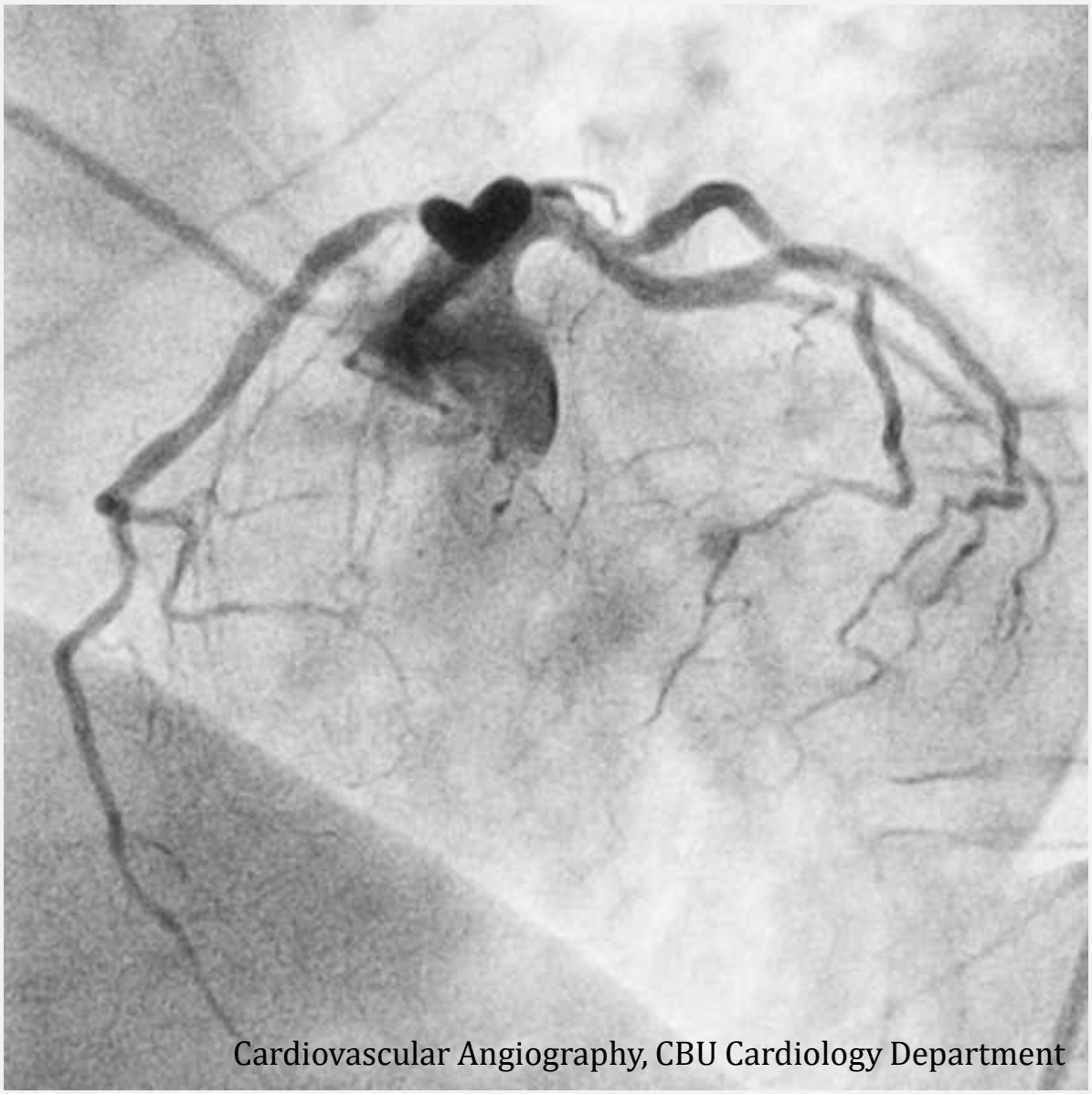
MSD
MEDICAL SCIENCE & DISCOVERY
ISSN: 2148-6832
Lycia Press LONDON UK



Cardiovascular Angiography, CBU Cardiology Department

High Lights

- The effect of statin over lens density
- Usage of some enzymes for diagnosis and detection of breast cancer by ROC curve
- Prevalence of early repolarization pattern in young healthy men
- Paraplegia and synthetic cannabinoid bonsai abuse

International Journal of
Medical Science and Discovery
Open Access Scientific Journal
June 2016, Vol.3, No.6
www.medscidiscovery.com

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ISSN: 2148-6832 (Print) E-ISSN: 2148-6832 (Online)
Category: Multi Disciplinary Health Science Journal
Abbreviated key title: Med. Sci. Discov.
Frequency: Monthly
Review System: Double Blind Peer Review
Circulation: Globally, Online, Printed
Article Processing Charge (APC): US$ 100
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Editor-in-Chief: Assoc. Prof. Dr. Zafer AKAN
Established: 30.04.2014
Address: CBU, Faculty of Medicine Dept. of Biophysics, Manisa, TURKEY
Web address: www.medscidiscovery.com; http://dergipark.ulakbim.gov.tr/msd
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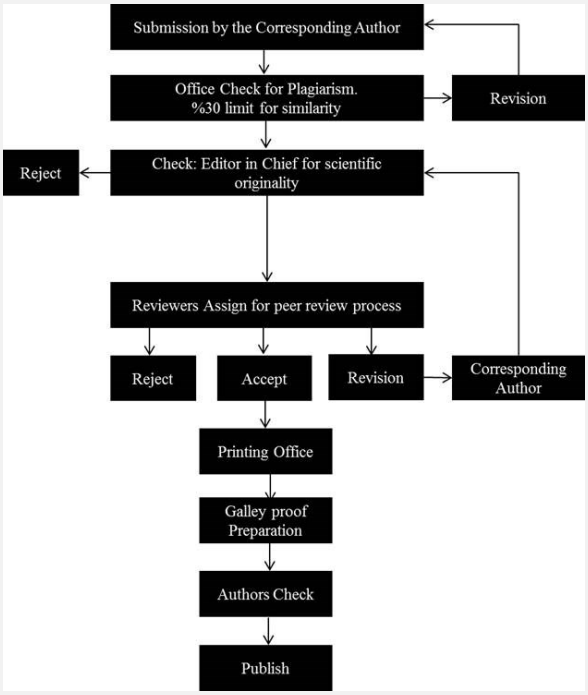

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Medical Science and Discovery, June 2016, Vol.3, No.6

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Case Reports

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ISSN: 2148-6832

MSD Medical Science and Discovery
2016; 3(6):245-9

Review Article Doi: 10.17546/msd.34944

The effect of statin use on lens density as assessed by pentacam hr® lens densitometry in adults

Akin Cakir[1*], Alptug Tokatli[2], Taner Kar[3], Yildiray Yildirim[3], Eyup Duzgun[4], Emrah Erdal[5]

Abstract

Objective: To investigate the association between statin use and cataract development by using Pentacam HR Scheimpflug camera system.

Material and Methods: Total 208 age and sex matched participants were included in this prospective, case-control study. 104 long-term statin users as study group and 104 non-statin users as control group constituted to the study. Lens densitometry measurements were performed with the Pentacam HR® (Oculus, Wetzlar, Germany) from three areas (anterior, nuclear, and posterior). These measurements were compared between the groups.

Results: There were no significant difference at the basic clinical variables (p=0.121, p=0.778). The mean duration of statin use of the study group was 5,4±4,3 years. The mean value of lens densitometry measurement at nuclear area tended to be lower in statin users than controls, but difference was not statistically significant (9.20±1.00 vs 9.35±1.20; p:0.346). However, the mean lens densitometry values at the anterior and posterior areas of statin users were found to be significantly higher than controls (10.44±1.32 vs 9.16±1.09;p<0.001; 7.86±0.49 vs 7.63±0.38; p<0.001,respectively).

Conclusion: The current study finds that long-term statin use seems to be associated with an increased risk of cortical cataracts (anterior-posterior) but not incidence of nuclear cataracts

Key Words: Statin, Cataract, Pentacam HR®.

Introduction

Cataract is the leading cause of low vision and blindness in the world (1). Because of the growing elderly population, the incidence of cataract is likely to increase. However, the exact mechanisms and pathogenesis of cataract formation are not completely understood. Therefore, investigating the risk factors for cataract development is crucial with its both medical and economical aspects (2).

Statins are widely prescribed drugs for their lipid-lowering effect. They also may have antioxidant effects and anti-inflammatory actions on the lens (3). While oxidative damage to the lens epithelium may induce cataract formation, statins may be protective against cataractogenesis. However, recent studies showed controversial results, with some suggesting an increased risk of cataracts with statin use (2,4,5) while others appear to show a beneficial effect of statins on cataract risk (5,6,7).

Another large study demonstrated that recent longer-term statin use was protective against cataract surgery in younger patients (50-64 years of age), while shorter-term use was associated with an increased risk of surgery (8). In the current study, we aimed to evaluate the effect of statin use on lens density by using Pentacam HR Scheimpflug camera system which provides objective and quantitative lens densitometry (LD) analysis..

Material and Methods

The 104 long-term statin users (study group) and 104 non-statin users (control group) were enrolled in this prospective, case-control study. The participants were matched for age and sex between groups. Informed consent was obtained from all the volunteers. The study was carried out in accordance with the tenets of the Declaration of Helsinki, and was approved by the institutional ethical committee.

Received 19-04-2016 **Accepted** 09-05-2016 **Available Online** 15-06-2016
1 GATA Haydarpasa Training Hospital, Naval Academy Clinic, Dept. of Ophthalmology, Tuzla, 34940, Istanbul, Turkey.
2 Golcuk Military Hospital, Dept. of Cardiology, Golcuk, 41650, Kocaeli, Turkey.
3 GATA Haydarpasa Training Hospital, Dept. of Ophthalmology, Uskudar, 34660, Istanbul, Turkey.
4 Gumussuyu Military Hospital, Dept. of Opthalmology, Taksim, 34420, Istanbul, Turkey.
5 Diyarbakir Military Hospital, Dept. of Cardiology, Diyarbakir, Turkey.
* Corresponding Author: Akin Cakir E-mail: dracakir@gmail.com Phone: +90 (216) 542 2020

The exclusion criteria were; the presence of any pathology of the anterior segment and fundus, any refractive errors within ± 3 diopters spherical equivalent, the presence of systemic diseases such as diabetes mellitus, hypertension and connective-tissue diseases, the history of ocular trauma and ocular surgery, the history of chronic ocular diseases such as glaucoma, keratoconus and uveitis, the history of smoking or current smokers, the history of long-term (more than 1 month) use of systemic or ocular corticosteroids.

