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Can physical education state policies impact on youth's health behaviors? A natural experiment study

Instrução normativa da educação física impacta nos comportamentos de saúde? Um experimento natural

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ABSTRACT

Since 2011 an educational policy promulgated that public schools from Pernambuco have had to offer physical education (PE) class in the same shift where students are enrolled. This study examined the impact of the implementation of an educational policy on PE offering and students' participation, and whether health related behaviors could be moderated by PE participation. It was a natural experiment study performed with data obtained from two cross-sectional studies (2007 and 2012) of a sample (n = 715) of high-school students from Caruaru. PE offering was assessed by asking students if they had PE class and adapted questionnaire was used to assess health-related information. It was observed that before policy implementation, most of the students (♂: 72.4%; ♀: 69.0%) was not engaged in any PE class during the week. After policy implementation the proportion of students who had at least one PE class/week increased (♂: 68.7%; ♀: 68.9%). Having ≥1 PE classes was not associated with the amount of physical activity either before (♂: OR = 1.47 (95%CI: 0.78 – 2.76)); ♀: OR = 1.02 (95%CI: 0.61– 1.72) or after (♂: OR = 0.90 (95%CI: 0.51 – 1.58); ♀: OR = 1.06 (95%CI: 0.63 – 1.80)) policy implementation. Fruit consumption was the only health-related behavior associated to PE class (♂: OR = 1.55 (95%CI: 1.01 – 2.70); ♀: OR = 1.48 (95%CI: 1.02 – 2.10)). PE offering and participation of students improved and it seems that implementation of new policies for PE might impact on students' behaviors, although, regarding to some limitations, not sufficiently to impact on overall students' health behaviors.

Keywords: Adolescents; Physical activity; Health status indicators.

RESUMO

Desde 2011 uma instrução normativa estipulou que escolas públicas de Pernambuco ofertassem aulas de educação física (EF) no turno em que estudantes estão matriculados. Este estudo examinou o impacto dessa normativa na oferta de aulas de EF, na participação e sua associação com comportamentos de saúde dos estudantes. Tratou-se de um experimento natural realizado com dados de dois estudos transversais (2007 e 2012) de uma amostra (n = 715) de estudantes do ensino médio de Caruaru. A oferta de EF foi avaliada perguntando aos estudantes se eles tinham aula de EF, e um questionário adaptado foi utilizado para avaliar comportamentos de saúde. Verificou-se que antes da implementação da normativa a maioria dos estudantes não tinha aulas de EF (♂: 72,4%; ♀: 69,0%). Depois da implementação, a proporção de estudantes que tiveram uma aula de EF/semana aumentou (♂: 68,7%; ♀: 68,9%). Ter ≥1 aula de EF não foi associado o nível de atividade física em adolescentes antes (♂: RO= 1,47 (95%IC: 0,78–2,76); ♀: RO = 1,02 (95%IC: 0,61 – 1,72)) ou depois (♂: RO = 0,90 (95%IC: 0,51 – 1,58); ♀: RO = 1,06 (95%IC: 0,63 – 1,80)) da implementação. Consumo de frutas foi o único comportamento de saúde associado com as aulas de EF (♂: RO = 1,55 (95%IC: 1,01 – 2,70); ♀: RO = 1,48 (95%IC: 1,02 – 2,10). Após implementação da normativa houve melhorias na oferta de aulas de EF e participação dos estudantes. Parece que isto pode impactar comportamentos de saúde em adolescentes, porém, devido limitações, é insuficiente para impactar os comportamentos de saúde no geral.

Palavras-chave: *Adolescentes; Atividade física; Indicadores básicos de saúde.*

Introduction

Physical education (PE) class is a key component for promoting health-related behaviors among youth. During the past decades, evidence has accumulated on the positivie association between PE and several outcomes, such as motor skills development[1], physical fitness[2], improvement in daily physical activity[3], health perception and nutrition[4]. In addition, participation in PE class can improve cognitive and academic skills[5], positive attitude[3,6] and health behaviors[3]. It was found by a Brazilian state representative study[4] that high-school students enrolled in PE classes had higher chances to be physically active and to fruit consumption.

