UNIVERSIDAD AUTÓNOMA DE GUERRERO
FACULTAD DE CIENCIAS QUÍMICO-BIOLÓGICAS
FACULTAD DE MEDICINA
UNIDAD DE INVESTIGACIÓN ESPECIALIZADA EN MICROBIOLOGÍA
MAESTRÍA EN CIENCIAS BIOMÉDICAS
Laboratorio de Investigación en obesidad y Diabetes

Tesis

Estudio longitudinal de la composición corporal y del perfil metabólico en personas positivas para *Adenovirus 36*.

Que para obtener el grado de Maestría en Ciencias Biomédicas, presenta:

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Chilpancingo Guerrero; diciembre de 2019.

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ACTA DE APROBACIÓN DE TESIS

En la ciudad de Chilpancingo, Guerrero, siendo los 11 días del mes de julio de dos mil diecinueve se reunieron los miembros del Comité Tutoral designado por la Academia de Posgrado de la Maestría en Ciencias Biomédicas, para examinar la tesis titulada "**Estudio longitudinal de la composición corporal y del perfil metabólico en personas positivas para** *Adenovirus* 36", presentada por la alumna **Adriana Oricel Cástulo Arcos**, para obtener el Grado de Maestría en Ciencias Biomédicas. Después del análisis correspondiente, los miembros del comité manifiestan su aprobación de la tesis, autorizan la impresión final de la misma y aceptan que, cuando se satisfagan los requisitos señalados en el Reglamento General de Estudios de Posgrado e Investigación Vigente, se proceda a la presentación del examen de grado.

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El presente trabajo de Investigación fue realizado en el Laboratorio de Investigación en Obesidad y Diabetes de la Facultad de Ciencias Químico Biológicas de la Universidad Autónoma de Guerrero.

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Durante el tiempo que cursó la Maestría en Ciencias Biomédicas, la C. Adriana Oricel Cástulo Arcos recibió una beca (No. De registro 857645) otorgada por el Consejo Nacional de Ciencia y Tecnología (CONACyT) a los programas de Posgrados de Calidad (PNPC).

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DEDICATORIAS

A quienes son magia en mi vida. ϟ

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¡EN DIOS NADA ME FALTA!

Longitudinal study of body composition and metabolic profile in positive subjects for *Adenovirus 36*

ÍNDICE

Longitudinal study of body composition and metabolic profile in positive subjects for Adenovirus 36

I. RESUMEN

Introducción: El término infectobesidad hace referencia a la implicación que tienen algunos microorganismos en el desarrollo de la obesidad. *Adenovirus 36* (Ad-36) se ha relacionado con la obesidad en estudios en modelos animales y en humanos. **Objetivo:** Analizar la relación de la positividad para el Ad-36 con la composición corporal y el perfil metabólico durante un año de seguimiento en jóvenes guerrerenses. **Métodos:** Se realizó un estudio longitudinal en 150 jóvenes guerrerenses, se integraron dos grupos de acuerdo a la seropositividad para Ad-36, mediante la determinación de anticuerpos específicos contra Ad-36 por el ensayo inmunoabsorbente ligado a enzima (ELISA). También se realizó la detección del DNA viral utilizando el DNA genómico obtenido de leucocitos. Se determinó el perfil bioquímico y la composición corporal en todos los participantes, al inicio, a los seis y doce meses. **Resultados:** Al inicio del estudio, la seroprevalencia del Ad-36 fue del 69%. En la medición inicial las personas seropositivas presentaron menor riesgo de presentar alteraciones en los niveles de colesterol y triglicéridos (OR: 0.39, p= 0.01 y 0.28, p=0.03, respectivamente); y a los doce meses no se observaron cambios significativos. Las personas positivas al DNA viral presentaron menor peso (70.1 vs 62.3) e IMC (26.5 vs 24.2), al inicio del estudio y a los seis meses (peso: 68.8 vs 63.2; IMC 26.1 vs 24.1) en comparación con el grupo control. **Conclusión:** Los jóvenes seropositivos para Ad-36 presentaron un menor riesgo de hipercolesterolemia e hipertrigliceridemia al inicio del estudio, pero no a los seis y doce meses**.** Con la presencia del DNA viral se observó una tendencia a la disminución en la adiposidad corporal, en los primeros seis meses del estudio.

Palabras clave: infectobesidad, Adenovirus 36, estudio longitudinal, perfil bioquímico, antropometría.