The following examinations were applied to all the participants in the study; visual acuity, anterior segment biomicroscopy, intraocular pressure, dilated fundus examination and Scheimpflug anterior segment analysis.

LD measurements were performed with Pentacam HR after maximal dilation with 2,5% phenylephrine and 1% tropicamide ophthalmic solution. Only the right eyes of subjects were included in study. The right eye of each subject was scanned twice by one experienced observer.

To minimize operator-dependent variables, Pentacam's automatic release mode was used. In this mode, the instrument automatically determines when correct focus and alignment with the corneal apex have been achieved and then performs a scan. Automatic release and 50-picture 3-dimensional (3D) scan modes were used for measurements. Images of 90-270º were assessed.

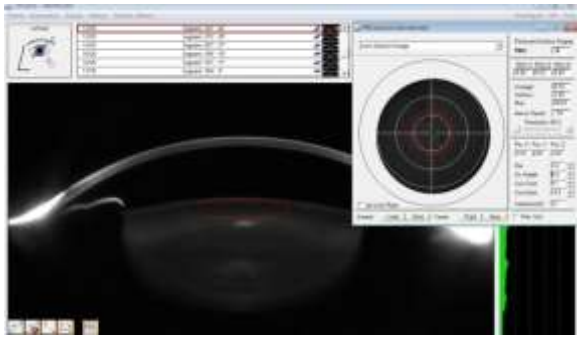

Figure 1: Anterior area (anterior subcapsular and cortical) measurement is shown.

Figure 2: Nuclear area measurement is shown in the figure.

While it is difficult to identify the exact boundaries of anterior subcapsular and cortical areas from the images, we measured the lens density from three areas. These areas were anterior (anterior subcapsular and anterior cortical), nuclear, and posterior (posterior subcapsular and posterior cortical).

To avoid measurement artefacts and perform a standard scan, we used 2,4 mm (horizontal) x 0,8 mm (vertical), 3,6 mm (horizontal) x 2.1 mm (vertical) and 2,4 mm (horizontal) x 0,6 mm (vertical) 3D body for anterior (Figure 1), nuclear (Figure 2) and posterior areas (Figure 3), respectively. After this process, Pentacam HR software calculated average values of 3D lens density automatically as seen in the figures in the top right corner.

All statistical analysis was carried out using SPSS 17.0 (SPSS, Chicago, Illinois, USA). Continuous variables were given as mean ± SD and categorical variables were defined as percentages. Normality of continuous variables' distribution was tested using Kolmogorov-Smirnov test. Normally distributed continuous variables were compared using independent samples t test. Other continuous variables were compared using Mann-Whitney U test. All analysis were two sided and considered significant at a p value of < 0.05.

A multiple linear regression model was used to identify independent predictors of LD. Also the effects of gender and age on LD were adjusted using two-way ANOVA and ANCOVA tests, respectively.

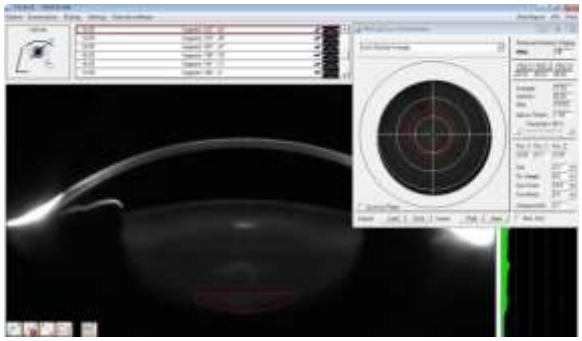

Figure 3: Posterior area (posterior subcapsular and posterior cortical) measurement is shown.

Results

Our study sample involved 104 long-term statin users and 104 healthy non-statin users. The study and control groups were similar in terms of gender and age. Study populations' baseline characteristics and demographics are given in table 1. The mean age of the study population was 58,4±8.4 years. Mean duration of statin use was 5,4±4,3 years. Among the statin users, a total of 56 patients (53.8%) had been recently using atorvastatin and 48 patients (46.1%) had stated rosuvastatin use.

Table 2 shows the results of mean LD values in the three areas in the study and control groups. The mean value of LD measurement at nuclear area tended to be lower in statin user group than controls, but difference was not statistically significant (9.20±1.00 vs 9.35±1.20; p:0.346). However, the mean LD value at the anterior area of study group was found to be statistically significantly higher (10.44±1.32 vs 9.16±1.09; p<0.001).

The mean LD value at the posterior area was also significantly higher in statin users (7.86±0.49 vs 7.63±0.38; p<0.001). In the subgroup analysis of LD measurements according to statins' type, no statistically significant difference existed between atorvastatin and rosuvastatin (Table 3). In order to eliminate the effect of gender and age on LD measurements, we have performed two-way ANOVA and ANCOVA tests, respectively. We found that while age has a statistically significant effect on the LD values of three areas (p<0.001), gender has not a statistically significant effect on the posterior area LD (p:0.089) but has an effect on the anterior and nuclear areas' LD measurements (p<0.001). Therefore, we performed a multiple linear regression model to identify regression coefficients of LD measurements. We observed that statin use was found to be more associated with LD values than gender and age (Table 4).

Table 1: Demographic characteristics of study and control groups.

	Study Group (N=104)	Control Group (N=104)	P value
Age (Mean [SD])	58,4 (8.4)	56,8 (6.0)	0.121
Sex (Male [%])	63 (60.6)	61 (58,7)	0.778*
Statin use (year)	5,4±4,3	-	
Statin agent type			
Atorvastatin (N[%])	56 (53.8)	-	
Rosuvastatin (N[%])	48 (46.1)	-	

*Mann-Whitney U

Table 2: LD measurements in the three areas in both groups.

	Study Group (N=104)	Control Group (N=104)	P value**
Anterior area (mean[SD])	10.44±1.32	9.16±1.09	<0.001
Nuclear area (mean[SD])	9.20±1.00	9.35±1.20	0.346
Posterior area (mean[SD])	7.86±0.48	7.63±0.38	<0.001

**Student t test

Table 3: Subgroup analysis of LD measurements in the three areas according to statins' type.

	Atorvastatin (N=56)	Rosuvastatin (N=48)	P value***
Anterior area (mean[SD])	10.41±1.29	10.47±1.36	0.816
Nuclear area (mean[SD])	9.16±0.88	9.25±1.14	0.672
Posterior area (mean[SD])	7.92±0.50	7.78±0.46	0.150

***Student t test

Table 4: Regression coefficients of LD measurements in the three areas in a multiple linear regression model.

	Anterior area		Nuclear area		Posterior area	
	B (Std Error)	P value	B (Std Error)	P value	B (Std Error)	P value
Statin use	1.262 (0.158)	<0.001	0.175 (0.140)	0.211	0.223 (0.059)	<0.001
Gender	0.594 (0.162)	<0.001	0.374 (0.142)	0.009	0.099 (0.060)	0.102
Age	0.037 (0.009)	<0.001	0.049 (0.008)	<0.001	0.013 (0.003)	<0.001

Discussion

There are several studies about the relationship between statin use and cataract formation. However, the findings in these studies are inconsistent and controversial. While some of them did not report an association between statin use and cataract (9,10,11,12), some found a protective effect of statins on cataract (5,6,7). Recently, few studies demonstrated an increased risk of cataract associated with statins (2,4,5,13,14). The majority of these studies are observational and dependent on the medical records derived from the national databases. Since cataract is not an ''all-or-nothing disease: there is a continuous spectrum of severe cataract to minor opacities that would only be found on detailed examination'' (12), it is likely therefore that early cataracts would have been missed in these studies. This may have caused a '' misclassification bias''. Therefore, we believe that our study differs with its unique design from these studies.

The results of this study suggest that statin use is significantly associated with anterior and posterior lens opacities (anterior-posterior subcapsular or cortical) but not incidence of nuclear cataracts. This difference may be due to the reality that varied etiological factors are related to specific cataract subtypes (5). For example, nuclear cataract is associated with oxidative stress, especially in the pathogenesis of age related cataract, cortical cataract is associated with UV exposure (15), and posterior subcapsular cataract with steroid use (16). Thus, it is possible that protective factors may also differ (5).

As previously reported, a protective effect of statins on nuclear cataract is biologically plausible because oxidative stress (17,18) and inflammation (19) have been shown to be related to nuclear cataract, and statins have been reported to counter such effects (20,21).

On the other hand, in another study it was shown that statins were differentially distributed in the lens, particularly a higher concentration was achieved in the cortex than in the nucleus of the beagles' lenses (22). This finding may explain why higher cortical cataract incidence was found in our study. In addition; Cenedella (23) hypothesized that the inhibition of cholesterol biosynthesis by statin medications prevents proper epithelial cell development within the crystalline lens. The lens' epithelial cells require high cholesterol levels to maintain transparency of the lens. Increased rates of cataract among animals and humans with hereditary cholesterol deficiency have been observed in another study (24). Recently, lanosterol synthase deficiencies in two families with extensive congenital cataracts were reported (25). The researchers (25) mentioned that lanosterol treatment could reduce cataract severity and increase transparency in dissected rabbit cataractous lenses in vitro and cataract severity in vivo in dogs. Since lanosterol is synthesized by lanosterol synthase in a key cyclization reaction of a cholesterol synthesis pathway, statins may be leading to cataract formation by inhibiting this pathway.