Given the importance, institutional initiatives and

policies[7] from countries around the world have advocated to guarantee high quality PE classes. Such as the United Nations Educational, Scientific and Cultural Organization (UNESCO) and partners, who in 2015 released a guideline to support governments to develop and implement inclusive physical education quality policy[7].

In Brazil, PE is a mandatory curricular component in primary and secondary education[8]. Although the draft law by the Federal Senate of Brazil (art. 26 of Law nº 9,394/96) had established a minimum of 2 hours/week of PE classes, not all public schools are regularly offering throughout elementary and high school[9]. Besides, several municipal, state and national-level studies have also reported low participation of students in PE classes[10]. Additionally, non-participation is higher among high school students from the Northeast region, when compared to other Brazilian regions[11] and to elementary and middle school[9,12].

It should be noted that there are many reasons for this lack of participation among high school students. Indeed, several associated factors have been listed to explain the low participation in PE class, such as students' motivation[10], quality-based classes (i.e. teaching methodologies and lack of materials)[10,13], low status of PE[14] and school environment (i.e. infrastructure and schedule)[15,16].

Nonetheless, there is a wide variability of participation between states and regions, which may be related to a macro-level influence (e.g. policy), more than intra or interpersonal factors. In 2008, Pernambuco State government promulgated an educational policy that regulated several aspects of the public-school systems. These included the implementation of the new educational curricula, which since 2011 occurred to PE class to be offered in the same shift where students are enrolled[17]. Feitosa and colleagues[18] have previously indicated the importance to offer PE class during the same shift where high-school students are enrolled.

This situation provided an opportunity to conduct a natural experiment to examine the impact of state educational policy on the offer of PE class and students' participation. Natural experiment studies are often recommended to evaluate and understand the impact of policies (i.e. health, and educational) on individuals or cluster behavior[19]. Although, to date, there is no evidence reporting the impact of ongoing educational policies implementation on the offer of physical education class and students' participation. For that matter, in this study, it is expected to find an increase of the offer of PE classes in high school and also an increase of students' participation since then. Furthermore, if there was an increase in the offer of PE class, would that be positively impacting on students' health outcomes (e.g. physical activity, reduced TV viewing, fruit and vegetable consumption) when comparing to the period before educational policy implementation?

Thus, the aims of this study were to examine the impact of educational policy on physical education offer and high school students' participation, and whether health related behaviors could be moderated by PE participation. We hypothesized that there was an increase on access to PE class and it had a differential impact on students' health outcomes from 2011 onwards.

Methods

The present study employed a natural experiment to examine the impact of a state educational policy on physical education class offers and participation, and then on students' health behaviors. All data were obtained from two cross-sectional studies of a representative sample of high-school students from Caruaru city, located in the dry region of Pernambuco state, 120 kilometers away from Recife, the capital city. The studies comprised data before (2007) the educational policy (2011), and one-year after (2012). All studies were approved by the ethics committee of Associação Caruaruense de Educação Superior (CAAE: 07296612.3.0000.5203 (2007, 2012), and previous publication can be found elsewhere[18].

The population comprised high-school students of both sexes attending public schools from Caruaru city [2007 (n = 8,333), 2012 (n = 9,405)]. All surveys used the same procedure to calculate the sample size and sampling process. The following parameters were used to estimate sample size: (a) 95% confidence interval; (b) maximum tolerable error of 5%; (c) sampling effect of 1.5. (d) The prevalence was estimated at 50% since multiple outcomes were considered. Additionally, due to possible losses and refusals, 20% of calculated sample was added. A two-stage cluster sampling procedure was performed.

In the first stage, the schools were considered as the sample unit and a random selection using following stratification criteria were considered: (a) school and student density in each micro region of the municipality; and (b) school size (small: less than 200 students, average: 200 to 499 students, and large: 500 students or more). In the second stage, the proportion of students enrolled per class was considered, respecting the

proportion of students enrolled in schools and regions. The final sample size was composed by 600 and 711 students in 2007 and 2012, respectively.