II. ABSTRACT

Introduction: Infectobesity refers to the implication that some microorganisms have in the development of obesity. Adenovirus 36 (Ad-36) has been linked to obesity in studies in human and animal models. **Aim:** We evaluated the relationship of seropositivity for Ad-36 with body composition and metabolic profile during a year of tracking in young people from the state of Guerrero. **Methods:** A longitudinal study was conducted in 150 young individuals from of the state of Guerrero, two groups were integrated according to seropositivity for Ad-36, by enzyme-linked immunosorbent assay (ELISA). Viral DNA was detected using genomic DNA obtained from leukocytes. The biochemical profile and body composition were determined in all participants, at the beginning of the study, at six and twelve months. **Results:** At the beginning of the study, the seroprevalence of Ad-36 was 69% and seropositive individuals had a lower risk of presenting alterations in cholesterol and triglyceride levels (OR: 0.39, *p*=0.01 and 0.28*, p*=0.03, respectively); and at twelve months no significant changes were observed. Positive individuals to viral DNA had lower weight (70.1 vs 62.3) and BMI (26.5 vs 24.2) at the beginning and at six months after (weight: 68.8 vs 63.2; BMI 26.1 vs 24.1) in comparison to the control group. **Conclusion:** At the beginning of the study, seropositive individuals had lower risk of hypercholesterolemia and hypertriglyceridemia, however at six and twelve months this low risk was not maintained. Positive individuals to the presence of viral DNA Ad-36, tend to lower body fat in the first six months of the study.

Key words: Infectobesity, Adenovirus 36, longitudinal study, biochemical profile, anthropometry.

III. INTRODUCTION

Obesity is the abnormal or excessive accumulation of fat that can be harmful to health (WHO). For its diagnosis, the calculation of the body mass index (weight in kg / height in m^2) is used, and a body mass index major than or equal to 30 is considered as obesity (Genoni *et al*., 2014). The presence of this disease is related to the development of comorbidities, including diabetes, hypertension, dyslipidemias, among others (González *et al*., 2011). It is currently known that obesity can have two phenotypes, obesity with metabolic disorders and metabolically healthy obesity (Gruberg *et al*., 2002; Lavie *et al*., 2009). The absence of metabolic alterations can be attributed to the distribution of adipose tissue and the absence of systemic inflammation (Antonopoulos *et al*., 2017; Jung, 2017).

Several factors are involved in the development of obesity, among them, the genetic one, since numerous studies have shown that the predisposition to obesity, and their comorbidities, are more common among genetically related individuals than in those unrelated, in addition diverse environmental factors, such as lack of physical activity and a hypercaloric diet; Also, the obesity has been related to presence of some infectious agents, mainly of viral origin (Dhurandhar *et al*., 1997; Dhurandhar *et al.,* 2002; Voss and Dhurandhar 2017).

The infectious agent that has been most related to the development of obesity in humans is Adenovirus type 36 (Ad-36), whose genome consists of a single molecule of double stranded linear DNA (35, 152 base pairs). This virus can cause respiratory, ocular and gastrointestinal infections (Saha, Wong and Parks 2014; J-H Nam *et al*., 2014). The association between Ad-36 and obesity has been observed in the American, Italian and Korean adult population (Atkinson *et al*., 2005; Na *et al*., 2012; Trovato *et al*., 2009). In a meta-analysis of studies conducted in adults, it was observed that seropositive for Ad-36 have a higher risk of being overweight and obese (Xu *et al*., 2015). In the US and Swedish population, seropositivity for the virus has been particularly related to the metabolically healthy obesity phenotype (Vander Wall *et al*., 2013; Almgren *et al*., 2014)

Furthermore, in chickens, mice and nonhuman primates have shown that Ad-36 can promote obesity. There is evidence that in the 3T3-L1 cell line, Ad-36 induces differentiation of preadipocytes to mature adipocytes (Rathod *et al*., 2007). In mice of strain C57BL / 6J, has been showed that Ad-36 decreases insulin resistance, regardless of dietary fat intake or adiposity. It has been proposed that Ad-36 increases glucose cell uptake through activation of the Ras-mediated phosphatidyl inositol 3-kinase (PI3K) signaling pathway, which decreases hyperglycemia in mice (Dhurandhar *et al*., 2011).

Since most of the studies in which the seropositivity relationship for Ad-36 with obesity is evaluated, are of a transversal type, we carry out a tracking study, with a duration of one year, in order to determine the changes in metabolic profile and body composition in seropositive and negative individuals for Ad-36 or viral DNA.

IV. MATERIALS AND METHODS

4.1 Participants

The protocol was designed as case and control study for evaluate the relation between seropositivity for Ad-36 and the changes of clinical and anthropometric measures over a year of tracking. One hundred fifty young university students from the Guerrero State, between 18 and 30 years old were included. According to seropositivity for Ad-36, two groups were formed, one group was integrated for 104 seropositive individuals (cases group) and the other group included 46 seronegative individuals (control group). Individuals who agreed to participate in the study were signed an informed consent. During the tracking study, at the beginning, at six- and twelve-months surveys were applied to each of the participants, which included sociodemographic data, family history of diseases and lifestyle.