Actually, the results of our study do not conflict entirely with the other studies' results, on the contrary, the results are complementary with them. For example, The Beaver Dam Eye Study reported a lower risk of nuclear cataract in statin users (5). A similar risk reduction was observed in the Blue Mountains Eye Study (6). However, a insignificant increased risk of cortical cataract (5) and posterior subcapsular cataract (6) were also reported in these studies respectively in a similar way to our results. Our study differs from these trials with our cataract assessment technique and in our opinion this is one of the strengths of our study. Having not been using data from medical records, we may have reduced the risk of '' misclassification bias''. Sure, encountering with some artefacts in the LD analysis process is possible but we have tried to avoid from these artefacts by using a standardization method in the measurement procedure. Nonetheless, some factors such as back scattering of light from the anterior lens area, shadows and light attrition may decrease the amount of light transmitted to the posterior pole of the lens and this may have caused nuclear and posterior area to appear less dense (26). Another limitation of the current study is that it is a not prospective cohort study. We have performed two-way ANOVA, ANCOVA and multiple regression analysis in order to evaluate the effect of gender and age on LD. Although statin use was still found to be more effective on LD than age and gender, our results may have been influenced by some other unpredictable factors. We have excluded the patients with any refractive errors within ± 3 diopters spherical equivalent and any systemic diseases such as diabetes mellitus, hypertension and connective-tissue diseases. Nevertheless, we know that anti-hyperlipidemic treatment is used for cardiovascular disorders and the patients usually have comorbidities. Surely, some factors like these may have affected our results

Conclusion

In conclusion, statins seem to be associated with an increased risk of cortical cataracts (anterior-posterior) but not incidence of nuclear cataracts. Nonetheless, the exact mechanism of the effect of statins in cataractogenesis is still unknown. Certainly, statins are very effective medications and the mainstay in the treatment of heart disease; therefore, side effects are expected. To identify the exact relationship, further double-blinded randomize controlled trials must be designed.

Conflict of Interest: The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical issues: All Authors declare that Originality of research/article etc... and ethical approval of research, and responsibilities of research against local ethics commission are under the Authors responsibilities. The study was completed due to defined rules by the Local Ethics Commission guidelines and audits.

Aknowledgement: None

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http://www.medscidiscovery.com
OPEN ACCESS JOURNAL

ISSN: 2148-6832

MSD Medical Science and Discovery
2016; 3(6): 250-4

Original Article Doi: 10.17546/msd.43631

The evaluation of diagnostic performance of some enzymes for diagnosis and detection of breast cancer by ROC curve

Halit Demir[1*], Sıddık Keskin[2], Canan Demir[3], Halis Gokyer[1]

Abstract

Objective: In this study, it was aimed to examine the performance of Arylesterase (ARE), Paraoxonase-1 (PON-1) and Adenosine deaminase (ADA) enzymes to identify cancer patients and their stages with ROC curve in serum samples taken from patients diagnosed as breast cancer in the Medical Oncology services at Yüzüncü Yıl University.

Material and Methods: The 25 healthy controls and 25 breast cancer patients are included in this study. 8 patients are in stage-1, 9 patients are in stage-2 and 8 patients are in stage-3. Adenosine deaminase (ADA), Paraoxonase-1 (PON-1) and Arylesterase (ARE) activities were detected in serums with Spectrophotometric method. Descriptive statistics for the traits studied were presented as median, mean, standard deviation, minimum and maximum value. In terms of these traits, Kruskal-Wallis test was performed to find out whether there are no differences between stages and control group. In addition, ROC analysis was performed to evaluate the performances of enzymes to classify patient and control group.

Results: The differences between stage-1, stage-2, stage-3 and control group for Adenosine deaminase (ADA) enzyme activities were found statistically significant ($p<0.01$). Besides, the difference between stage-2 and stage-3 was found insignificant for Paraoxonase-1 (PON-1) and Arylesterase (ARE) enzyme activity. Last, the difference between them and stage-1 and control group was found statistically significant.

Conclusion: It can be inferred that sensitivity and specificity values of the enzymes are favorable high and they have outstanding performance to identify tumor stage as well as patient and control group. Therefore, these enzymes should be suggested as a "diagnostic test" to classify patients with breast cancer and control group.

Key words: Arylesterase, adenosine deaminase, sensitivity, specificity.

Introduction

Cancer is a serious disease which is the result of cells' abnormal growth, multiplying out of control, spreading to other tissues and which results mostly in death. Cancer causes non-functionality of the organs and body. The exact reasons are unknown yet. However, there are two risk factors for this disease. While the first group risk factor depends on age, gender, lifestyle and family history; the second one consists of environmental factors. Cigarette, use of alcohol, air pollution, some viruses, exposure to extensive sunlight, overdose radiation and X- rays, and chemicals such as benzene, coloring materials and asbestos can be ranked among these environmental factors. It is stated that after lung cancer, breast cancer (%23) occurrence in women is the second most common kind of cancer in the world (1, 2).

One out of every eight women is under the risk of getting cancer according to a research conducted in the US in 2011 (3).

There are some tumor markers, such as carcino-embryonic antigen (CEA) alpha-fetoprotein (AFP) calcitonin (CT) and alkaline phosphatase (ALP) to evaluate parenchyma. In other words, tumors respond to treatment and to detect metastasizes early (4). In breast cancer, the decrease in superoxide dismutase (SOD) and antioxidant enzyme (catalase) activity was detected. In the same study, it is reported that there is a correlation between the increase in blood lipid peroxidation and SOD and decrease in catalase activity (5, 6). Paraoxonase-1 (PON-1), which is calcium-dependant ester hydrolase in glycoprotein structure, is an enzyme which has both arylesterase

Received 19-04-2016 **Accepted** 20-05-2016 **Available Online** 15-06-2016
1 Yuzuncu Yil University, Faculty of Science, Department of Chemistry/Biochemistry-Van, Turkey
2 Yuzuncu Yil University, Faculty of Medicine, Department of Biostatistics-Van, Turkey
3 Yuzuncu Yil University, Vocational School of Health Services-Van, Turkey
* Corrospending Author: Halit Demir E-mail: halitdemi@gmail.com Phone: +904322251806

(ARE) (E.C. 3.1.1.2) and paraoxonase (E.C.3.1.8.1) serum activity (7, 6). Adenosine deaminase (ADA) is a key enzyme in mammal purine salvage pathway and catalyses the inosine transformation of adenosine (6). In recent studies, it has been reported that oxidative stress emerging with regard to the changes in the level of prooxidant-antioxidant in patients with breast, colon and prostate cancer is related to malign diseases (6).

Diagnostic tests used in medicine to make decisions are generally laboratory tests or observations or measurements made via devices. Clinical performance of a diagnostic test is evaluated with general accuracy rate or according to whether or not it classifies ill individuals as "ill" and healthy individuals as " healthy". Most of the laboratory findings used for diagnosis of many diseases is biochemical traits which have are continuous variables. As a diagnostic test, in order to find out continuous variable performance, ROC (Receiver operating characteristic) curve is one of the commonly used methods to classify ill and healthy individuals by detecting cut-off points. ROC is the curve which is obtained by taking measured value of continuous variable as cut-off points and with marking Sensitivity level on Y axis, 1- Specificity level on X axis. The total area under the curve is "1". This indicates that classification power of the trait is 100%. However, when the area under the curve is 0.50, this means that there is no classification power for the trait. ROC curve summarizes accuracy of the test with a numerical value.

In this study, it was aimed to examine the performance of Arylesterase (ARE), Paraoxonase-1 (PON-1) and Adenosine deaminase (ADA) enzymes to identify cancer patients and their stages with ROC curve in serum samples taken from patients diagnosed as breast cancer in Yüzüncü Yıl University Medical Oncology services.

Material and Methods

The data of this study were taken from the study conducted by Gökyer (2015) (6). 25 healthy controls and 25 breast cancer patients are included in the study. 8 patients are in stage-1, 9 patients are in stage-2 and 8 patients are in stage-3. Before taking blood samples, the approval of Yüzüncü Yıl University, Medical Faculty, Education and Research and Application Hospital Clinic and Laboratory Research Local Ethics Committee was received. 3 ml venous blood samples were taken from each patient involved in the study. These samples were centrifuged in 5000 rpm/min for nearly ten minutes and serums were separated and Adenosine deaminase (ADA), Paraoxonase-1 (PON-1) and Arylesterase (ARE) activities were detected in serums with Spectrophotometric method.

Statistical Analysis: Descriptive statistics for the traits included in the study was presented as median, mean, standard deviation, minimum and maximum value. For these traits, Kruskal-Wallis test was performed to find out whether there are any differences between stages and control group. In addition, ROC curve analysis was also carried out to evaluate the performances of enzymes to identify the patient and control group. Statistical significance level was considered as 5% and SSPS (ver: 13) statistical software was used for all statistical computations.

Results

Descriptive statistics and comparison results are given in Table 1 for Arylesterase (ARE), Paraoxonase-1 (PON-1) and Adenosine deaminase (ADA) enzyme activities. As shown in Table 1, the differences between stage-1, stage-2, stage-3 and control group for Adenosine deaminase (ADA) enzyme activities were found statistically significant ($p<0.01$). While the difference between stage-2 and stage-3 was not significant for Paraoxonase-1 (PON-1) and Arylesterase (ARE) enzyme activity, the differences of these from stage -1 and control group was found (statistically) significant. Accordingly, mean value of Adenosine deaminase (ADA) enzyme activity was found 24.89 EU/L. Mean values were found 37.41 EU/L in stage-1, 45.07 EU/L in stage-2 and 58.34 EU/L in stage-3. When the mean values of Paraoxonase-1 (PON-1) enzyme activity were considered, these values were 4.46 EU/L in stage-1, 0.51 EU/L in stage-2 and 0.25 EU/L in stage-3, while 13.11 EU/L in control group. Arylesterase (ARE) enzyme activity means were 236.38 kEU/L in control group, 41.68 kEU/L in stage-1, 1.32 kEU/L in stage-2 and 0.18 kEU/L in stage-3.

Table 1. Descriptive statistics and comparison results

The results of ROC analysis conducted to evaluate the performance of Adenosine deaminase (ADA) enzyme for the classification of healthy and diseased individuals and the stages of diseased individuals from each other are given in Table 2. One of the criterions to evaluate the performance of the tests for giving the true positive decision is the area under the ROC curve. This area ranges from the value of maximum 1 to minimum 0.5. The area under the ROC curve was found 0.987 for patient and control-group in Table 2. While this value was found 0.960 for stage -1 and stage -2, the areas under the curve were 1 for the other groups. For determining the best cut-off point values by means of ROC curve, these values are observed as 34.039 for patient and control-group, 42.380 for stage-1 and control-group, 47.805 for stage-2 and control-group, 51.411 for stage -3 and control-group, 34.039 for stage-1 and stage -2 , 40.126 for stage-1 and stage-3 , 45.55 for stage-2 and stage-3.

