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Effects of an Exercise and Nutritional Intervention on Circulating Biomarkers and Metabolomic Profiling During Adjuvant Treatment for Localized Breast Cancer: Results From the PASAPAS Feasibility Randomized Controlled Trial

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Abstract

Purpose: Exercise has been shown to improve physical and psychological conditions during cancer therapy, but mechanisms remain poorly understood. The purpose of the present study was to report the results of cancer-related biomarkers and metabolomics outcomes from the PASAPAS feasibility study. **Methods:** In the PASAPAS randomized controlled trial, 61 women beginning adjuvant chemotherapy for localized breast cancer were randomized in a 6-month program of weekly aerobic exercises associated with nutritional counseling versus usual care with nutritional counseling. In the present analysis of 58 women for whom blood samples were available, first, circulating levels of biomarkers (ie, insulin, insulin-like growth factor 1, estradiol, adiponectin, leptin, interleukin-6, and tumor necrosis factor α) were measured at baseline and 6-month follow-up. Changes in biomarkers were compared between exercisers (n = 40) and controls (n = 18) using mixed-effect models. Second, serum metabolites were studied using an untargeted ^{1}H nuclear magnetic resonance spectroscopy, and orthogonal partial least squares analyses were performed to discriminate exercisers and controls at baseline and at 6 months. **Results:** Over the 6-month intervention, no statistically significant differences were observed between exercisers and controls regarding changes in biomarkers and metabolomic profiles. **Conclusion:** The present analysis of the PASAPAS feasibility trial did not reveal any improvement in circulating biomarkers nor identified metabolic signatures in exercisers versus controls during adjuvant breast cancer treatment. Larger studies preferably in women with poor physical activity level to avoid ceiling effect, testing different doses and types of exercise on additional biological pathways, could allow to clarify the mechanisms mediating beneficial effects of physical exercise during cancer treatment. **Trial registration:** ClinicalTrials.gov Identifier: NCT01331772. Registered 8 April 2011, https://clinicaltrials.gov/ct2/show/NCT01331772?term=pasapas&rank=1

Keywords

breast cancer, exercise, chemotherapy, biomarkers, metabolomics

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Introduction

Physical exercise during breast cancer treatment has shown positive effects on physical and psychological outcomes in numerous meta-analyses.[1,2] Observational studies have also suggested that physical activity (PA) after cancer diagnosis

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may improve prognosis among breast cancer survivors.[3] The mechanisms linking PA to breast cancer outcomes are insufficiently understood, but could involve changes in biomarkers that have been associated with breast cancer progression.[4] Several randomized trials have assessed the effect of exercise on insulin, insulin-like growth factors (IGFs), adipokines and inflammation in breast cancer patients after treatment completion.[5] Few studies, however, have examined the effects of exercise on cancer-related biomarkers during cancer treatment. To our knowledge, only 1 study evaluated the effect of exercise on inflammatory biomarkers during adjuvant breast cancer chemotherapy, showing no effect of exercise on interleukin-6 (IL6), interleukin-1-receptor antagonist (IL1ra), and the IL6/IL1ra ratio.[6]

A growing number of metabolomic studies carried out on diverse types of biological samples, more particularly on biological fluids (ie, serum and urine), aim at highlighting biomarkers to distinguish early breast cancer and relapses[7,8] or subclasses linked to treatment response.[9,10] Although some metabolomic studies have investigated the effects of PA,[11–13] to our knowledge, none has studied its effects in the cancer setting.

The randomized, controlled, single-center, open-label PASAPAS trial was designed to evaluate the feasibility of implementing an individualized aerobic exercise program and nutritional counseling during adjuvant treatment of localized invasive breast cancer.[14] The primary outcomes of the PASAPAS trial were compliance and adherence. The overall median adherence rate to the 6-month program (supervised and non-supervised sessions) was 85% despite poor compliance to twice-weekly supervised sessions.[15] To apprehend the benefits of initiating exercise from the onset of adjuvant breast cancer treatment, the purpose of the present study was to report secondary biomarkers and metabolomic outcomes from the PASAPAS feasibility trial. First, we explored the intervention effect on changes in circulating levels of insulin, IGF1, estradiol, adiponectin, leptin, IL6, and tumor necrosis factor α (TNFα) during adjuvant chemotherapy. Second, serum nuclear magnetic resonance (NMR) spectroscopy analyses were performed to examine metabolomic profiles.