4.2 Clinic and anthropometric measurements

During the tracking study, an analysis of body composition was performed on all participants. Measurements of weight and height were performed to calculate the body mass index (BMI). A TANITA MC-7800U scale was used for the evaluation of

body composition (fat mass, muscle mass, and visceral fat level), a portable stadiometer was used to measure height (Seca, Hamburg, Germany). Blood pressure was measured using an automatic Baumanometer (OMROM).

4.3 Laboratory determinations

Venous blood sample was obtained from each participant after they fasted for 8 h to determine serum glucose, total cholesterol, HDL cholesterol, LDL cholesterol and serum triglycerides by standard enzymatic colorimetric methods (Spinreact). The blood samples were obtained during the tracking study in at the beginning, at six and twelve months.

4.4 Seropositivity detection for Ad-36

The qualitative determination of specific antibodies against Ad-36 was carried out by means of an enzyme-linked method based on the principle of sandwich type ELISA of the MyBioSource brand (No. Catalog: MBS9310682); according to the manufacturer instructions. Serum samples were processed and classified as seronegative or seropositive according to recommended cut-off absorbance.

4.5 Viral DNA detection

A blood sample was used for DNA extraction from leukocytes, using the modified Miller technique. A fragment corresponding to the gene coding for the hexon viral protein was amplified using the following primers: sense (5'- AGT CAG TGG ACT GAC AAA GAA CG -3´) and antisense (5'- GTC TGC ATA TAT CTC TTC TTC ACC-3 '). The PCR product was visualized by electrophoresis in 6% polyacrylamide after that gel was stained with 0.3% silver nitrate.

4.6 Statistical analysis

The statistical analysis of the results was performed in the STATA v.14.0 (College Station, Texas USA). After checking the normality of the data, means and standard deviations were obtained as measures of central tendency and dispersion, respectively. The comparison of the groups was made using the student's *t*-test. The

comparison between the measurements obtained at the beginning of the study (initial) and tracking (6 and 12 months) was performed using repeated measures ANOVA with random effects. The changes in biochemical parameters according to serological status were estimated using a simple linear regression model. To identify risk factors associated with metabolic disorders such as: fasting impaired glucose (> 100 mg / dL), hypercholesterolemia (> 200 mg / dL) and hypertriglyceridemia (> 150 mg / dL), a multiple logistic regression was performed. A value of $p<0.05$ was considered significant.

V. RESULTS

At the beginning if the study, 150 young individuals were included, 104 individuals (69%) were seropositive for Ad-36 and 46 (31%) were negative. In the second measurement, at six months, 92 (61.3%) were seropositive and 36 (24%) negative individuals were analyzed. In the last measurement one year later, 68 (45.3%) seropositive and 29 (19.3%) negative individuals were evaluated. The monitoring diagram is shown in Figure 1.

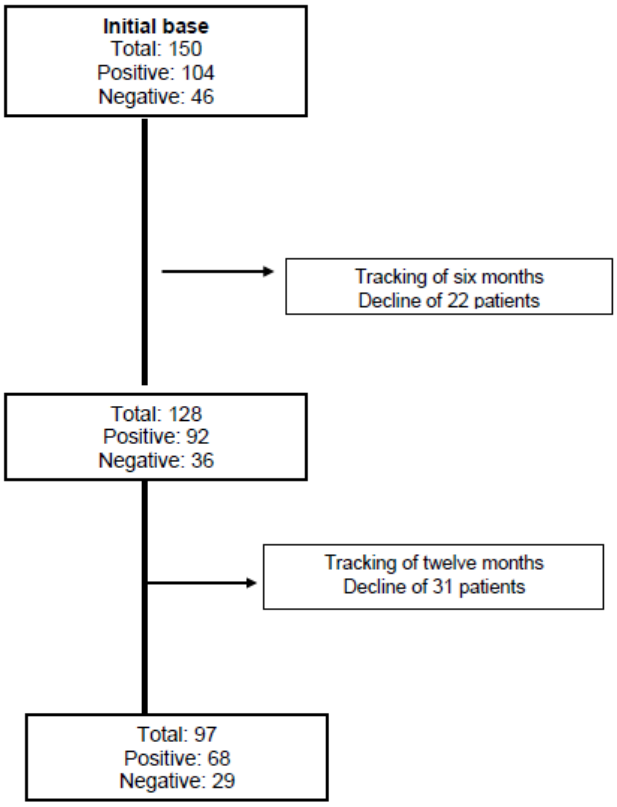

Figure 1 Diagram of tracking that shows the changes in the number of positive and negative patients throughout the study.

Biochemical profile and anthropometric measurements of study groups according to seropositivity for Ad-36

The clinical and metabolic characteristics between the seropositive and negative groups for Ad-36, in the three stages of the tracking study were summarized in the table 1. Significant differences were found in glucose levels, triglycerides and cholesterol, as well as weight in these parameters the seropositive people to Ad-36 were obtained lower values compared to seronegative patients (p = 0.04, <0.01, p <0.01 and 0.05, respectively).