Table 1. Descriptive statistics and comparison results

Groups		Stage-1	Stage-2	Stage-3	Control
Adenosine deaminase (ADA)	Median	36.07 c	44.64 b	59.58 a	24.35 d
	Mean ± SD	37.41 ± 3.42	45.07 ± 1.87	58.34 ± 3.59	24.89 ± 7.88
	Min.	34.27	42.83	53.68	14.43
	Max.	41.93	49.14	64.68	37.42
Paraoxonase-1 (PON-1)	Median	5.12 b	0.48 c	0.24 c	13.42 a
	Mean ± SD	4.46 ± 3.22	0.51 ± 0.16	0.25 ± 0.16	13.11 ± 2.07
	Min.	0.78	0.30	0.06	10.11
	Max.	9.35	0.78	0.54	17.45
Arylesterase (ARE)	Median	21.85 b	1.45 c	0.13 c	228.56 a
	Mean ± SD	41.68 ±43.40	1.32 ± 0.42	0.18 ± 0.07	236.38 ±34.72
	Min.	1.84	0.66	0.13	198.95
	Max.	112.52	1.84	0.26	335.43
p		0.01	0.01	0.01	0.01

Table 2. Results of ROC analysis for ADA

Groups	Cut-off value	The area under the curve	Std. Error	Sensitivity	Specificity	p
Patient- Control	34.039	0.987	0.011	1.000	0.920	0.001
Stage 1-Control	42.380	1.000	0.001	1.000	1.000	0.001
Stage 2- Control	47.805	1.000	0.001	1.000	1.000	0.001
Stage 3- Control	51.411	1.000	0.001	1.000	1.000	0.001
Stage 1- Stage 2	34.039	0.960	0.032	1.000	0.920	0.001
Stage 1- Stage 3	40.126	1.000	0.001	1.000	1.000	0.001
Stage 2- Stage 3	45.550	1.000	0.001	1.000	1.000	0.001

Table 3. Results of ROC analysis for PON-1

Groups	Cut-off value	The area under the curve	Std. Error	Sensitivity	Specificity	p
Patient - Control	9.730	1.000	0.001	1.000	1.000	0.001
Stage 1- Control	0.750	0.993	0.011	1.000	0.890	0.001
Stage 2- Control	0.660	1.000	0.001	1.000	1.000	0.001
Stage 3- Control	0.240	0.882	0.086	1.000	0.500	0.008
Stage 1- Stage 2	9.730	1.000	0.001	1.000	1.000	0.001
Stage 1- Stage 3	5.445	1.000	0.001	1.000	1.000	0.001
Stage 2- Stage 3	5.325	1.000	0.001	1.000	1.000	0.001

Table 4. Results of ROC Analysis for ARE

Groups	Cut-off value	The area under the curve	Std. Error	Sensitivity	Specificity	p
Patient- Control	155.735	1.000	0.001	1.000	1.000	0.001
Stage 1- Control	1.710	0.993	0.014	1.000	0.890	0.001
Stage 2- Control	1.052	1.000	0.001	1.000	1.000	0.001
Stage 3- Control	460	1.000	0.001	1.000	1.000	0.001
Stage 1- Stage 2	155.735	1.000	0.001	1.000	1.000	0.001
Stage 1- Stage 3	100.397	1.000	0.001	1.000	1.000	0.001
Stage 2- Stage 3	99.607	1.000	0.001	1.000	1.000	0.001

According to these cut-off values, it can be noted that the sensitivity of the test which is the ability to identify the individuals as patient equal "1" for all groups. The specificity value of the test, which is the ability to identify the individuals as healthy, is 0.920 for patient and control-groups and stage-1 and stage-2. However, this value is 1 for the other groups.

For Paraoxonase-1 (PON-1) enzyme activity, ROC analysis results concerning patient and control groups along with patients' stages are given in Table 3. While considering Table 3, it can be stated that the area under the curve is 0.993 for stage-1 and control-group, 0.882 for stage-3 and control-group and 1 for the other groups. Accordingly, the best cut-off point values were found 9.730 for patient and control group, 0.750 for stage-1 and control group, 0.660 for stage-2 and control group, 0.240 for stage-3 and control group, 9.730 for stage-1 and stage-2, 5.445 for stage-1 and stage-3 and 5.325 for stage-2 and stage-3. According to these cut-off values, while sensitivity was found 1 for all groups, specificity values were recorded as 1 for the other groups except stage-1 and control group (0.890) and stage-3 and control group (0.50).

For Arylesterase (ARE) enzyme activity, results of ROC analysis concerning patient and control groups along with patients' stages are given in Table 4. Findings in Table 4 indicate that the area under curve is 1 for the all groups excepting for stage-1 and control group. Accordingly, the best cut-off values were found 155.735 for patient and control group, 1.710 for stage-1 and control group, 1.052 for stage-2 and control-group, 0.460 for stage-3 and control group, 155.735 for stage-1 and stage-2, 100.397 for stage-1 and stage-3 and 99.607 for stage-2 and stage-3. Sensitivity values of the diagnostic test were found 1 for all groups and specificity values were found 1 for all groups except stage-1 and control group (0.890).

Discussion

Although cancer occurs as a result of malignant transformation of the cells, there is very little difference between normal and cancer cells in terms of genotypic expression. The mutations causing cancer are unlikely to be able to change either genetic or phenotypic expression except cell growth regulation. Breast cancer is a type of cancer that forms in breast tissue and differs significantly between geographical regions. The incidence of breast cancer is more frequent in developed countries compared to less developed countries in East Asia. When categorized in terms of age, the rate of getting breast cancer is 76.7 in one hundred thousand in North America while this rate is 23.5 in one hundred thousand in East Asia (8). According to scientific studies, the rate of getting breast cancer is higher in low-income countries and the women in those countries are under bigger risk.

In biological and biochemical systems; catalase (CAT), arylesterase (AR), paraoxonase-1 (PON-1), peroxidise (POD), glutathione reductase (GR) and superoxide dismutase (SOD) are enzymes which have antioxidant effects. Antioxidant defense system protects cell from the oxidative damage of free radical or other reactive molecules. For this reason, antioxidant enzymes such as CAT, PON-1, ARE, GR and SOD have great importance in defense system. Detrimental effects of free radicals are controlled by antioxidant defense systems in cells (9, 6). In a study on breast cancer, serum PON-1 activities in patients were found lower compared to control-group (10).

Again, in a literature review, ADA activity was found higher in breast cancer patients compared to healthy control group (11). In literature, the activity of ARE may be a useful for study clinical in breast cancer (12).

Conclusion

ROC curve is one of the commonly used methods to evaluate any diagnostic tests, determine the most appropriate cut-off value and determine the efficiency of the test depending upon this value. In addition to numerical values, graphical presentation contributes to interpretation of the results.

It can be concluded that sensitivity and specificity values of the enzymes are favorable high and they have outstanding performance to identify tumor stage as well as patient and control group. Therefore, these enzymes should be suggested as a "diagnostic test" to identify breast cancer patients and control group.

Conflict of Interest: The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical issues: All Authors declare that Originality of research/article etc... and ethical approval of research, and responsibilities of research against local ethics commission are under the Authors responsibilities. The study was completed due to defined rules by the Local Ethics Commission guidelines and audits.

Acknowledgments: None

Author Contributions: H D: Hypothesis and design, S K: Hypothesis and Statistical Analysis, C D: Sample collection and tests, H G: Sample collection and tests

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http://www.medscidiscovery.com
OPEN ACCESS JOURNAL

ISSN: 2148-6832

MSD Medical Science and Discovery
2016; 3(6): 255-8

Original Article Doi: 10.17546/msd.94548

Prevalence of early repolarization pattern in young healthy men

Tarik Yildirim[1]*, Eyup Avci[1], Seda Elcim Yildirim[1], Gokhan Aydin[1], Ahmet Dolapoglu[2], Adil Ozan Gokuc[3]

Abstract

Objective: Although the early re-polarization pattern known as a benign condition, it can result in idiopathic ventricular fibrillation and sudden cardiac death. In this study, we aimed to evaluate the prevalence of early repolarization pattern in young healthy men.

Methods and Patients: Four hundred fourteen young healthy men were enrolled in the study. PR intervals, QRS durations, QT intervals and corrected QT durations were measured from electrocardiographic records. Early re-polarization pattern was defined as if an elevation J-point was greater than 0,1 mV according to the iso-electrical point.

Results: The prevalence of early repolarization pattern was 23,2 % (n=96). Of these, we found early re-polarization pattern in inferior, lateral and inferolateral leads with 14,7%, 2,2% and 6,3% respectively. There was a significant relation between heart rate and elevation of J point.

Conclusion: The prevalence of early repolarization pattern in young healthy men from Turkey was similar with the previously reported rates from different white populations.

Key Words: Early repolarization; Healthy men; Cardiology

Introduction

Early repolarization pattern (ER) is characterized by if elevation of J point, that is junction between the end of QRS complex and the beginning of the ST segment is greater than 0,1 mV. And it is required at least two subsequent derivation with ERP for diagnosis (1,2). ER pattern on precordial derivation is benign condition but there is a strong relation between idiopathic ventricular fibrillation (VF) and ER pattern that seems in inferior and/or lateral leads (3-8). ER syndrome is defined as sudden cardiac death or occurrence of VF/ polymorphic ventricular tachycardia (VT) that cannot be explained in patient with ER pattern (9). Hypothermia, cocaine usage, Brugada syndrome and hypertrophic cardiomyopathy should be considered on differential diagnosis. Prevalence of ERP is 1-13 % in normal population and 15-70 % in patients with idiopathic VF (10,11). The rate is 44 % in young athletes and most of them have ascending ST segment pattern after J point (12). Existence of strong evidence between the sudden cardiac death and ER pattern and due to absent of literature knowledge about ER pattern of young men in Turkey, we aimed to find the rate of ER pattern among young healthy men in Turkey.

Material and Methods

After a protocol was approved by the ethical committee, we identified 414 young healthy men who applied for police high-school and had physical examination in cardiology department of Balikesir State Hospital. None of participants had diabetes mellitus and hypertension. The participant's ages were shown in table 1. Data were collected retrospectively including electrocardiographic (ECG) records. All subjects underwent 12-leads standard ECG in the supine position. A paper speed of 25 mm/s and a calibration of 10 mm/mV were used (Nihon-Kohden, ECG-9010D Japan). All records were evaluated by two special cardiologists for the presence of early repolarization pattern. Patients with complete right bundle branch block (RBBB), complete left bundle branch block (LBBB), J point elevation in V1-V3 leads, long QT interval and arrhythmia, Wolf-Parkinson-White syndrome were not included in the study.

