It is worth noting that, while the serum glucose level gradually increased in the saline-treated diabetic group, it changed in the opposite direction in the extract-treated control and diabetic animals. In other words, the longer the animals received the extract, the lower serum glucose levels they presented.

During the four-week experimental period, body weights were reduced in the saline- and extract-treated diabetic rats; however, the extent of reduction was lower in extract-treated diabetic rats so that their weights were significantly higher than those of the saline-treated diabetic animals at the end of the fourth week. The failure of STZ-induced diabetic rats to gain weight has already been reported (16–18). Administration of the extract restored these levels toward a normal range. The ability of the extract to partially restore body weight seems to be because of its ability to reduce hyperglycemia (16, 19). This may also be due to the protective effect of the extract in controlling muscle wasting, i.e., reversal of gluconeogenesis (20). As oxidative stress has been shown to play a key role in the pathogenesis of diabetes, antioxidants may play a role in the alleviation of diabetes (21). It is possible that treatment with the extract might lead to a better utilization of nutrients in the diet and thus a gain in weight.

The results of the current study provide evidence for the marked decrease in cardiac function parameters as a result of diabetes before and after ischemia. These findings are consistent with the results presented by Ravingerova et al. (22) and Nemeth et al. (23), who showed that 4- and 8-week diabetes significantly decreases baseline heart rate and coronary flow. Although the actual mechanisms are not completely understood, some possible factors that might be involved in the progression of cardiovascular malfunction in diabetic rats include decreased hepatic glucokinase and hexokinase activity, increased oxidative stress due to excessive production of oxygen free radicals, reduced antioxidant defense systems (8, 24, 25), decreased endothelial nitric oxide (NO) release or production, decreased reactivity of vascular smooth muscle to NO in diabetic animals, and increased production of superoxide anions (26).

In addition to the partial improvement of blood glucose concentrations, the results of our study are indicative of improvement of cardiac functional parameters of the diabetic rats in the baseline and post-ischemic periods following extract administration. Therefore, some part of the improvement can be explained by the efficacy of the extract in improving the glucose metabolism, but some of it may also result from the antioxidant and other therapeutic properties of the ingredients.

It has been reported that the hypoglycemic effect of shallot extract may be induced by its flavonoid compounds, especially quercetin, which is a highly potent antioxidant flavonoid (4, 5). Studies have shown that quercetin is capable of increasing both glucose tolerance and hepatic glucokinase activity in STZ-induced diabetic rats (27–29). Diets with a high content of quercetin have been associated with decreased risk of cardiovascular diseases (30–32). Consistent with this idea, in vivo treatment with shallot extract reduces the end-products of lipid peroxidation (33, 34). Furthermore, the lipid-oxidation-lowering effect of shallot extract may be induced via its direct superoxide scavenging properties or indirectly increased NO synthesis (35). Moreover, Persian shallot is full of organosulfur compounds, especially allicin. These compounds may be responsible for some beneficial properties of this plant (36, 37). In agreement with our findings, several studies have shown the hypoglycemic effect of garlic, which is attributed mainly to allicin-type compounds (38, 39). Therefore, the hypoglycemic and the beneficial effects of ethyl acetate fraction of shallot on the cardiac function of diabetic rats can be explained by its antioxidant potency, which needs to be elucidated in the future studies.

The vessels of diabetic patients show marked abnormalities in endothelial function, characterized by reduced NO bioactivity and increased superoxide production (40). Impaired NOS activity and reduced NO bioavailability are common initiators of cardiovascular dysfunction (41). Some medicinal plants may improve cardiovascular function through nitric oxide modulation (42). In our study, the protective effect of the extract might have been caused by increased NO bioactivity, resulting in improvement of cardiac function parameters.

**Study limitations**

Lack of quantitative analysis of nitric oxide pathway and other biochemical parameters can be considered as a limitation of this study.

**Conclusion**

Besides lowering blood glucose level, oral administration of ethyl acetate fraction of *Allium hirtifolium* significantly improved the baseline and post-ischemic cardiac function parameters in the STZ-induced diabetic rats. Therefore by extension it can be used in the management of diabetic and cardiac dysfunction. Further studies are recommended to investigate the underlying mechanisms.
Acknowledgements: This research was supported by the Medical Biology Research Center, Kermanshah University of Medical Sciences, Kermanshah, Iran.

Conflict of interest: None declared.

Peer-review: Externally peer-reviewed.

Authorship contributions: Concept – M.H.; Design – M.H., S.Kh.; Supervision – D.Sh., A.M.; Materials – M.H., D.Sh., A.M.; Data collection &/or processing – S.Kh., M.H., A.G., D.Sh.; Literature search – S.Kh., A.G.; Writing – S.Kh., M.H., A.G.; Critical review – D.Sh., A.M.