The results also were showed that seropositive individuals had lower levels of triglycerides, total cholesterol, LDL cholesterol and visceral fat, without statistical significance (Table 1).

Table 1. Comparison of biochemical profile, clinical and anthropometric measurements between groups according to seropositivity to Ad-36.

Parameter	Ad-36 (-) N=46	Ad-36 (+) N=104	*p value*
	Initial		
Glucose (mg/dL)	81.1±10.1	77.8±8.8	**0.04**
Triglycerides (mg/dL)	110.5±59.7	86.3±41.7	**<0.01**
Cholesterol (mg/dL)	184.2±41.8	163.9±37.7	**<0.01**
HDL-C (mg/dL)	41.1±11.7	39.6±3.6	0.21
LDL-C (mg/dL)	92.6±32.4	97.4±34.6	0.42
Weight	70.6±18.5	64.6±16.8	0.05
BMI	26.5±5.8	24.9±5.3	0.12
% of fat	27.6±9.8	28.8±7.6	0.40
Waist circumference	82.7±18.5	79.1±13.2	0.18
Hip circumference	100.1±14.1	97.9±10.8	0.31
Visceral fat	4.6±4.2	3.6±3.3	0.10
SBP	110±13	107±15	0.19
DBP	67±11	64±9	0.12
	Six months		
	N=36	**N=92**	
Glucose (mg/dL)	73.5±7.1	73.7±7.4	0.90
Triglycerides (mg/dL)	112.2±54.3	99.6±59.9	0.27
Cholesterol (mg/dL)	166.4±27.5	160.7±31.9	0.34
HDL-C (mg/dL)	37.3±2.9	38.5±9.1	0.45
LDL-C (mg/dL)	105.2±25.9	95.3±30.6	0.09
Weight	69.1±15.3	65.3±15.9	0.22
BMI	25.9±4.6	24.8±5.1	0.26
% of fat	26.1±8.3	27.6±7.4	0.33
Waist circumference	87.2±11.7	83.1±13.5	0.12
Hip circumference	100.2±9.4	98.5±11.1	0.42
Visceral fat	4.1±3.2	3.4±3	0.11
SBP	111±11	109±11	0.50
DBP	66±9	68±11	0.33
	Twelve months		
	N=29	**N=68**	
Glucose (mg/dL)	83±11.3	82.4±12.7	0.85
Triglycerides (mg/dL)	113.6±55.5	110.2±55.2	0.79
Cholesterol (mg/dL)	161.7±30.9	153.4±31.7	0.25
HDL-C (mg/dL)	38.4±3.5	38.1±5.2	0.73
LDL-C (mg/dL)	94.5±33.4	84.5±27.7	0.13
Weight	67.8±13.2	64.3±15.9	0.32
BMI	25.3±4.1	24.8±4.9	0.63
% of fat	27.2±8.3	28.1±7.5	0.63
Waist circumference	83.5±12.2	81.1±16.4	0.48
Hip circumference	99.4±7.9	96.0±14.9	0.27
Visceral fat	3.9±2.6	3.4±3	0.47
SBP	110±13	106±15	0.31
DBP	68±10	69±11	0.82

Values are shown as mean ± standard deviation.

p: Student's *t*-test.

HDL high density lipoprotein, LDL low density lipoprotein, BMI body mass index, SBP systolic blood pressure, DBP diastolic blood pressure.

With the aim to emphasize the most important differences that were observed in metabolic profile between seropositive and negative individuals, the parameters of glucose, total cholesterol and triglycerides were graphed. The results were showed that seropositive individuals have a better metabolic profile, compared to seronegative individuals (figure 2).