Statistical Analyses: The statistical analyses were performed using the Statistical Package For Social Studies (SPSS version 20.0 USA). P value <0.05 was accepted statistically significant.

Received 14-03-2016 **Accepted** 18-05-2016 **Available Online** 15-06-2016
1 Balikesir State Hospital, Balikesir Department of Cardiology, Balikesir, Turkey
2 Balikesir State Hospital, Balikesir Department of Cardiovascular Surgery, Balikesir, Turkey
3 Balikesir State Hospital, Balikesir Department of Anesthesiology, Balikesir, Turkey
* Corrospending Author: Tarik Yildirim E-mail: kdrtarik@gmail.com Phone: +90266223510

http://dx.doi.org/10.17546/msd.94548

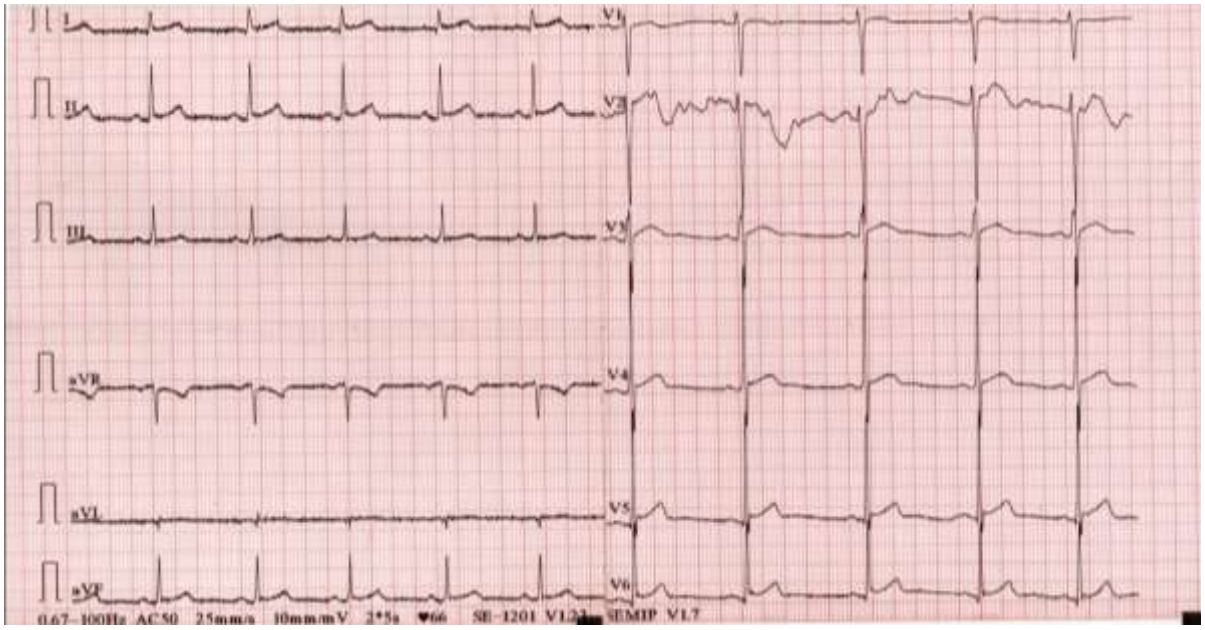

Figure 1. Inferolateral J point elevation

Results

Ninety-six patients had ER pattern and the prevalence was 23,2 %. Of these, individuals with J point elevation in the inferior, lateral and infero-lateral leads were 61 (14,7%), 9 (2,2%) and 26 (15,9%) respectively. There was no significant difference in QRS duration and PR interval between patients with ER pattern and without it. The ages were similar between patients with ER pattern in different lead pattern (inferior, lateral and infero-lateral leads). Eighty-four subjects had the J point elevation of 0,1 mV and 12 subjects had 0,2 mV. There was a significant link between heart rate and J point elevation. When the heart rate was reduced the elevation of J point was increased significantly. ECG example has been shown in figure 1.

Table 1. The age range of participants

Age	Number*	%
21	1	0,2
22	11	2,7
23	41	9,9
24	73	17,6
25	80	19,3
26	91	22,0
27	73	17,6
28	44	10,6
Total	414	100,0

Discussion

Haruto et all found the prevalence of ERP in their study 23,9 % and also they claimed that ER pattern was appeared in mostly second decade. Another study conducted by Haisaguerre et all showed the prevalence of 1-13 % and this may result from the intermittent occurrence of ER pattern (3,11). Our results were similar with Haruto`s study. On the other hand, patients enrolled our study were mostly in their second decades and this is confirming the Haruto`s finding.

In another study, Tikkanen et all found that the prevalence of ER pattern was 5,8 %. Of them, 3,5 % were seen in inferior leads, 2,4 % in lateral leads and 0,1 % in both leads. They also estimated the incidence of 0,33 when the J point elevation was greater than 0,2 mV. In that study there was a relation between J point elevation and prolonged QT interval and this was stronger predictor than left ventricular hypertrophy (13). Our ER pattern prevalence were 14,7 %, 2,2 % and 6,3 % in inferior, lateral and infero-lateral leads respectively.

Siner et all showed that cardiac sudden death occurred 2-4 times more in patients with ER pattern. Also ERP seemed more in inferior and lateral leads in patients with idiopathic VF. Sudden cardiac death rate was high 2,9 times more when the J point elevation was greater than 0,2 mV and the this rate increased 1,4 times whit the J point elevation of 0,1 mV. J point elevation along with horizontal or descending ST segment increased the rate of death from arrhythmias but up sloping ST segment had benign prognosis (3,10,12,13). Based on these findings, patients with ER pattern should be closely investigated for history of syncope and family history of sudden death.

ER pattern is affected by autonomic tonus and heart rate and it is related with long QRS duration, short QT duration and left ventricular hypertrophy. It is seen more in male and second decade. The prevalence goes lower after second decade and the reason might be the hormones which change by aging process (14-16).

Although the mechanism of J point and ER pattern are not understood yet completely, Osborn first tried to explain the effect of ions in cell membrane during hypothermia study. After this, another studies showed that the increase repolarization between the endocard and myocardium is responsible for the main pathology (17).

Conclusion

We report here the prevalence of ERP in young healthy men. Patient with ERP has a higher incidence of ventricular arrhythmia and sudden cardiac death than normal population. We suggest that all young patients with ERP need to be questioned for previous syncope history and family history of sudden cardiac death. More clinical and experimental studies should be performed for explain the relation in ERP and VF.

Conflict of Interest: The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical issues: All Authors declare that Originality of research/article etc... and ethical approval of research, and responsibilities of research against local ethics commission are under the Authors responsibilities. The study was completed due to defined rules by the Local Ethics Commission guidelines and audits.

Acknowledgement: None

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OPEN ACCESS JOURNAL

ISSN: 2148-6832

MSD Medical Science and Discovery
2016;3(6):259-65

Original Article Doi: 10.17546/msd.40471

Effect of a carbohydrate rich diet in vas deferens contractility in rats

Ekrem Akdeniz[1*], Suleyman Sirri Bilge[2], Mustafa Suat Bolat[1], Fatih Firat[3], Arzu Erdal Agri[2], Omer Alici[4], Onder Cinar[1], Fikret Erdemir[5]

Abstract

Objective: We aimed to investigate the effects of a Carbohydrate-rich diet on vas deferens contractility and histology.

Methods and Patients: Twenty mature male rats were subjected into two groups. The control group received regular food and water and the study group received regular food and a carbohydrate-rich liquid diet mixture of 37–40% fructose, 30–36% sucrose and 27–30% glucose instead of water and prepared stock diet was diluted with 50% water. Animals were weighted and sacrificed after six weeks and the vas deferens has been evaluated with in vitro studies. Five rats from both the control and study groups, were separated into subgroups for dapoxetine administration 4 hours before they were sacrificed

Results: At the beginning of this study, the mean body weights were 228.6±6.7 g (range: 205-237) and 231.4±9.6 g (range: 202-243) in control and study groups, respectively. The mean body weights were 247.1±4.8 g (236-252) in the control group and 318.8±9.3 g (314-326) in the study group ($p<0.001$). Weight gain was more distinct in the study group when compared to control group ($p<0.001$) (9% vs 27.4%). Contractile responses were recorded in each group, including the dapoxetine subgroups, to various noradrenaline concentrations; all groups had similar results ($p>0.05$). Responses to Adenosine Three Phosphate (ATP) were also not significant ($p>0.05$). The contractile responses at the same frequencies of electrical field stimulation (EFS) were similar in both groups ($p>0.05$). Histological examinations showed no abnormalities in either group. .

Conclusion: Carbohydrate-rich diet caused no increase in fibrotic activity, and pharmacological and histological properties remained stable, but caused significant weight gain over a short time period. Furthermore, dapoxetine had no effect on the contractility of the vas deferens

Key Words: Dapoxetine, Carbohydrate, Obesity, Vas deferens

Introduction

The ejaculation process requires coordinated inputs from both the central and peripheral nervous systems to produce the expulsion of semen from the urethra. Three distinct phases of semen transportation have been defined: emission, closure of the bladder neck and ejaculation. Potentialization of the cerebral cortex stimulates the prostatic smooth muscle, seminal vesicles and the vas deferens via sympathetic nerves in the thoracolumbar axis. Thus, the deposition of pre-ejaculatory fluid into the posterior urethra (emission phase) facilitates. Sympathetic innervation initiates the bladder neck closure and finally, the propelling of semen occurs. There are many reasons why this delicate mechanism may be easily affected.

Obesity is defined as abnormal or excessive fat accumulation that increases health risks. By 2030, almost half of the population will be clinically overweight or obese (1). Obesity threatens the public health on a worldwide scale and is associated with gastrointestinal, cardiovascular, neuromuscular, genitourinary and oncological diseases, which may cause high morbidity and mortality (2).

Obesity may occur for various reasons that include excessive intake of fatty foods, gallbladder disease, hypercholesterolemia, atherosclerosis, heart disease, hypertension, stroke, depression, sleep apnoea syndrome, urinary infections, lumbar postural disorders and neurological, metabolic and endocrinologic diseases.