Methods

The methodology of the PASAPAS trial has been previously described.[14] The trial (https://www.clinicaltrials.gov, NCT01331772) was approved by the French Sud-Est IV Ethics Committee (No.11-023) and written informed consent was obtained from all participants.

Briefly, 61 women ≥18 years old with a first primary invasive non-metastatic breast cancer scheduled for adjuvant chemotherapy at the comprehensive Léon Bérard Cancer Centre (Lyon, France), with no contraindications to exercise, living within a 60-km perimeter of the cancer center, and able to support travel expenses to attend PA sessions, were randomized in a 2:1 ratio to an exercise intervention or a control group using a computer-generated allocation. Participants in both groups received usual care, including dietary and PA counseling delivered by a registered dietician and a certified adapted PA trainer, according to guidelines for cancer survivors and the recommendations of the French National Health and Nutrition Program (Plan National Nutrition Santé 2, PNNS2)[14,16] as well as print material (entitled "Tips for a healthy diet and practicing physical activity" and labelled by the PNNS2). Personalized dietary care was possible on request for 6 months. The patients in the control group had no restriction for PA. In the intervention group, the 6-month PA program consisted in sessions of 45 to 60 minutes of moderate-to-vigorous intensity (ie, ≥3 metabolic equivalent of task [MET]) aerobic exercises (Nordic walking outdoors, aerobic fitness indoors), performed in small groups, twice a week during chemotherapy (ie, 12-18 weeks in total) and 3 times a week after chemotherapy (including during radiotherapy). Physical activity intensity and duration and type of exercise were individualized under the supervision of a certified adapted PA trainer according to initial PA level.[14,15] In addition to supervised sessions, participants were encouraged to perform non-supervised exercise sessions at home and to record sessions in a PA diary, collected at each follow-up visit. The total duration of participation for all patients was 12 months (ie, for the 6-month intervention program, then a 6-month follow-up). There were 4 evaluation times in the

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study (at baseline, 9 weeks after chemotherapy initiation, at 6 months, and at 12 months)[15]; biomarkers and metabolites analyses have been performed twice from fasting blood samples drawn before the first cycle of chemotherapy (baseline) and at the end of the intervention (6 months after the start of chemotherapy).

Biomarkers Analyses

Serum concentrations of biomarkers were quantified following manufacturer's instructions, using commercial Luminex multiplex kits (insulin, leptin, IL6 and TNFα: HADK2MAG-61K, USA; adiponectin: HADK1MAG-61K, USA; estradiol: STTHMAG-21K, USA). The procedure consisted in incubating 25 µL of serum for 2 hours at room temperature in the presence of magnetic beads labeled with an antibody directed against the parameter to be assayed. After incubation and washing, the beads were incubated for 1 hour with the revelation antibody and then for 30 minutes with the revelation solution (Streptavidin/phycoerythrin). The fluorescence of the beads was read on a luminous automaton (Luminex System, Bio-Rad Laboratories, Germany) and quantified by Luminex software (Bio-Rad laboratories version 4.2). Total IGF1 was quantified using an ELISA kit (DL-IGF1-Hu, China). Duplicate measurements were made for each sample, and the mean of the duplicate measurements was assigned as the sample value.