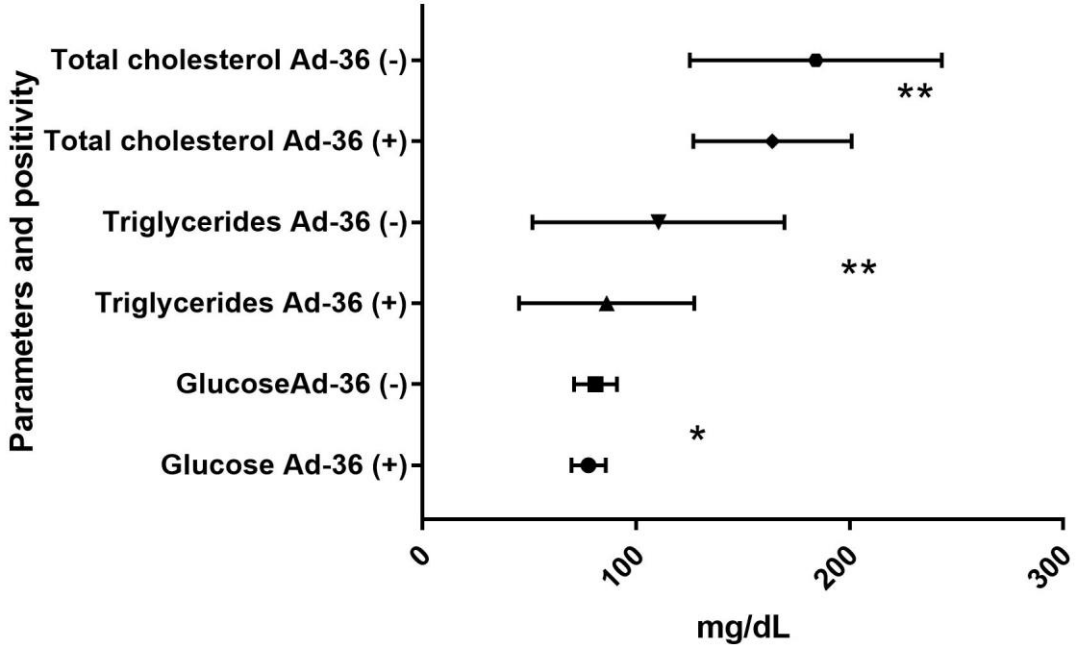

Figure 2 Biochemical parameters in which statistically significant differences were observed between the groups in the initial measurement.

Association between seropositivity for Ad-36 and metabolic alterations

A logistic regression analysis was performed to determine the association between Ad-36 status and the presence the metabolic alterations in the three stages of tracking study. The results showed that seropositive individuals have a lower risk of alterations in cholesterol levels (OR: 0.39, $p = 0.03$) and triglycerides (OR: 0.28, $p = 0.01$) in the initial measurement (Table 2).

Table 2. Association between seropositivity for Ad-36 and metabolic alterations.

Variable	OR (CI 95%)	p
	Initial	
Glucose ≥100 mg/dL	0.16 (0.01-1.6)	0.12
Cholesterol ≥200 mg/dL	0.39 (0.17-0.90)	**0.03**
Triglycerides ≥150 mg/dL	0.28 (0.10-0.79)	**0.01**
LDL-C ≥100 mg/dL	1.5 (0.70-3)	0.32
	Six months	
Glucose ≥100 mg/dL	*	*
Cholesterol ≥200 mg/dL	1.1 (0.31-3.7)	0.91
Triglycerides ≥150 mg/dL	0.69(0.26-1.8)	0.46
LDL-C ≥100 mg/dL	0.60(0.27-1.3)	0.21
	Twelve months	
Glucose ≥100 mg/dL	*	*
Cholesterol ≥200 mg/dL	0.55 (0.08-3.6)	0.52
Triglycerides ≥150 mg/dL	0.55(0.18-1.7)	0.29
LDL-C ≥100 mg/dL	0.63(0.24-1.67)	0.35

Logistic regression. Models adjusted for age and gender.
LDL low density lipoprotein.
* None individual had alterations

Changes in biochemical profile and anthropometry throughout tracking study in seropositive and negative individuals

To evaluate changes in glucose, total cholesterol, triglycerides and LDL-C levels and the anthropometry respect to the initial measurement to one year later in both groups, the delta was calculated, the results were indicate that glucose levels decreased in both individuals seropositive and negative (p <0.01), however at twelve months the values in both groups were increased again, however, only in the seropositive individuals, this increase was statistically significant. Triglyceride levels increased in both groups, but in seropositive the change was greater. Total cholesterol levels decreased in both groups, with a greater effect on seronegative

individuals. LDL cholesterol only decreased in the seropositive group, and at twelve months the change in LDL cholesterol values were statistically significant (Table 3).

Table 3. Changes observed in the metabolic profile with respect to the initial measurement

	Initial	6 months			12 months		
Parameter	**Media ± DE**	**Media ± DE**	Δ	***p***	**Media ± DE**	Δ	***p***
Glucose							
Negative	81.1 ± 10	73.5 ± 7	-8.0	**<0.01**	83±11	1.8	0.45
Positive	77.8 ± 9	73.7 ± 7	-4.0	**<0.01**	82.4 ± 12	4.6	**<0.01**
Triglycerides							
Negative	110.5 ± 59	112.2 ± 54	3.6	0.77	113.6 ± 55	6.6	0.62
Positive	86.3 ± 42	99.6 ± 59	12.4	0.10	110.2 ± 55	22.2	**<0.01**
Cholesterol Total							
Negative	182 ± 41	166.4 ± 27	-17	**0.03**	161.7 ± 30	-21.5	**0.01**
Positive	163.9 ± 37	160.7 ± 31	-3.8	0.44	153.4 ± 31	-11.1	**0.04**
LDL-C							
Negative	92.6 ± 32	105.2 ± 25	11.9	0.09	94.5 ± 33	1.8	0.81
Positive	97.9 ± 10	95.3 ± 30	-1.6	0.71	84.5 ± 27	-12.8	**0.01**

Table shows the Δ in studied population. Simple linear regression. LDL low density lipoprotein

Detection of viral DNA in leukocytes

Ad-36 DNA was detected from peripheral blood leukocytes to evaluated whether the presence of viral DNA in these cells was associated with changes in the biochemical and / or anthropometric profile during tracking study. At the beginning of the study the viral DNA was detected in 46% of the population, at six months the prevalence decreased to 19% and at twelve months it increased to 41% (Figure 3).