Received 04-05-2016 **Accepted** 11-06-2016 **Available Online** 15-06-2016
1 Samsun Training and Research Hospital, Department of Urology, Samsun, Turkey
2 Ondokuz Mayıs University, Department of Pharmacology, Samsun, Turkey
3 Tokat State Hospital, Urology Clinic, Tokat, Turkey
4 Gazi Osman Pasa University, Department of Pathology, Tokat, Turkey
5 Gazi Osman Pasa University, Department of Urology, Tokat, Turkey
*Corrospending Author: Ekrem Akdeniz E-mail: ekremakdeniz@yahoo.com Phone: 090 362 311 1500

Ejaculatory function may be affected by a variety of reasons with a commonly reported pathology being premature ejaculation (PE). Even though PE is not a life-threatening disturbance, it frequently causes lower satisfaction in sexual relationship, difficulty in relaxing during intercourse and less frequent intercourse (3). The prevalence of PE is 4–39% (4). Drugs used for lower urinary tract symptoms (LUTS), such as alpha blockers, may cause ejaculatory dysfunction (5). Hormonal disorders (hypo-or hyperthyroidism, diabetes mellitus, and hypogonadism), genital infectious diseases and psychiatric disturbances may also play a role in ejaculatory disturbances (6). Anejaculation is a another condition that involves the complete absence of ejaculation and may result from several causes including androgen deficiency, sympathetic denervation, pharmacologic agents and bladder neck/prostatic surgery.

The role of obesity in sexual function has gained importance in recent years. Not only high fat diets but also high amounts of carbohydrate consumption may have a negative effect on sexuality. Although obesity is considered as cause of PE, there is no strong evidence based on prospective studies in the literature (7). Sexual dysfunction, particularly PE may have related with smooth muscle contraction of vas deferens. While ACh (Acetylcholine) is responsible from stabile smooth muscle contraction, ACh and ATP both initiate nerve-mediated over-active smooth muscle contractions (8). These neurotransmitters rapidly break downs by ACh esterase. When ACh esterase activity reduces, purinergic component may be predominated and this may cause abnormal contractions. We used ATP and NA stimulation tests in order to show the contractile strength of vas deferens in the groups (9). Similarly, total contractile strength was measured using EFS, which represent the release of all neurotransmitters into the neuromuscular synaptic space (10).

In this present study, we aimed to investigate the effects of a Carbohydrate-rich diet on vas deferens contractility and histology in rats..

Material and Methods

Twenty mature male Sprague-Dawley rats 9-12 weeks of age were obtained from the Gazi Osman Paşa University vivarium sources. All procedures and protocols were conducted in accordance with the Guidelines for the Care and Use of Laboratory Animals (NIH application 865-23, Bethesda, MD, USA). Experiments were approved by the Gazi Osman Paşa University, Animal Care and Use Committee.

Rats were housed in a temperature and humidity-controlled room (22 °C and 60±5%, respectively) with a 12-hour light/dark cycle and were randomized into 2 groups of 10 animals each. The control group rats received regular food and water ad libitum. The study group rats received regular food and a liquid mixture of 37–40% fructose, 30–36% sucrose and 27–30% glucose instead of water, and stock nutrition was diluted with 50% water for feeding. At the beginning of this study, the mean body weights were 228.6±6.7g (range: 205-237) and 231.4±9.6 g (range: 202-243) in control and study groups, respectively. The mean body weights were 247.1±4.8 g (236-252) in the control group and 318.8±9.3 g (314-326) in the study group at the end of this study ($p<0.001$). Weight gain was higher in the study group than control group (9% vs 27.4%). After six weeks, rats were sacrificed for in vitro studies. Dapoxetine were administered to five rats from the control and study groups, 4 hours before scarification.

In Vitro Experiments

The vas deferens was prepared in 10 mm strips and transferred into organ baths containing 10 ml Krebs Henseleit solution (composition in mM: NaCl: 118, KCl: 5.6, CaCl2:2.5, MgSO4:1.2, KH2PO4: 0.9, NaHCO3: 25, and glucose: 11). The solution was kept in the standard conditions (5% CO2, 37.2 °C and pH 7.4). Each preparation was treated through a ring electrode (3 mm internal diameter, 1 cm apart) (MLA0305/8, AD Instruments, UK) connected to a stimulator (Grass, USA). The lower end of the preparation was attached to a holder, and the other end was attached to an isometric force transducer (MLT0201, AD Instruments, UK) coupled to a Quad-Bridge amplifier (ML118, AD Instruments, UK) that was connected to a digital recorder PowerLab/4SP (AD Instruments, UK). Strips were allowed to equilibrate for 1 hour followed by the application of 1 g of tension. The Krebs solution was refreshed every 15 minutes. The functional viability of the preparation was assessed by the addition of acetylcholine (Ach) and noradrenaline (NA) for vas deferens.

Vas deferens contractions

NA (Sigma, USA) (10-8–10-5 M) and adenosine triphosphate (ATP) (Sigma, USA) (10 7–10- 4 M) were administered in a cumulative manner. The frequency-response curves were constructed as follows: square wave pulses (60 V, 0.5 ms) were delivered for 20 seconds at increasing frequencies (2–64 Hz).

Statistical Analysis

All data are expressed as the mean±SEM. Data analyses were performed using GraphPad Instat software (v 3.0) (GraphPad, USA).

Following the assurance of a normal distribution of data, a one-way analysis of variance (ANOVA) with Tukey-Kramer post hoc tests were used for multiple comparisons. Values of $p<0.05$ were regarded as statistically significant

Results

Body weights were recorded after 6 weeks and the mean body weight was 247.1±4.8 g in the control group and 318.8±9.3 g in the study group (27.4% increase). At the beginning of this study, the mean body weights were 228.6±6.7g (range: 205-237) and 231.4±9.6 g (range: 202-243) in control and study groups, respectively. After six weeks the mean body weights were 247.1±4.8 g (236-252) in the control group and 318.8±9.3 g (314-326) in the study group at the end of this study ($p<0.001$).

Pre-treatment with dapoxetine did not change the NA-induced vas deferens contractions in either group (Fig.1). The cumulative addition of ATP (10-7-10-4) elicited rapid, transient, concentration dependent contractions in both groups. However, no significant differences were seen between the control and study groups ($p>0.05$). The addition of dapoxetine did not change the ATP-induced vas deferens contractions in either group ($p>0.05$) (Fig.2).

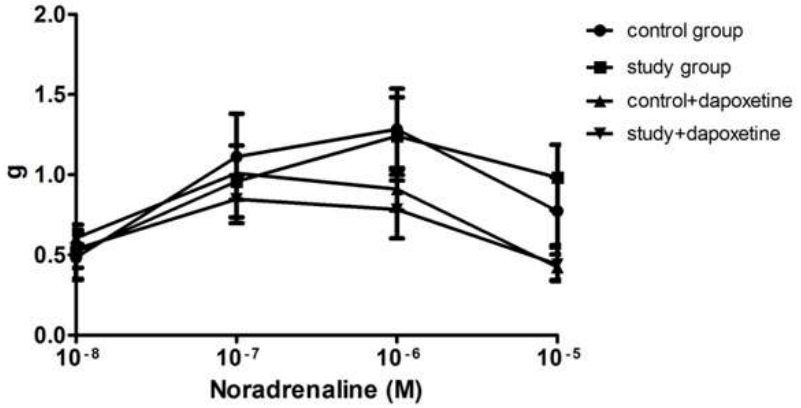


Figure 1: Contractions induced by Noradrenaline (10^{-8}-10^{-5}M) in vas deferens. Values have shown as the means±SE.

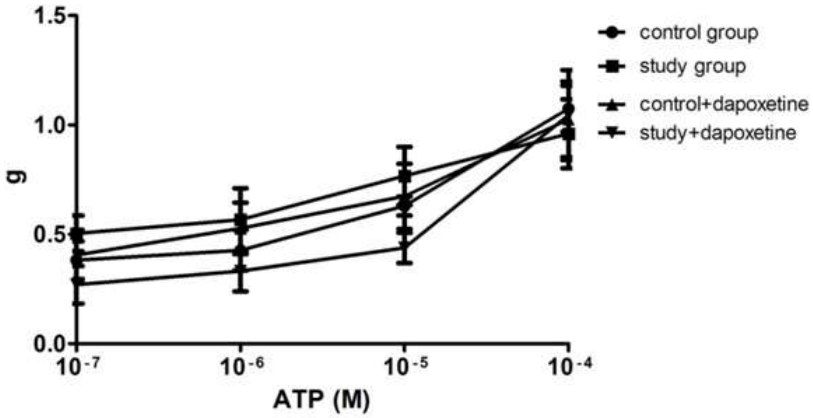


Figure 2: Contractions induced by ATP (10^{-7}-10^{-4} M) in vas deferens. Values have shown as the means±SE.

The cumulative addition of NA (10-8-10-5M) induced repetitive phasic, concentration-dependent contractions in the vas deferens of both the control and study groups. There were no significant differences between the control and study groups ($p>0.05$).

Electrical stimulation of the vas deferens strips of the normal fed rats were recorded in frequencies of 2-64 Hz. The contractile responses at the same frequencies of electrical field stimulation (EFS) were similar in both groups ($p>0.05$).

Pretreatment with dapoxetine did not change the EFS induced vas deferens contractions in either group (p>0.05) (Fig.3). Histological examinations showed no abnormalities in the control group with hematoxylin and eosin (H&E), and Masson's trichrome stain revealed no increase in subepithelial and intercellular fibrotic activity (Pic.1a and b, respectively). A carbohydrate-rich diet showed no increase in fibrotic activity in vas deferens (Pic.1c and d).