^{1}H NMR Metabolomic Approach

For NMR analysis, 200 µL of each sample were diluted with 400 µL of buffer solution (0.142 Na_2HPO_4 wt/vol, NaN_3 4% vol/vol, D_2O 10% vol/vol) in a microtube. Samples were centrifuged for 5 minutes at 4°C at 12 000*g*. About 550 µL of supernatant were transferred into 5 mm NMR tubes. NMR spectra collection, processing and annotation were carried out according to previously described protocols.[17] Serum samples were maintained at 4°C before NMR acquisition. One-dimensional ^{1}H nuclear Overhauser effect spectroscopy (NOESY) and Carr-Purcell-Meiboom-Gill (CPMG) NMR spectra were recorded for each serum sample on a Bruker Avance III spectrometer operating at 800.14 MHz ^{1}H NMR frequency. The temperature was regulated at 27°C throughout the experiment. Prior to NMR data acquisition, automatic tuning and matching, frequency locking on D_2O and 1D automatic gradient shimming were performed on each sample. A total of 128 transient free induction decays were collected for each experiment with a spectral width of 20 ppm. For both sequences the relaxation delay was set to 2 s. The NOESY mixing time was set to 10 ms and the CPMG spin-echo delay to 300 µs, with total length of spin-echo train of 80 ms, allowing an efficient attenuation of the lipid NMR signals. The 90° pulse length was automatically calibrated for each sample at around 9 µs. To monitor the good reproducibility of NMR data acquisition over time, additional quality control (QC) samples were prepared according to the same protocol. Serum QC samples were obtained by aliquoting serum from 1 healthy blood donor provided by Établissement Français du Sang, Lyon, France. In practice, 2 QC samples were introduced at the beginning and the end, respectively, of each sample rack corresponding to 1 day of NMR throughput (~40 samples per day). All spectra were referenced to the α-glucose anomeric proton signal ($\delta = 5.23$ ppm) and were manually corrected for baseline using Topspin 3.5 (Bruker GmbH, Rheinstetten, Germany). Additional 2-dimensional NMR spectra (^{1}H-^{13}C HSQC, ^{1}H-^{1}H TOCSY, and ^{1}H J-Resolved) were recorded on a set of representative samples to achieve assignment of the NMR signals observed in the ^{1}H 1-dimensional fingerprints to metabolites. The measured chemical shifts were compared to reference shifts of pure compounds using the HMDB and ChenomX NMR Suite v. 7.1 (Chenomx Inc., Edmonton, Canada) databases. Supplemental Figure S1 shows the mean CPMG spectrum with metabolite assignments. The identification of full spin systems allowed annotation of 50 metabolites illustrated in Supplemental Table S1. After processing and calibration, each 1D NMR spectrum was reduced into bins of 0.001 ppm width over a chemical shift range of 0 to 10 ppm using the AMIX software (Bruker GmbH, Rheinstetten, Germany), giving a total number of 9999 NMR variables. Residual water signal (4.51-5.11 ppm) and polyethylene glycol signal (3.69-3.71 ppm), a contaminant that migrates from plastic vial to the serum, were removed. The spectra were normalized to their total intensity and pareto scaled. All spectra were aligned using the module Icoshift in Matlab (The Mathworks Inc., Natick, MA).

Physical Activity Level

The PA level was assessed by the French version of the validated International Physical Activity Questionnaire (IPAQ, long form).[14,18,19] The IPAQ long form contains 27 items designed to collect detailed information on specific types of activities (walking, moderate-intensity, and vigorous-intensity activities) across 4 domains of PA (occupational, transport, household, and leisure) as well as sedentary behavior over the past 7 days. Total weekly PA was computed by weighting each type of activity by its energy requirement expressed in MET to yield an IPAQ score in MET-minutes/week by multiplying the MET score of an activity by the minutes performed. The IPAQ allowed to classify individuals into categories as follows: high (total PA $\geq$ 3000 MET-minutes/week), moderate (total PA $<$ 3000 and $\geq$600 MET-minutes/week), and low level of PA (total PA $<$ 600 MET-minutes/week).