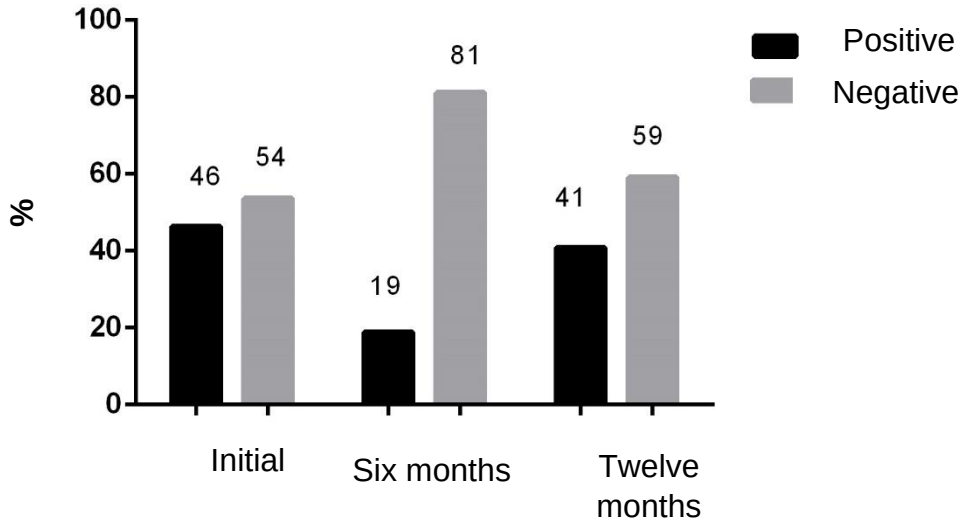

Figure 3. Frequency of Ad-36 DNA detection at initial measurement, at 6 and 12 months

Characteristics of biochemical profile and anthropometric measurements of study groups according to presence of viral DNA during tracking study.

To determine the relationship of the metabolic profile with the presence of Ad-36 DNA in the year of the tracking study, several biochemical parameters were evaluated. The results showed that individuals positive for viral DNA had lower glucose levels (p=0.02) with respect to the individuals negative to the viral DNA in the initial evaluation, however after six and twelve months no significant differences were observed between the study groups.

Regarding the anthropometric measurements, in the three measurements carried out it was observed that individuals positive to viral DNA have lower values of visceral fat, BMI, fat percentage, weight, as well as waist and hip circumference (Table 4).

Table 4. Comparison of biochemical profile, clinical and anthropometric measurements between groups according to the presence of viral DNA.

Parameter	Ad-36(-) N=80	Ad-36 (+) N=70	*p* value
	Initial		
Glucose (mg/dL)	80.3±10.3	77±7.8	**0.02**
Triglycerides (mg/dL)	94.4±48.8	92.8±49.6	0.85
Cholesterol (mg/dL)	175.9±40.3	163.4±38.8	0.05
HDL-C (mg/dL)	39.4±3.3	40.7±9.8	0.25
LDL-C (mg/dL)	95.3±35.7	96.7±31.9	0.79
Weight	70.1±18.1	62.3±15.9	**<0.01**
BMI	26.5±5.9	24.2±4.8	**0.01**
% of fat	29.1±8.5	27.7±8.1	0.31
Waist circumference	82.3±14.1	77.9±15.8	0.06
Hip circumference	100.7±11.9	96±11.6	**0.01**
Visceral fat	4.4±4	3.3±3	0.08
SBP	108±16	107±12	0.63
DBP	65±10	65±9	0.64
	Six months		
	N=99	**N=23**	
Glucose (mg/dL)	73.7±7.3	73±7.3	0.84
Triglycerides (mg/dL)	103±64.3	103.5±50.6	0.96
Cholesterol (mg/dL)	160±32.9	165.3±27.8	0.33
HDL-C (mg/dL)	38.6±9.9	37.6±3.5	0.50
LDL-C (mg/dL)	93.8±32.3	103.6±24.8	0.06
Weight	68.8±15.8	63.2±15.3	**0.04**
BMI	26.1±5.2	24.1±4.4	**0.01**
% of fat	27.9±7.8	26.2±7.5	0.21
Waist circumference	85.6±13.2	82.6±12.8	0.19
Hip circumference	100.6±11	96.9±9.9	0.05
Visceral fat	4±3.2	3.1±2.8	0.06
SBP	110±12	109±10	0.85
DBP	67±9	67±12	0.82
	Twelve months		
	N=55	**N=38**	
Glucose (mg/dL)	81.6±15.3	83.7±7.1	0.42
Triglycerides (mg/dL)	103.4±51	120.5±58.7	0.13
Cholesterol (mg/dL)	152.6±35.4	159.7±25.8	0.28
HDL-C (mg/dL)	38.7±5.3	37.5±3.8	0.19
LDL-C (mg/dL)	88.7±32.3	86.2±26.5	0.68
Weight	67.1±14.2	63.1±16.2	0.21
BMI	25.5±4.6	24.2±4.6	0.21
% of fat	28±8.4	27.6±6.9	0.79
Waist circumference	81.7±6.9	81.1±13.1	0.99
Hip circumference	97.4±16.1	96.6±8.9	0.77
Visceral fat	3.6±2.7	3.5±3.1	0.08
SBP	106±16	109±12	0.21
DBP	67±11	70±9	0.16