Data from both cross-sectional studies were collected in October (2007 and 2012) in the randomly selected classes and schools. Information on the health behaviors and health status were obtained through an interview using the adapted "*Risk Behaviors used in Adolescents in Santa Catarina State* questionnaire *(COMCAP)"*, previously validated (ICC = 0.62 to 0.98) from students of the public Brazilian school system[20]. Additionally, students answered questions related to PE, such as the school´s offer of PE classes (yes, in the curricular program; yes, as an after-school program; No). Moreover, an open question was included: Give three contents that you most like and three that you dislike in PE classes.

The study variables were divided into demographic and socioeconomic (sex [boy; girl], age [in years], civil status [single; married; other], living zone [urban; rural], family monthly income [≤ R$ 1,000.0; > R$ 1,000.0]), school-related (grade [1st; 2nd; 3rd], shift [morning, night], PE class offer [Yes, same shift; yes, other shift; No], PE class participation [No; Yes], PE school offer [yes, in the curricular program; yes, as an afterschool program; No]) as independent variables, and health related behaviors (physical activity, fruit and vegetable consumption, and TV viewing) as dependent variables.

- **Physical activity** – The habitual physical activity was computed in minutes/week by multiplying both following questions: During a typical or usual week, on how many days do you practice moderate to vigorous physical activity? (0 – 7 days) and "In the days that you practice moderate to vigorous physical activity, how long time you spend?". Youth were classified as physically activity if they reported ≥300 minutes/week of moderate to vigorous physical activity[4].
- **Television time**: It was computed in minutes/week using both following questions: "During weekdays, how much time do you spend per day watching television? and "During weekend days, how much time do you spend per day watching television? The following equation was applied to determine the time using television during the week. [(Time in minutes during weekdays * 5) + (Time in minutes during weekend days * 5/7)]. Adolescents who reported less than 3 hours were classified as non-exposure[4].
- **Fruit consumption:** In a typical week, on how many days do you eat fruit at least once a day? Adolescents who reported 5 times/week or more were classified as adequate eating habits[4].
- **Vegetable consumption:** In a typical week, on how many days do you eat vegetables at least once a day? Adolescents who reported 5 times/week or more were classified as adequate eating habits[4].
- **Health perception:** In general, how do you consider your health? The answers were on a Likert scale, who reported "excellent", or "good" were classified as a positive perception, and who reported "regular" or "bad" were classified as negative perception[4].

Data were tabulated using EpiData program (version 3.1), through duplicate file comparison aiming to identify and to correct possible typing errors based on original values of variables. Statistical analysis procedures were performed using SPSS software (version 18) for Windows. Descriptive and inferential analysis were made by using frequency distribution. Differences between categorical variables were assessed through the chi-square tests. Binary logistic regression was conducted to analyze possible associations between the dependent variable "PE participation" and independent variables related to health behaviors (physical activity, TV viewing, fruit consumption, vegetable consumption and health status perception), stratified by sex. Analysis was adjusted for age, marital status, living zone, family income and school grade. Significance differences or relations were accepted at $p<0.05$.

Results

As shown on Table 1, the sample was composed of 715 students who properly completed the questionnaire, which 56.8% and 61.3% were girls aged 15-17 in 2007 and 2012, respectively. Both samples had higher percentual of students who belonged to low-income families.

Regarding frequency, Figure 1A and 1B show that in 2007 most of the boys and girls did not have any PE class/week, estimated in 72.4% and 69.0%, respectively. When compared to 2012, there was a significant decrease. Otherwise, the proportion of boys and girls who had only one PE class/week in 2012 was highly increased compared to 2007. Conversely, in 2012 there was also a decrease on the proportion of boys and girls who had at least 2 PE classes/week (4.3% and 2.3%, respectively). The percentage of students (boys and girls), who had 3 PE classes was very small either in 2007 or 2012.

Table 2 shows a similar behavior of students both in 2007 and 2012, however there were differences between boys and girls. The most liked activity reported by the boys in both years was soccer. Conversely, it was also the activity girls least liked. Volleyball was the activity girls most liked differently from boys. Although, leading to a significant change in 2012, theory class took an important increase as one of the activities they least liked in PE class.