Statistical Analyses

The sample size of this feasibility trial was fixed to meet its primary objective (ie, the feasibility of implementing an exercise intervention).[14,15] A total sample size of 60 participants (or 40 participants in the exercise group according to the 2:1 allocation ratio) was considered sufficient to assess the primary endpoints of compliance and adherence, and fell within the range of sample sizes recommended in the literature for such trials.[20]

Linear mixed-effect models with subject-specific random intercepts were used to analyze group differences in biomarkers changes from baseline to 6 months. Log-10 transformation was applied to the biomarkers concentrations to account for non-normal distributions. Interaction term between group (exercisers, controls) and time (baseline, 6-month follow-up) was used to examine between-group differences in biomarker changes during the intervention. Estradiol concentrations were modeled using mixed-effect model with binary response based on lower detection limit (ie, 0.1 ng/L). Models were adjusted for age and baseline body mass index. As adjustment for covariates did not modify the results, only unadjusted results are presented. Linear mixed-model analysis was also used to assess between-group differences in change in IPAQ score from baseline to post-intervention.

Analyses of the NMR data were conducted using both unsupervised (principal component analysis [PCA]) and supervised (orthogonal partial least squares discriminant analysis [O-PLS-DA]) multivariate statistical methods to build models for sample classification and extract group-specific metabolic signatures. An initial PCA was used to provide an overview of all observations in the CPMG NMR dataset, check sample homogeneity and identify potential biological and technical outliers. O-PLS-DA was then employed for sample class discrimination. These analyses were conducted to discriminate populations by regressing a supplementary data matrix Y, containing for example information about the intervention and control groups, onto the X NMR data matrix. The goodness-of-fit parameters R^2 and Q^2, which relate to the explained and predicted variance, respectively, evaluated models performance. CV-ANOVA was used to evaluate the significance of each O-PLS-DA model.

All tests were 2-sided and *P*-values $<.05$ were considered statistically significant. Statistical analyses were performed using R (version 3.4.2) and SIMCA-P14 (Umetrics, Umea, Sweden) softwares.

Results

The analyses were performed on 58 out of 61 participants for whom blood samples were available at baseline and 6-month follow-up (40 exercisers, 18 controls).

Baseline characteristics were similar between groups (Table 1). Half of patients were postmenopausal and mean body mass index was 25.1 kg/m^2. The great majority had a diagnosis of invasive ductal carcinoma with Scarff-Bloom-Richardson (SBR) grade II/III. All women were treated with taxane-based chemotherapy and most women (86%) had planned radiotherapy after chemotherapy.

Baseline PA level based on IPAQ score was balanced between groups (Table 1). Women in both groups were physically active at baseline with at least moderate PA level corresponding to recommendations of 30 minutes of moderate-intensity PA on most days. Among patients randomized in the exercise intervention, total PA level increased from an average 2440 (standard deviation, SD: 1152) MET-minutes/week at baseline to 3127 (SD: 1511) MET-minutes/week at 6 months. In controls, the average total PA level was 2920 (SD: 1702) and 2868 (SD: 1333) MET-minutes/week at baseline and 6-month follow-up, respectively. Over the 6 months, no significant difference in change of total PA level was observed between exercisers and controls.

Baseline and 6-month biomarker concentrations are presented in Table 2. No significant between-group differences were observed for any biomarker at baseline. Insulin decreased more in exercisers than in controls but the difference between groups was not statistically significant ($P=.29$). The small increase in IGF1 from baseline to 6 months did not differ between groups ($P=.98$). Inflammatory biomarkers increased slightly in both exercisers and controls, and no significant differences between groups were observed ($P=.63$ and $P=.90$ for between-group differences in changes of IL6 and TNFα, respectively). Leptin and adiponectin decreased in both groups, and the differences comparing exercisers and controls were not statistically significant. Circulating level of estradiol at baseline was over the detection limit (ie, 0.1 ng/L) for 65% of exercisers and 61% of controls compared to 35% and 44% at 6 months in exercisers and controls, respectively ($P=.39$ for between-group difference in changes over time).