Values are shown as mean ± standard deviation.
p: Student's t-test.
HDL high density lipoprotein, LDL low density lipoprotein, BMI body mass index, SBP systolic blood pressure, DBP diastolic blood pressure.

To emphasize the most important differences that were observed in the anthropometry between positive and negative individuals to Ad-36 DNA, the parameters evaluated were graphed. Positive individuals to Ad-36 had lower adiposity values compared to negative individuals (Figure 4).

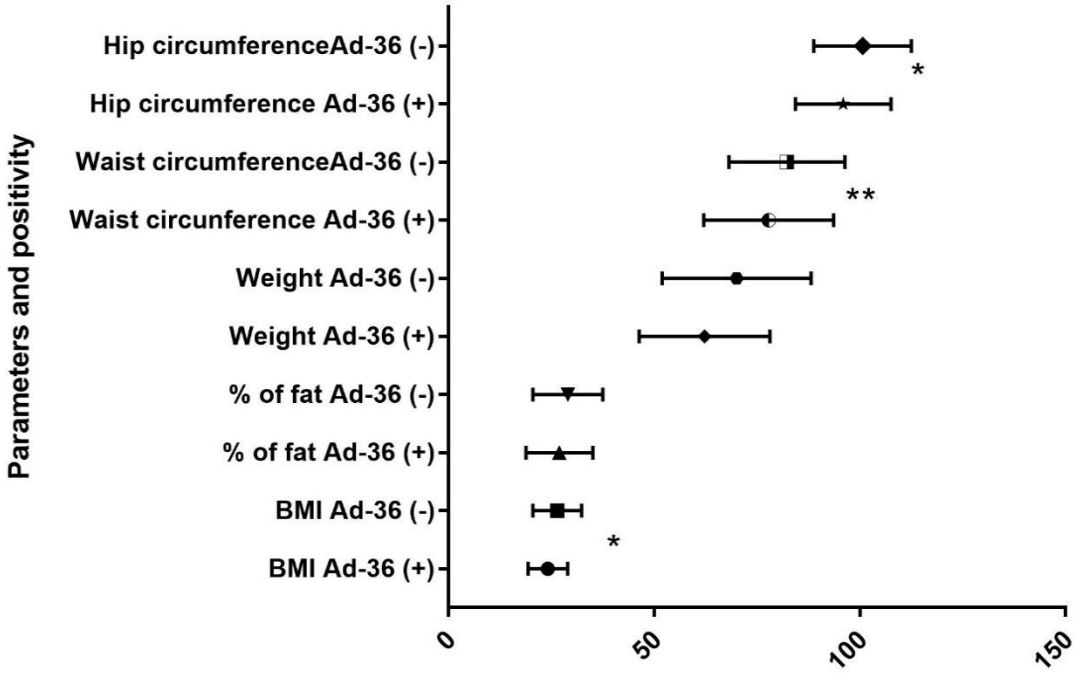

Figure 4 Anthropometric parameters in which statistically significant differences were observed between the groups in the initial measurement.

A logistic regression was performed to determine the association between the viral DNA presence and metabolic alterations in the three measurements carried out during tracking study, but no significant results were found (data not shown).

VI. DISCUSSION

In the present tracking study, in the initial measurement, seropositive individuals to Ad-36 presented a lower risk of having alterations in cholesterol and triglyceride levels, in addition to having lower levels of glucose and LDL cholesterol, these data

coincides with studies carried out by Almgren in 2012 and by Atkinson in 2005 in Swedish and US populations, respectively, they found that seropositive people had lower lipid levels contrary to observed in the Italian population, the cholesterol levels were higher in the seropositive group, characteristics also described in children by H-N Na and collaborators in 2010, they found that cholesterol and triglycerides were higher in seropositive infants. In animal models, to Ad-36 infection, there also decreases in blood cholesterol and triglyceride levels a few days after the inoculation of the virus (Dhurandhar, *et al*., 2002; Dhurandhar, *et al*., 2006). In the cross-sectional, studies, a better metabolic profile has been observed in seropositive individuals, however the moment in which they acquired the infection is unknown. In the present study, after six months of tracking, the decreased levels in the lipid profile are not maintained. According to the metabolic characteristics observed in the animal models, in our study group it could be inferred that the seropositive individuals had acquired the infection to Ad-36 in relatively recent way, because the favorable metabolic changes have been observed only at the beginning of the viral infection.