Overall health behavior is presented in figure 2. For boys, physical activity was similarly estimated in 2007 and 2012. Otherwise, for girls, in 2012 it decreased from 32.5% to 21.9%. Meeting TV viewing recommendation was estimated in 20.6% in 2012 for boys, which suffered a decrease from 2007. Meanwhile for girls it sustained 30.0% in 2012. Surprisingly, most of the students had a positive health status perception, obtaining 89.6% (2007) and 91.0% (2012) for boys and 84.5% (2007) and 73.8% (2012) for girls. This last one presented a concerning decrease over the years. Overall, girls had decreases on every health-related outcome investigated. The same happened to boys, except for physical activity and health status.

Table 3 shows the associations of overall health behavior and PE class for boys and girls, respectively. Having ≥ 1 PE classes was not associated with amount of physical activity either for boys or girls in 2007. TV viewing, as presenting < 3 hours/day, was not significantly associated with having ≥ 1 PE classes, for both sexes.

Reporting fruit consumption for at least 5 days/week was also not associated with having ≥ 1 PE classes in 2007 for both sexes. Similarly, vegetable consumption was not associated with having ≥ 1 PE classes for both sexes. Overall, positive perceived health status was not significantly associated with ≥ 1 PE classes.

In 2012 all outcomes related to health behavior

Table 1 – Students' demographic and socioeconomic characteristics.

Variables	Boys			Girls		
	2007	2012		2007	2012	
	% (n)	% (n)	p	% (n)	% (n)	p
Age						
15 - 17	52.4 (118)	54.1 (152)	0.72	56.8 (213)	61.3 (266)	0.19
18 - 20	47.6 (107)	45.9 (129)		43.2 (162)	38.7 (168)	
Civil status						
Single	97.3 (219)	95.0 (267)	0.24	94.1 (353)	95.4 (414)	0.32
Married	0.4 (1)	2.1 (6)		3.2 (12)	1.6 (7)	
Other	2.2 (5)	2.8 (8)		3.0 (13)	2.7 (10)	
Living zone						
Rural	14.7 (33)	7.1 (20)	0.01	6.9 (26)	9.7 (42)	0.16
Urban	85.3 (142)	92.9 (261)		93.1 (349)	90.3 (392)	
Family monthly income						
≤ R$ 1,000.0	69.8 (157)	62.3 (175)	0.07	75.5 (283)	69.1 (300)	0.04
> R$ 1,000.0	30.2 (68)	37.7 (106)		24.5 (92)	30.9 (134)	
School grade						
1st	44.0 (99)	48.8 (137)		48.0 (180)	41.7 (181)	
2nd	32.9 (74)	34.2 (96)	0.22	32.8 (123)	30.4 (132)	0.01
3rd	23.1 (52)	17.1 (48)		19.2 (72)	27.9 (121)	

Figure A. Boys

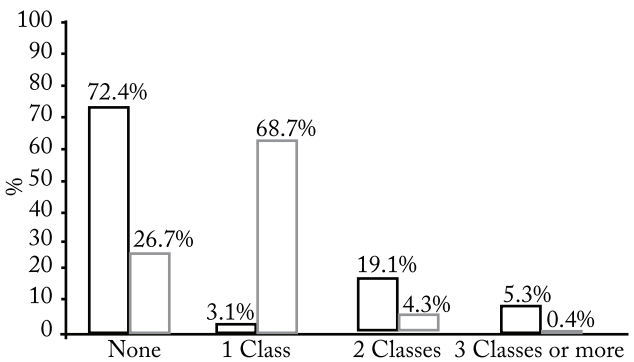

Figure B. Girls

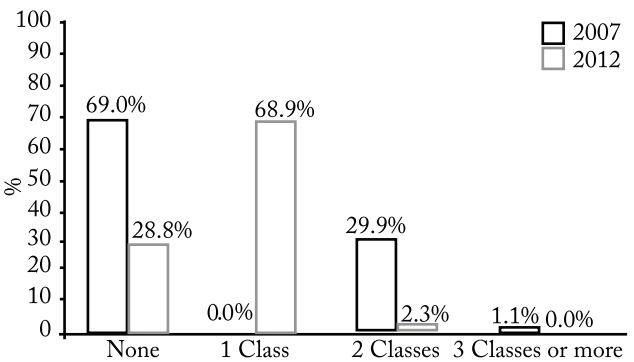

Figure 1 – Students' participation in PE class

remained not significantly associated with having ≥ 1 PE classes. Except for fruit consumption, which became significantly associated for both sexes. This result means that the number of PE class/week is not significantly associated with health-related outcomes, except for fruit consumption.