^{1}H NMR fingerprints provided a detailed molecular characterization of the patients' sera, including a range of around 50 metabolites from various classes: amino acids, organic acids, sugars, and fatty acids mainly (Supplemental Figure S1, Table S1). Supervised multivariate analysis of the NMR metabolic profiles (O-PLS-DA models) did not reveal any significant difference between exercisers and controls' profiles at baseline and at 6 months (Table 3), characterized by negative Q^2 values. Yet, an evolution of the metabolic profiles for the full cohort of exercisers and control subjects was observed at 6 months as compared to baseline, as shown by O-PLS-DA cross validation ($Q^2=0.135$; CV-ANOVA $P=.003$). Corresponding loadings revealed increased levels of fatty acids and decreased levels of glucose, pyruvate, 3-hydroxybutyrate, glycerol, acetoacetate, histidine, alanine

Table 1. Characteristics of Participants at Baseline (n = 58).

	Exercisers (n = 40)	Controls (n = 18)
Age (years)*	53.8 (10.9)	48.9 (11.1)
BMI (kg/m^2)*	25.2 (5.8)	25.1 (6.1)
Menopausal status**		
Postmenopausal	20 (50.0)	9 (50.0)
Perimenopausal	2 (5.0)	0 (0.0)
Premenopausal	15 (37.5)	7 (38.9)
Unknown	3 (7.5)	2 (11.1)
SBR grade**		
I	4 (10.5)	2 (11.8)
II	18 (47.4)	8 (47.1)
III	16 (42.1)	7 (41.2)
Missing data	2	1
HER2 status**		
Positive	9 (22.5)	4 (22.2)
Negative	31 (77.5)	14 (77.8)
Estrogen receptor status**		
Positive	28 (70.0)	15 (83.3)
Negative	12 (30.0)	3 (16.7)
Progesterone receptor status**		
Positive	25 (62.5)	14 (77.8)
Negative	15 (37.5)	4 (22.2)
Chemotherapy**		
Anthracycline and taxane	18 (45.0)	8 (44.4)
Taxane	22 (55.0)	10 (55.6)
Total physical activity		
IPAQ score (MET-minutes/week)*	2440 (1152)	2920 (1702)
Categories**		
High level [IPAQ score $\geq$3000]	15 (37.5)	7 (38.9)
Moderate level [IPAQ score $\geq$600 and $<$3000]	25 (62.5)	11 (61.1)
Low level [IPAQ score $<$600]	0 (0.0)	0 (0.0)

Abbreviations: BMI, body mass index; SBR, Scarff-Bloom-Richardson; HER2, human epidermal growth factor receptor 2; IPAQ, international physical activity questionnaire; MET, metabolic equivalent of task.
*Mean and standard deviation in parentheses.
**Frequency and percentage in parentheses.

and glutamine in the sera of patients at 6 months as compared to baseline (Figure 1).

Discussion

The present analysis of the PASAPAS feasibility trial did not reveal any improvement in circulating biomarkers nor identify metabolic signatures in exercisers versus controls during adjuvant breast cancer treatment. Keeping in mind the small sample size, the 6-month aerobic exercise program was not able to induce a significant decrease in insulin, IGF1, estradiol, leptin, IL6 and TNFα levels, nor an increase in adiponectin. This is consistent with the absence of significant between-group difference observed for changes in PA level during the study intervention. Given that women in both groups were physically active at baseline, further improvements may have been difficult to achieve, indicating a potential ceiling effect.

This feasibility trial was original since the intervention started at chemotherapy onset and continued for the duration of the adjuvant chemotherapy and radiotherapy treatment. All patients received taxane and nearly half also had anthracycline, and the majority had radiotherapy after chemotherapy. Anthracycline- and taxane-containing adjuvant chemotherapy or radiotherapy have been suggested to increase biomarkers of glucose metabolism (eg, insulin and insulin resistance), IGF1, and biomarkers of inflammation, including IL6 and TNFα.[21–25] In addition, high levels of insulin, IGF1, inflammatory biomarkers, estradiol and leptin, as well as low levels of adiponectin have been associated with increased risk of breast cancer recurrence and mortality.[26–30] While post-treatment

Table 2. Baseline and 6-Month Follow-Up Geometric Means (95% Confidence Intervals) for Concentrations of Biomarkers in Exercisers and Controls (n = 58).