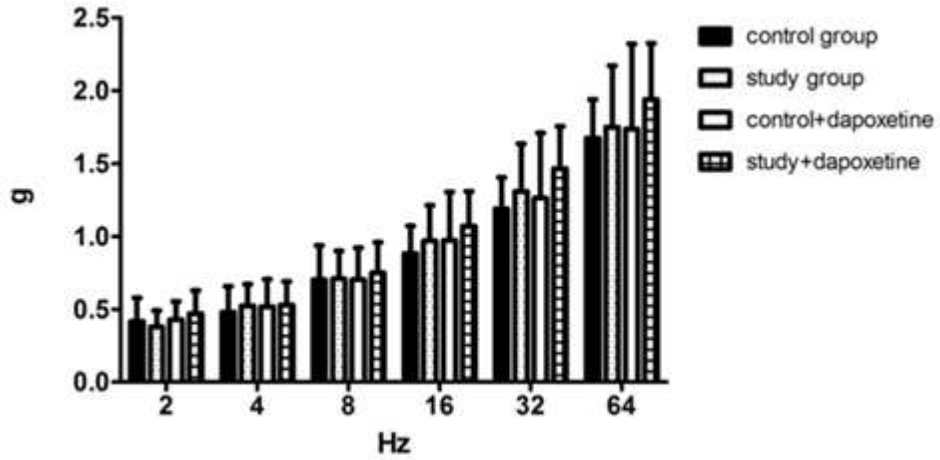
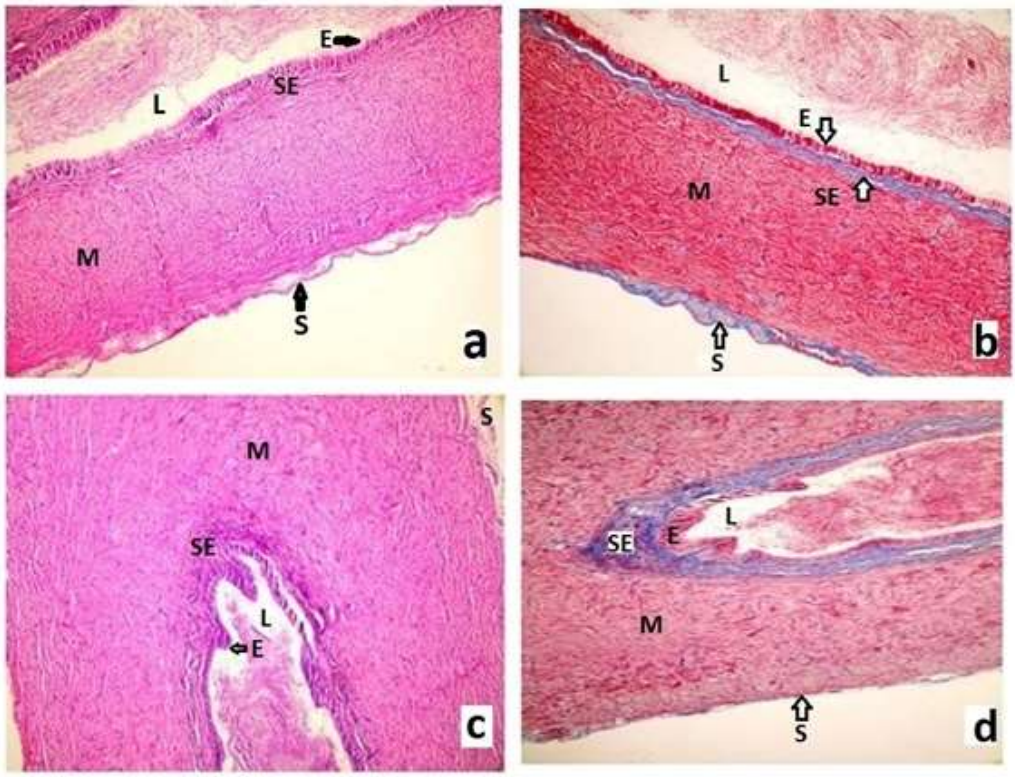


Figure 3: Contractions induced by EFS (60V, 0,5ms, 2-64 Hz) in vas deferens.
Values have shown as the means ± SE.



Picture 1: Histopathological appearance of vasdeferens of the groups (H-E stain x100) (a and c); Masson's trichrome stain in both groups, respectively (b and d). L, lumen SE, subephitelium M, muscular layer of the vasdeferens S, serosa. Note that there is no difference between groups.

Discussion

The consumption of carbohydrate-rich diets has increased considerably in both developed and developing countries. Fructose is one of the important carbohydrate which is not easily metabolized in mammals. The proportional increase in fructose consumption far exceeds the increases in intake of other food groups such as carbonated beverages and other sweetened drinks, baked goods, candies, canned fruits, jams, jellies and dairy products (11, 15). The food industry often prefers carbohydrate, particularly fructose to sucrose since they are inexpensive to produce and transport, and fructose mixes well in many foods. In the last four decades, fructose-rich diet consumption has seen a 79.7-fold increase (12).

Several short-term clinical studies have shown that small amounts of fructose have no deleterious effects on metabolism (13,14). Because natural fructose find in relatively small amounts in fruits and vegetables, it is not severe harmful to consume even it be excluded from standart diets (15). High amounts of fructose consumption have been shown to cause hypertriglyceridemia, hyperuricemia and induce insulin resistance. Excessive consumption of fructose may promote obesity, type 2 diabetes mellitus and fatty liver disease (16). In this present study, we showed that a carbohydrate-rich diet which consists of 37–40% fructose caused marked weight gain (27.4%) in the study group whereas it was 9% in the control group after six weeks period. Because fructose does not require insulin for uptake into the cells, the feeling of satiety has been delayed by the fructose. Natural fructose finds in small amounts in fruits and vegetables, and consumption of these foods, not adversely affect blood-glucose levels. Bantle et al. stated that short-term fructose replacement in the diabetic diet improves glycaemic control (17), whereas other studies found that fructose had deleterious effects on other aspects of metabolism (18).

The negative effects of obesity on erectile and bladder function have been investigated in many clinical studies. Increased intraabdominal pressure due to weight gain which provokes stress on the pelvic floor, impaired neuromuscular function (urge incontinence) and the vascular damage due to systemic oxidative stress origin may be given as pathophysiological mechanisms (19,20). This study was designed to investigate whether carbohydrate-rich diet had negative effects on contractility of the vas deferens. In addition, changes on vas deferens histology and response to dapoxetine were also studied.

Adrenergic nerve fibres predominantly finds in the vas deferens. NA is the main neurotransmitter, whereas vasoactive intestinal polypeptide (VIP), somatostatin and leu enkephalin are the other putative substances (21).

Perivasal ganglionic cells also consist of nonadrenergic, noncholinergic, purinergic nerve fibres (22). In this study, the contractile response of the vas deferens to NA and ATP were investigated. Similar responses in both the main groups and subgroups support the fact that obesity does not contribute to vas deferens dysfunctions. In 2013, Mosli et al. reported that Basal Mass Index (BMI) increases were associated with sexual disorders. Although premature ejaculation PE was thought to be reasoned by obesity (23), our study showed that different mechanisms might play a role in ejaculation other than those having a vas deferens origin.

According to the literature, numerous disturbances can be encountered in the aetiology of ejaculatory dysfunction including retroperitoneal lymph node dissection, major pelvic surgeries, aortoiliac reconstruction, inguinal herniorrhaphy, multiple sclerosis, transverse myelitis and diabetes (24-31). As previously reported by Risely in 1963, the distribution of contractile cells and sympathetic nerves within the vas deferens are responsible for the rhythmic peristaltic movements in the vas deferens during emission that help the movement of sperm through the epididymis.

Total contractile strength, which can be measured using EFS, represents the release of all transmitters into the neuromuscular synaptic space. Our EFS results showed no differences in the control and study groups. The addition of dapoxetine in some subgroups showed similar responses to EFS (Fig.3).The selective serotonin reuptake inhibitor, dapoxetine, was originally approved for the on-demand treatment of PE (32). For this reason, dapoxetine was administered in an on-demand fashion in the groups prior to sacrification of the rats (33). Studies that have focused on histopathological changes of the vas deferens are extremely rare (34). In the present study, we showed no histopathological or pharmacological changes in the vas deferens due to carbohydrate-rich diet in rats.

The short follow-up period is a limitation of this study. Further studies are required to aid in better understanding the effects of a carbohydrate-rich diet on the vas deferens.

Conclusion

In conclusion, although the carbohydrate-rich diet caused weight gain over a short period of time, pharmacological and histological properties remained stable and dapoxetine had no effect on the contractility of the vas deferens. Even though carbohydrate-rich diet have no negative pharmacological or histological effects on the vas deferens, it should be kept in mind that other parts of the male genital tract may have been negatively affected.

Conflict of Interest: The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical issues: All Authors declare that Originality of research/article etc... and ethical approval of research, and responsibilities of research against local ethics commission are under the Authors responsibilities. The study was completed due to defined rules by the Local Ethics Commission guidelines and audits.

Acknowledgement: Author contributions??

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ISSN: 2148-6832

MSD Medical Science and Discovery
2016;3(6):266-9

Case Report Doi: 10.17546/msd.94547

An important paraplegia case due to synthetic cannabinoid bonsai abuse

Serkan Demir[1], Hakan Simsek[2], Mehmet Guney Senol[1], Ahmet Eroglu[3], Ergenekon Karagoz[4], Hakan Tekeli[1], Rifat Erdem Togrul[1]

Abstract

Objective: The number of synthetic derivatives of the world's most popular and commonly abused drug, cannabis is rapidly growing day by day. Although the drug has certain applications in the medical field, due to its wide and easy access and cheapness, more people start trying the drug commonly referred to as 'Bonsai'. This synthetic form of the drug has many different names in different countries. The drug is most commonly referred as "Bonsai" in Turkey, "Spice" in UK and "K2" or "Jamaica" in the US. The synthetic cannabinoids are mainly smoked using a pipe, a hookah (or a bong), or wrapped in cigarette papers. Various cases, including the result of death, have been reported related to 'Bonsai' smoking. We present here a case of 'Bonsai' drug abuse with unusual presentation of paraplegia

Key Words: Bonsai, Cannabinoid, Paraplegia

Introduction

Cannabis was used for both medical and pleasure-inducing purposes in the history. Today, it is still the most popular and commonly abused illegal drug (1). Ibn-I Sina is among the early researchers who discussed the medical utilization of cannabis in his book "El Kanun Fit-Tıbbi" in 10 A.D2.

Cannabis has a wide variety of uses in medicine. By interacting different receptors, it can show anti-ischemic, anxiolytic, analgesic, anti-diabetic, anti-bacterial, anti-epileptic, spasmolytic, anti-tumoral, and anti-inflammatory effects. However, because of its likelihood of being pleasure-inducing, it is also used illegally (2, 3, 4, 5).

Marijuana is the dried leaves of the cannabis plant. It is the most popular drug abused in the US. The production of synthetic cannabinoids, as an alternative to Marijuana, initially started in 2004. The synthetic alternatives have then started to spread rapidly because of their cheapness and commercial availability. This synthetic form of the drug has many different names in different countries. The drug is most commonly referred as "Bonsai" in Turkey, "Spice" in UK and "K2" or "Jamaica" in the US. The synthetic cannabinoids are mainly smoked using a pipe, a hookah (or a bong), or wrapped in cigarette papers. There are many reports of the synthetic drug taken orally as an alternative (6, 7).