Discussion

The present study investigated the impact of educational policy on the offer of physical education classes and

Table 2 – Activities that students most and least liked during PE class

Activities	2007						2012					
	Most liked			Least liked			Most liked			Least liked		
	All	Boys	Girls	All	Boys	Girls	All	Boys	Girls	All	Boys	Girls
	(%)			(%)			(%)			(%)		
Soccer	37.2[a]	58.5	22.0	52.4[a]	21.5	78.0	44.1[a]	61.8	20.0	41.2[a]	32.4	47.6
Basketball	5.2	6.3	4.7	13.4[a]	19.5	8.0	3.8	4.3	3.0	2.1	2.9	1.1
Handball	8.1	8.3	8.0	6.1	7.5	5.0	2.2	2.4	1.9	0.8	0.7	0.9
Volleyball	27.3[a]	9.0	41.0	16.5[a]	35.4	1.5	22.1[a]	9.4	43.8	4.0[a]	10.4	0.8
General exercise	22.2[a]	17.9	24.3	11.6[a]	16.1	7.5	21.2[a]	18.4	24.8	3.2	3.8	2.7
Theory class	0.0	0.0	0.0	0.0	0.0	0.0	6.6	6.8	6.5	48.7	50.1	46.9

a = p < 0.05 between boys and girls

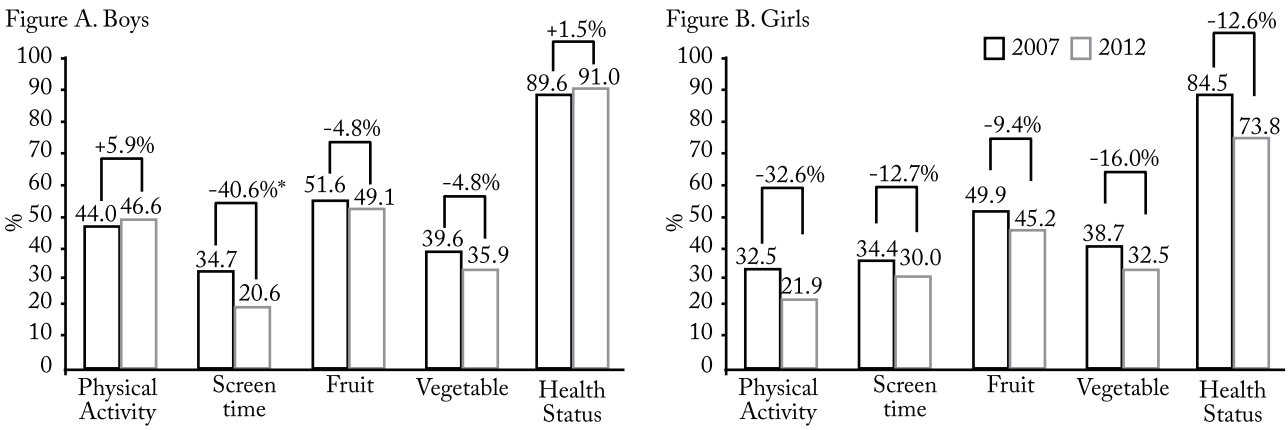

Figure 2 – Percentual of boys and girls who meet recommendations (physical activity, fruit and vegetable consumption, TV viewing and health status) per year

Table 3 – Association of health-related behaviors with PE class among boys and girls