References

1. Kannel WB, McGee DL. Diabetes and cardiovascular disease. The Framingham study. JAMA 1979; 241: 2035-8.
2. Dubrey SW, Reaveley DR, Seed M, Lane DA, Ireland H, O’Donnell M, et al. Risk factors for cardiovascular disease in IDDM. A study of identical twins. Diabetes 1994; 43: 831-5.
3. Levy C, Seefeld LD, Lindor KD. Use of herbal supplements for chronic liver disease. Clin Gastroenterol Hepatol 2004; 2: 947-56.
4. Asadi-Samavati S, Azimi N, Fasahi A, Alineja-Ahandani E, Rafieian-Kopaei M. Medicinal plants with hepatoprotective activity in Iranian folk medicine Asian Pac J Trop Biomed 2015; 5: 146-57.
5. Kazemi S, Asgary S, Mostaghanj J, Rafieian M, Adelina A, Shamsi F. Liver-protective effects of hydroalcoholic extract of Allium hirtifolium Boiss. In rats with alloxan-induced diabetes mellitus. ARYA Atheroscler 2010; 6: 11-15.
6. Mink PJ, Scrafford CG, Barraj LM, Harnack L, Hong CP, Nettleton JA, et al. Flavonoid intake and cardiovascular disease mortality: a prospective study in postmenopausal women. Am J Clin Nutr 2007; 85: 895-909.
7. Asgari S R-kM, Pourheysari B, Ansari-Samani R, Deris F, Shahin-Fard N, Hojjati MR, et al. Allium hirtifolium Boiss: Radical scavenging property and the lowering effects on blood fibrinogen and factor VII. Life Sci J 2012; 9: 1793-8.
8. Mahmoodi M, Zarei S, Rezaeian M, Arababadi MK, Ghasemi H, Khoramdelazad H, et al. Persian Shallot (Allium hirtifolium Boiss) Extract Elevates Glucokinase (GCK) Activity and Gene Expression in Diabetic Rats. Am J Plant Sci 2013; 4: 1393-9.
9. Jalal R, Bagheri SM, Moghim H, Rasuli MB. Hypoglycemic effect of aqueous shallot and garlic extracts in rats with fructose-induced insulin resistance. J Clin Biochem Nutr 2007; 41: 118-123.
10. Seyfi P, Mostafaie A, Mansouri K, Arshadi D, Mohammadi-Motlagh HR, Kiani A. In vitro and in vivo anti-angiogenesis effect of shallot (Allium ascalonicum): a heat-stable and flavonoid-rich fraction of shallot extract potently inhibits angiogenesis. Toxicol In Vitro 2010; 24: 1655-61.
11. Famil Samavati S, Mohammadi-Motlagh HR, Mostafaie A. A highly pure sub-fraction of shallot extract with potent in vitro anti-angiogenic activity. Int J Mol Cell Med 2014; 3: 237-45.
12. Pandhare RB, Sangameswaran B, Mohite PB, Khanage SG. Anti-diabetic activity of aqueous leaves extract of Sesbania sesban (L) Merr. in streptozotocin induced diabetic rats. Avicenna J Med Biotechnol 2011; 3: 37-43.
13. Singh R AM, Kazmi I, Anwar F Talc used in anticancer drugs is promoter for diabetes in hepatocellular carcinoma induced rats. Eur J Cancer 2014; 50: 247-8.
14. Kumar V AD, Gupta PS, Anwar F, Mujeeb M. Anti-diabetic, antioxidant and anti-hyperlipidemic activities of Melastoma malabathricum Linn. Leaves in streptozotocin induced diabetic rats. BMC Complement Altern Med 2013; 13: 222-41.
15. Reshadat S, Nikray R, Alord S, Shacklebadi D, Godini A, Hesari M. The effects of acetazolamide on ischemia reperfused isolated hearts of 2- and 8-week-old rabbits. Saudi Med J 2012; 33: 250-5.
16. Tahmasebpoor N DG, Hosseinpourfeizi MA, Esmaeili HA. Variation in body weight and some hematological parameters in streptozotocin-induced diabetic rats, treated with Teucrium orientale. Pharmacology online 2013; 3: 32-6.
17. Marrif HI, Ali BH, Hassan KM. Some pharmacological studies on Artemisia herba-alba (Asso.) in rabbits and mice. J Ethnopharmacol 1995; 49: 51-5.
18. Mansi K, Lahjam J. Effects of Artemisia sieberi Besser (A. herba-alba) on heart rate and some hematological values in normal and alloxan-induced diabetic. J Basic Appl Sci 2008; 4: 57-62.
19. Dehghan G TN, Tahmasebpoor N, Hosseinpourfeizi MA, Sheikhzadeh F, Banan Khojasteh SM. Hypoglycemic, antioxidant and hepatoprotective effects of Teucrium orientale in streptozotocin diabetic rats. Pharmacol online 2013; 1: 182-9.
20. Burke JP WK, Narayan KM, Leibson C, Haffner SM, Stern MP. A population perspective on diabetes prevention: whom should we target for preventing weight gain? Diabetes Care 2003; 26: 1999-2004.
21. Eslamizad TA, Mohammadirad AS, Abdullahi M. Study on the oxidative stress status among cement plant workers. Hum Exp Toxicol 2008; 27: 463-9.
22. Ravingerova T, Styk J, Panca D, Tribulova N, Sebokova J, Volkovová K, et al. Diabetic cardiomyopathy in rats: alleviation of myocardial dysfunction caused by Ca2+ overload. Diabetes Res Clin Pract 1996; 31: S105-12.
23. Nemeth J, Szilvassy Z, Orozzi G, Porszasz R, Jakab B, Szolcsányi J. Impaired capsaicin-induced decrease in heart rate and coronary flow in isolated heart of diabetic rats. Acta Physiol Hung 2001; 88: 207-18.
24. Oberley LW. Free radicals and diabetes. Free Radic Biol Med 1998; 5: 113-24.
25. Baynes JW. Role of oxidative stress in development of complications in diabetes. Diabetes 1991; 40: 405-12.
26. Giugliano D, Ceriello A, Paolisso G. Oxidative stress and diabetic vascular complications. Diabetes Care 1996; 19: 257-67.
27. Su HC, Hung LM, Chen JK. Resveratrol, a red wine antioxidant, possesses an insulin-like effect in streptozotocin-induced diabetic rats. Am J Physiol Endocrinol Metab 2006; 290: E1339-46.
28. Chi TC, Chen WP, Chi TL, Kuo TF, Lee SS, Cheng JT, et al. Phosphatidylinositol-3-kinase is involved in the antihyperglycemic effect induced by resveratrol in streptozotocin-induced diabetic rats. Life Sci 2007; 80: 1713-20.
29. Vessal M, Hemmati M, Vasei M. Anti-diabetic effects of quercetin in streptozotocin-induced diabetic rats. Comp Biochem Physiol C Toxicol Pharmacol 2003; 135C: 357-64.
30. Hertog MG, Feskens EJ, Hollman PC, Katan MB, Kromhout D. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen Elderly Study. Lancet 1993; 342: 1007-11.
31. Geleijnse JM, Launer LJ, Van der Kuip DA, Hofman A, Witteman JC. Inverse association of tea and flavonoid intakes with incident myocardial infarction: the Rotterdam Study. Am J Clin Nutr 2002; 75: 880-6.
32. Knekt P, Kumpulainen J, Jarvinen R, Rissanen H, Heliovaara M, Reunanen A, et al. Flavonoid intake and risk of chronic diseases. Am J Clin Nutr 2002; 76: 560-8.