Regarding longitudinal studies, in 2013 in a diabetic population of Hispanic origin, it was observed that seropositive people to Ad-36 had better glycemic control, as well as decreased fasting insulin levels (Lin *et al*., 2013). It should be mentioned that after 12 months, seropositive people presented a significant increase in glucose levels. However, in another study, seropositivity for the virus was common in individuals with normal glucose tolerance compared to type 2 diabetes mellitus patients. In that same study, women categorized as prediabetic, have a greater sensitivity to insulin (Almgren *et al*., 2014), this study suggested that sensitivity to insulin are related to the ability of the virus to increase glucose uptake independently of the proximal signaling of insulin, this part of signaling pathway is generally affected in people with type 2 diabetes (Kusminski *et al*., 2015; Shirani *et al*., 2017; Yoon *et al*., 2017).

In the determination of the presence of viral DNA in peripheral blood leukocytes, the frequencies were similar both at baseline and at 12 months, it is worth mentioning that this is the first study where the identification of Ad-36 DNA is carried out in this type of cells. Usually, the detection is carried out in samples of adipose tissue, in the

first study of this type in 2015, 49 samples from obese people and 49 with normal weight were included, however, in none of them was viral DNA identification achieved (Ergin *et al*., 2015). In another study by Ponteiro and collaborators they identified the DNA in four samples from adipose tissue, two people were obese and two overweight, all individuals had normal fasting glucose and insulin levels (Ponterio *et al*., 2015). In the present study, different frequencies in viral DNA detection were observed in the three measurements, which can be attributed to the differences in the immunological condition of the host, in addition to the fact that tropism is not exclusive for this type of cells and preferentially infects epithelial cells (Narvaiza *et al*., 2003).

In the population studied, the individuals positive to the viral DNA has lower BMI, visceral fat level, waist and hip circumferences, compared to negative DNA virus subjects. The decrease in these parameters has been observed in some populations where intervention has been carried out for weight reduction, in female one population of Czech origin this intervention was performed in seropositive and seronegative people to Ad-36, it was determined that there was a greater decrease in anthropometrical measures in seropositive people (Zamrazilova *et al*., 2015). In another intervention study conducted in children of Korean origin, seropositive and seronegative to Ad-36 were subjected to a supervised exercise regime, the decrease in BMI levels was greater in the seropositive to Ad-36 (Xu *et al*., 2015). However, in a one-year tracking study in adolescents of Korean origin, an increase in body fat was observed (Park *et al*., 2015).

In cross-sectional studies where the positivity to the Ad-36 has been identified, it has been consistently observed that seropositive people have greater weight, BMI, fat percentage, as well as waist and hip circumference with respect to seronegative individuals (Atkinson *et al*., 2005; Trovato *et al*., 2009; Ergin *et al*., 2015). This could be explained because *in vitro* studies, in cell lines and in cells from human tissue samples, has been shown that Ad-36 increases adipocyte differentiation and lipid accumulation thereby promoting the development of obesity (Dhurandhar, *et al*., 2016; McGraw, *et al*., 2016).

The mechanisms by which Ad-36 could influence biochemical and / or anthropometric parameters have not been elucidated, however, it is known that viruses can use and modify cellular machinery by altering the expression of different genes (Sang *et al*., 2018). In skeletal muscle cells obtained by biopsies from normal-weight male and infected with Ad-36; a microarray was carried out to evaluate the differential expression of genes of the infected and nor infected cells. After the corresponding evaluation it was determined that most of the altered genes in the cells that were infected are involved with the development, immune response, signal transduction, transcriptional regulation, as well as carbohydrate, lipid and protein metabolism (Wang *et al*., 2012).

At the beginning of this study, the seropositive population presented a lower risk of alterations in cholesterol and triglyceride levels, however, during tracking study (at 6 and 12 months) the groups have a similar metabolic profile. Furthermore, throughout the tracking study, body fat measures were kept constantly low in people positive to viral DNA. In the populations that have been studied to find the relationship of obesity, with the infection of Ad-36 and the changes in the metabolic profile, the results obtained have been inconsistent, this can be attributed to the method of detection of antibodies and viral DNA, to the diagnostic criteria of metabolic alterations and to the genetic background of the populations studied.

VII. CONCLUSION

At the beginning of the study, seropositive individuals had lower risk of hypercholesterolemia and hypertriglyceridemia, however at six and twelve months this low risk was not maintained. Positive individuals to the presence of viral DNA Ad-36, tend to lower body fat in the first six months of the study.

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