Health behavior	PE class	Boys				Girls			
		2007		2012		2007		2012	
		OR (95%CI)	p	OR (95%CI)	p	OR (95%CI)	p	OR (95%CI)	p
Physical activity									
<300 min/week	0 (Ref)	1	0.23	1	0.72	1	0.92	1	0.80
≥300 min/week	≥ 1	1.47 (0.78 – 2.76)		0.90 (0.51 – 1.58)		1.02 (0.61 – 1.72)		1.06 (0.63 – 1.80)	
Tv viewing									
≥3 hours/day	0 (Ref)	1	0.23	1	0.47	1	0.96	1	0.84
<3 hours/day	≥ 1	1.50 (0.77 – 2.94)		1.27 (0.66 – 2.44)		0.98 (0.58 – 1.66)		1.04 (0.65 – 1.67)	
Fruit consumption									
<5 days/week	0 (Ref)	1	0.80	1	0.04	1	0.31	1	0.04
≥5 days/week	≥ 1	0.92 (0.49 – 1.72)		1.55 (1.01 – 2.70)		1.28 (0.79 – 2.09)		1.48 (1.02 – 2.10)	
Vegetable									
<5 days/week	0 (Ref)	1	0.70	1	0.71	1	0.70	1	0.76
≥5 days/week	≥ 1	1.04 (0.63 – 1.10)		1.07 (0.69 – 1.68)		1.05 (0.63 – 1.70)		1.07 (0.67 – 1.70)	
Perceived health status									
Negative	0 (Ref)	1	0.38	1	0.43	1	0.85	1	0.90
Positive	≥ 1	1.18 (0.64 – 1.98)		1.22 (0.74 – 2.02)		1.06 (0.48 – 1.98)		0.94 (0.36 – 2.46)	

Adjusted analysis for age, marital status, living zone, family income and school grade.

high school students' participation. As the main result, we found that, after the implementation of state educational policies, the frequency of students in PE classes was improved. For girls, health-related behaviors decreased after the policy implementation. On the other hand, boys increased the physical activity levels and health status perception at the same time. Participation in PE class was not associated to health-related behaviors among students, except for fruit consumption. Finally, comparing 2007 to 2012 the number of students who had none PE class was decreased, whereas the number of students who had at least one PE class was increased.

Although this improvement on adherence to PE class may be extremely positive, it remains not ideal. Variability has been noticed in the offer of PE classes among Brazilian schools[15,21]. That could be explained by the fact of, historically in Brazil, PE classes used to occur in a different shift where the students were enrolled. It means that students enrolled in the morning shift, should go home and then go back to school in the afternoon for PE class. Similarly, the students enrolled in the afternoon shift should go to school in the morning only for PE. This practice was strongly characterized as a negative factor, since for a majority, who lived far from school, the fact of returning for a separated class would be discouraging[14]. On the other hand, when PE class is offered in regular school time, it increases the frequency, eliminating characteristics of an optional extracurricular activity that might be associated to PE class[22].

Another associated factor to participation in PE is the increase on physical activity levels[11]. Indeed, regular PE class should provide several benefits for youth's health and the quality of those classes should be endorsed together along with the offer of classes. In Pernambuco state, it is guaranteed one class/week which is equivalent to 50 minutes[17] but it has also been reported[23] that 26.7% of the duration time of PE classes are dispended on physical activity of moderate intensity and only 5.1% of vigorous intensity. On the other hand, sedentary activities and light intensity physical activity take over 22.6% and 44.7%, respectively, representing more than half of the regular PE class duration time. In other words, it is possible to assume that offer of PE is important but the quality of PE plays a large role on the participation.

Results of the present study showed that participation in at least one PE class was largely increased. However, after implementation of state educational policy in 2011[17], the percentual of participation of students in two or three classes was barely existent. This may indicate that just implementing state educational policies for PE class might not be enough to encourage changes on health behaviors of youth, if also the number of classes that should be offered by the schools is not taking into account. It is possible that only assuring for students to have PE class would not be significant for association of PE class and health-related behaviors of students. Nonetheless, the number of classes may have an impact on that. For that matter, there are policies for PE class, not only in Brazil but also among other countries, that establish the minimum number of classes/week[24].

International recommendations indicate that schools must provide at least 150 and 225 minutes of PE, in elementary and high school, respectively[25]. For instance, a specific legislation for PE was created in the United States and, since 2007, the number of laws created for physical activity during PE classes has also increased. PE classes has also increased. Furthermore, in the state of California schools must provide at least 200 minutes of PE classes each 10 day[27].

When it concerns to the quality of PE classes and benefits it may provide, it is indicated in the UNESCO report[7], based on scientific evidences concerning the association of PE with health outcomes, that those policies should be implemented to guarantee the offer of PE. In order to analyze this association, it was also an objective of this study to investigate whether health-related behaviors could be moderated by PE participation.