The greatest concern regarding the synthetic cannabinoids is their continuous change of structure. While the initially manufactured drugs mostly include JWH-018 and JWH-073, due to close regulations and safety restrictions, a large amount of side-products started to be produced (JWH-081, JWH-122, LWH-210, and AM-2201). According to the report by the European Monitoring Centre for Drugs and Drug Addiction in 2012, out of the 73 psychoactive chemicals, 30 were synthetic cannabinoids. European Union warning system in 2013 included 80 different synthetic cannabinoids (8, 9). When inhaled, the drug's effects start within 1-2 minutes, however oral consumption of the drug may take a longer period of time to start action (10).

In the past several years, a high number of deaths have been reported related to Bonsai' abuse (11). The drug has many harmful side effects including nausea, vomiting, diffuse alveolar damage, tachycardia, abdominal pain and edema. Many patients suffer neuropsychiatric complaints. These neurologic symptoms and findings may include ataxia, coma, confusion, imbalance, headache, muscle weakness, speech impairment, paralysis, epileptic seizure, tremor, dilated pupils and nystagmus.

Received 12-05-2016 **Accepted** 13-06-2016 **Available Online** 15-06-2016
1 GATA Haydarpasa Training Hospital, Department of Neurology, Istanbul, Turkey.
2 GATA Haydarpasa Training Hospital, Department of Neurosurgery, Istanbul, Turkey.
3 Van Military Hospital, Department of Neurosurgery, Van, Turkey.
4 Van Military Hospital, Department of Infectious Diseases and Clinical Microbiology, Van, Turkey
*Corrospending Author Ahmet Eroglu E-mail: drahmeteroglu@gmail.comPhone: +90 506 203 6231

In addition, psychiatric complaints including panic attack, anxiety, irritability, acute psychotic attack, hallucination, agitation, delusion and paranoia may be seen due to 'Bonsai' abuse (12, 13).

The patient who addicted to Bonsai usage for a while, he gave up using Bonsai due to different complaints. Although stopped drug abuse, his clinical status deteriorated and developed paraplegia. Because of its unusual effect and to get attention to potential harmful effects of Bonsai, the case has been thought as significant for presentation.

Case

The 48 year-old male patient came to our clinic with leg weakness and numbness. His occupation was a professional tourist guide. He explained that after six months of using Bonsai, he started to feel numbness in his legs and then stopped using the drug because of his complaints. He had certain pains and aches in his extremities when he first started using the drug, but ignored them. He became more wary of his condition when he started to feel numbness in his pains. He stated that his arms and upper body generally felt better compared to his legs. He also added that sometimes the numbness and the pain were so intense that he couldn't stand by his own.

His initial neurological examination revealed that his upper extremities had bilateral vague paresis (muscle strength 4/5). Lower extremities were paraplegic with the accompanying spasticity was noted.

His deep tendon reflexes were globally brisk, plantar reflexes were bilaterally extensor and Achilles klonus was present bilaterally with 4-5 beats. There were no autonomic findings in the patient. Spinal MRIs showed a symmetrical, non-contrast-enhancing lesion which impacts on bilateral pyramidal tracts (Figures 1,2,3).

Cerebrospinal fluid (CSF) analysis was normal but CSF protein level was 299 mg/dl (normal range 15-45 mg/dl). The blood tests regarding infective, rheumatological and vasculitic diseases revealed no abnormality. Because the patient was a professional tourist guide, 'tropical spastic paraparesis' was suspected.

HTLV 1-2 virus antibodies examined and found to be as negative. Methylprednisolone 1 gr/day was given intravenously for 10 days and a brief recovery period was observed. Intrathecal baclofen pump was used to reduce the spasticity in his legs and some progress has been achieved. The patient was also mobilized with the physical therapist. The diffuse lesions were observed to best able in the further control MRIs. Patient consent form has been approved by the patient for this presentation and article.

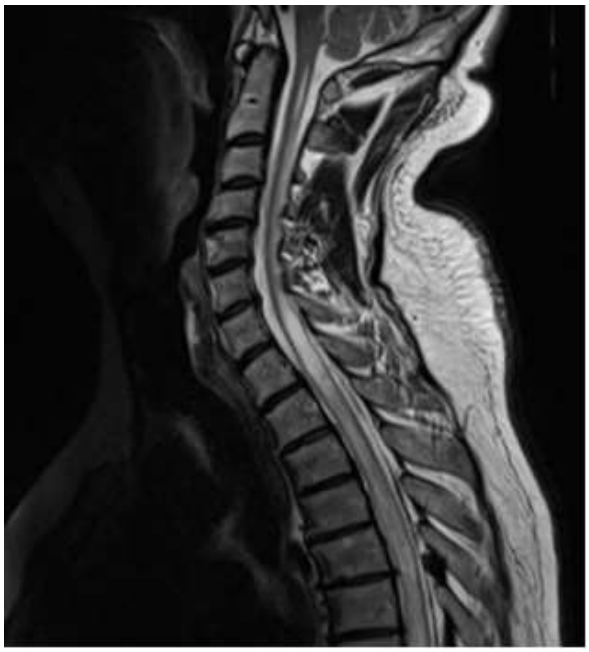

Figure 1: Cervical Spinal MRI showed a diffuse, non-contrast-enhancing lesion

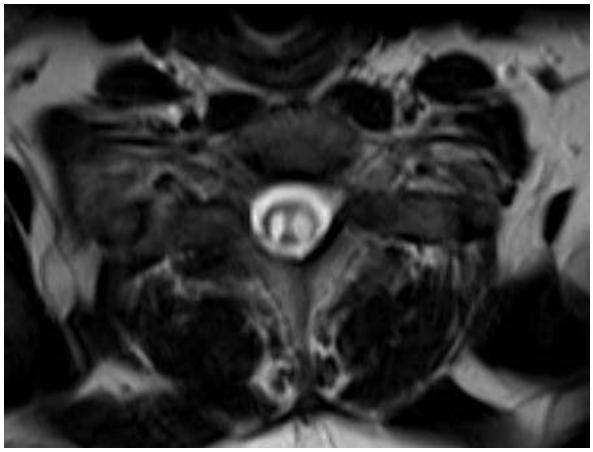

Figure 2: Cervical Spinal MRI showed a symmetrical, non-contrast-enhancing lesion which impacts on bilateral pyramidal tracts

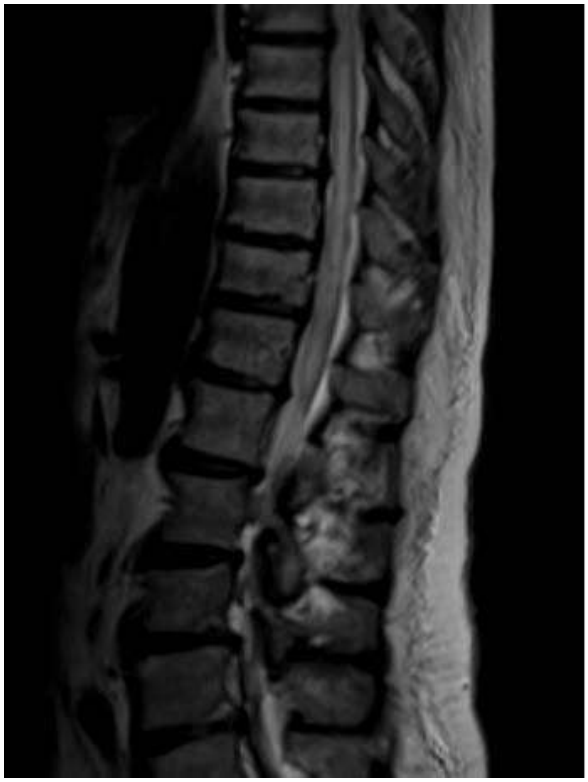

Figure 3: Dorsal and Lomber Spinal MRI showed a diffuse non-contrast-enhancing lesion which impacts on pyramidal tracts

Discussion

New synthetic forms of cannabis, a plant which was used for both medical applications and pleasure-inducing effects in the past, have been developed in recent years. Commonly referred as 'Bonsai' in our country, Turkey, these synthetic drugs are widely abused due to their cheapness and easy accessibility.

There are many reported cases of death because of the 'Bonsai' use. Texas Toxin Center's research with 464 participants revealed that 43% had cardiac, 61.9% had neuropsychiatric and 21.1% had gastrointestinal symptoms. 41% of 464 participants were below 19 years old and the majority of the patients were male (13). Another research in 2010 by the USA National Toxin Center indicated that the most common adverse sign was tachycardia with 37.7% in 1353 patients. In the same research it was reported that 88.2% of the complaints had acute onset and there was only one death (14). Many different clinical presentations found to be reported in the current literature. Our case was unique regarding the patient's complaints have a subacute onset, the patient presents a relatively rare neurologic deficit (paraplegia) and the patient is older in age (48 year old). According to the data from the research of Texas Toxin Center, only 0.2% of the patients had paralysis. As far as to be known, there is only one paper reports generalized paraplegia due to abuse of the synthetic drug "Ninja Strong" (15). It should be noted that serious neurological symptoms, such as muscle weakness, are quite rare.

Hereditary spastic paraplegia was ruled out because of the proven spinal involvement with imaging and his age. Another possible diagnosis, tropical spastic paraparesis was also ruled out since the HTLV 1-2 antibodies were negative. Slightly increased CSF protein levels has been reported, however our patient had a level that was 6 times above the average (16). Although we have noted that the pyramidal tracts were affected across the spine bilaterally and symmetrically in spinal MRIs, we are unable to figure out why the upper extremities were rather unaffected.

Conclusion

Our patient partially benefited from the administration of high dose IV corticosteroids. He also highly benefited from the intrathecal baclofen pump which was used to reduce the spasticity. He was able to walk with support after physical rehabilitation

Conflict of Interest: The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical issues: All Authors declare that Originality of research/article etc... and ethical approval of research, and responsibilities of research against local ethics commission are under the Authors responsibilities. The study was completed due to defined rules by the Local Ethics Commission guidelines and audits.

Acknowledgement: None

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MSD MEDICAL SCIENCE & DISCOVERY
ISSN: 2148-6832
Lycia Press LONDON UK

International Journal of
Medical Science and Discovery
Open Access Scientific Journal
June 2016, Vol.3, No.6
www.medscidiscovery.com