33. Kesavulu MM, Rao BK, Giri R, Vijaya J, Subramanyam G, Apparao C. Lipid peroxidation and antioxidant enzyme status in Type 2 diabetics with coronary heart disease. Diabetes Res Clin Pract 2001; 53: 33-9.

34. Leelarungrayub N, Chanarat N, Rattanapanone V. Potential activity of Thai shallot (Allium Ascalonicum L.) extract on the prevention of hemolysis and glutathione depletion in human erythrocyte from oxidative stress. CMU J 2004; 3(3): 225-34.

35. Chang KC, Chung SY, Chong WS, Suh JS, Kim SH, Noh HK, et al. Possible superoxide radical-induced alteration of vascular reactivity in aortas from streptozotocin-treated rats. J Pharmacol Exp Ther 1993; 266: 992-1000.

36. Amin M, Kapadnis BP. Heat stable antimicrobial activity of Allium ascalonicum against bacteria and fungi. Indian J Exp Biol 2005; 43: 751-54.

37. Segoviano-Murillo S, Sanchez-Gonzalez DJ, Martinez-Martinez CM, Cruz C, Maldonado PD, Pedraza-Chaverri J. S-allylcysteine ameliorates ischemia and reperfusion induced renal damage. Phytother Res 2008; 22: 836-40.

38. Chang ML, Johnson MA. Effect of garlic on carbohydrate metabolism and lipid synthesis in rats. J Nutr 1980; 110: 931-36.

39. Mathew PT, Augusti KT. Studies on the effect of allicin (diallyl disulphide-oxide) on alloxan diabetes. I. Hypoglycaemic action and enhancement of serum insulin effect and glycogen synthesis. Indian J Biochem Biophys 1973; 10: 209-12.

40. Guzik TJ MS, Gastaldi D, Sadowski J, Ratnatunga C, Pillai R, Channon KM. Mechanisms of increased vascular superoxide production in human diabetes mellitus role of NAD(P)H oxidase and endothelial nitric oxide synthase. Circulation 2002; 105: 1656-62.

41. Vanhoutte PM, Gao Y. Beta blockers, nitric oxide, and cardiovascular disease. Curr Opin Pharmacol 2013; 13: 265-73.

42. Karagöz A KS, Vural A, Usta M, Tezcan B, Semerci T, Teker E. Cardioprotective effects of Viscum album L. ssp. album (Loranthaceae) on isoproterenol-induced heart failure via regulation of the nitric oxide pathway in rats. Anatol J Cardiol 2016; 16: 923-30.

From Prof. Dr. Arif Akşit’s collection’s