It came to light that health-related behaviors were not associated with the offer of at least one PE class in this research, except fruit consumption. Regarding to fruit consumption, even though it was found positive association with PE, also found in previous study[4], it was observed that the proportion of students meeting the recommendations in 2007 was approximately 51.6% (boys) and 49.9% (girls), which for both sexes it slightly decrease in 2012 after the policy implementation[17]. In accordance to that, an ecologic study[28] reported that among low-middle-income-countries (of all continents), adolescents commonly do not meet the recommendations for fruit consumption. This outcome could be related to many factors, such as environmental, cultural or socioeconomic factors.

It was also found in the present study that, at least for boys, the level of physical activity had a small increase after policy implementation[17]. In spite of that, physical

activity level decreased for girls. However, regarding physical activity, no positive association with PE was observed. Conversely, other studies indicated different outcomes. They have found that students who had two[4] or three[29] PE classes/week, met physical activity level recommendations. Furthermore, the negative association of PE with physical activity might occur in accordance to intrinsic characteristics of quality of PE class such as intensity and duration of physical efforts in PE[23]. Which means that less activities with higher energy expenditure in PE class lead to reduction of its associations.

It is also possible to attribute the finding of negative association of PE class with physical activity to the fact that one class/week barely counts for 50 minutes of physical activity recommendations[23], diminishing the impact of increase of PE class offer. It may be emphasized by the type of activity proposed in class. As analyzed in the results, there was an increase in theory class after policy implementation, which was the activity students least liked. In fact, this kind of activity is characterized by low energy expenditure. In other words, even though students had offer of at least one PE class, it was probably insufficient to contribute to the overall amount of physical activity.

In addition, it is possible that students did not feel engaged or motivated to participate regarding the activity proposed due to their dislike. Interestingly, before implementation soccer was both the activity they most and least liked. The same occurred after policy implementation[17], when the activity they most liked was soccer and at the same time it was the second activity they least liked (41.2%). In particular, girls showed greater dislike concerning soccer in both years. According to an observational study[13] the content or activities proposed in class are the top of the list of characteristics they least appreciate in PE class and girls (74.9%) show higher dislike concerning PE activities than boys (66.2%).

Also, one of the reasons for them to do not participate would be the repetitive activities[13], it might accumulate natural avoidance over the years. For that matter, some studies from Brazil[13,30] have indicated that students are more likely to be engaged in PE classes when they participate by choosing the activity of the class, also students who dislike the type of activities are more likely to do not participate in the classes. On the other hand, stronger associations are still needed to reinforce this evidence. It was also observed that boys showed greater dislike concerning volleyball and and this should be examined in future studies, since it could be caused by cultural influence.

This study presents strengths regarding the sampling methods, which used parameters with higher chances of accuracy in order to calculate the sample size of the main variable. In spite of that, some limitations need to be highlighted. Although the use of directly measured data is more indicated for this type of research, the behaviors measured in this study were obtained by self-reported questionnaires. Moreover, despite the representative sample, the generalization of data must be done cautiously, as this study only reached high school students from public schools from one municipality of Pernambuco state. Additionally, students' occupation was not analyzed in this study whereas PE class is not obligatory for students with particular issues and it might also impact on students' participation.

Thus, the results indicated that the offer of PE class in high school and participation of students in at least one PE class were improved and substantially higher after the policy implementation in 2011[17]. However, the offer of PE was not associated with students' health-related behaviors, except for fruit consumption in that period of time. It seems that implementation of new policies might cause positive impact on students' behaviors although not sufficiently to cause changes on overall students' health behaviors. It is convenient to suggest further researches intending to consider other behaviors related to the health of this population and the use of different approaches which could allow a better assessment of cause-and-effect association of variables.

Conflicts of interest

The authors declare no conflict of interest.

Contribution of authors

Silva LB, participated in the conception and writing of the manuscript and interpretation of data. Tenório MCM, participated in the critical analysis and review of the manuscript. Martins CML, participated in the critical analysis and review of the manuscript. Silva CRM, participated in the writing and critical analysis of the manuscript and interpretation of data. Tassitano RM, participated in all stages of construction of the manuscript.

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