	Baseline	6 months	
	Mean (95%CI)	Mean (95%CI)	*P*-value*
Insulin (ng/L)			
Exercisers	199 (165-239)	148 (114-192)	0.29
Controls	184 (108-312)	171 (116-253)	
Total IGF1 (μg/L)			
Exercisers	48.8 (42.3-56.3)	56.7 (50.4-63.9)**	0.98
Controls	41.3 (30.3-56.4)	47.7 (38.9-58.7)	
Adiponectin (mg/L)			
Exercisers	35.1 (27.3-45.1)	27.8 (21.5-35.9)	0.89
Controls	31.5 (24.4-40.7)	25.3 (19.1-33.5)	
Leptin (μg/L)			
Exercisers	13.0 (9.9-17.1)	11.6 (8.8-15.3)	0.54
Controls	13.2 (9.5-18.5)	12.9 (9.9-16.9)	
IL6 (ng/L)			
Exercisers	1.1 (0.7-1.7)	1.6 (1.2-2.3)	0.63
Controls	1.4 (0.7-2.5)	1.7 (1.0-2.9)	
TNFα (ng/L)			
Exercisers	2.6 (2.1-3.2)	3.1 (2.6-3.6)	0.90
Controls	2.7 (2.0-3.6)	3.1 (2.2-4.5)	

Abbreviations: CI, confidence interval; IGF1, insulin-like growth factor 1; IL6, interleukin-6; TNFα, tumor necrosis factor α.
**P*-value for mixed-effect models comparing changes between groups from baseline to 6-month follow-up.
**One missing value.

Table 3. Goodness-of-Fit Model Parameters for Orthogonal Partial Least Squares Discriminant Analysis (O-PLS-DA) Models Discriminating Exercisers and Controls at Baseline and at 6 Months, and Profiles at 6 Months of Intervention Against Baseline for the Whole Cohort of Patients. Negative Q^2 Values Characterize an Absence of Discrimination Between Exercisers and Controls.

O-PLS-DA model	Samples	Number of samples	Number of components	R^2Y	Q^2	*P*-value
Exercisers vs Controls	Baseline	55	1+1+0	0.483	−0.047	1
	6 months	54	1+1+0	0.371	−0.361	1
Baseline vs 6 months	Exercisers and Controls	113	1+2+0	0.235	0.135	.003

exercise has been shown by meta-analyses to favorably affect insulin, IGF1, and inflammatory biomarkers in breast cancer survivors,[31–33] the effect on biomarkers of exercise during cancer treatment has been little studied to date,[6,25,34] and to the best of our knowledge none has investigated the effect of exercise on insulin and IGF1 levels during chemo- and radiotherapy.

In the present study, insulin levels decreased by respectively 26% and 7% in exercisers and controls, and the metabolomics analyses revealed decreased levels of glucose in the whole cohort of exercisers and controls. Changes in anthropometric measures and body composition have been previously published, showing non-significantly improved body composition (ie, decreased percentage of body fat during the 6-month intervention) in exercisers compared to controls, and stable body weight in the entire cohort of exercisers and controls,[15] although weight gain and deterioration of body composition are frequently reported in breast cancer women during treatment.[21] These results may suggest that aerobic exercise concomitant to adjuvant therapy could mitigate chemotherapy-induced effect on insulin, either through the stimulation of the exercise-mediated energy metabolism and insulin signaling, or through improved body composition.[31,35] However, a non-differential contribution of standard care dietary counseling cannot be excluded. Indeed, dietary recommendations provided as usual care in both groups, and personalized dietary care requested by respectively 3 and 2 women in the exercise and control groups,[15] may have impacted the insulin course alongside the exercise intervention.

The course of estradiol was similar with a more marked decrease in exercisers than in controls, albeit not

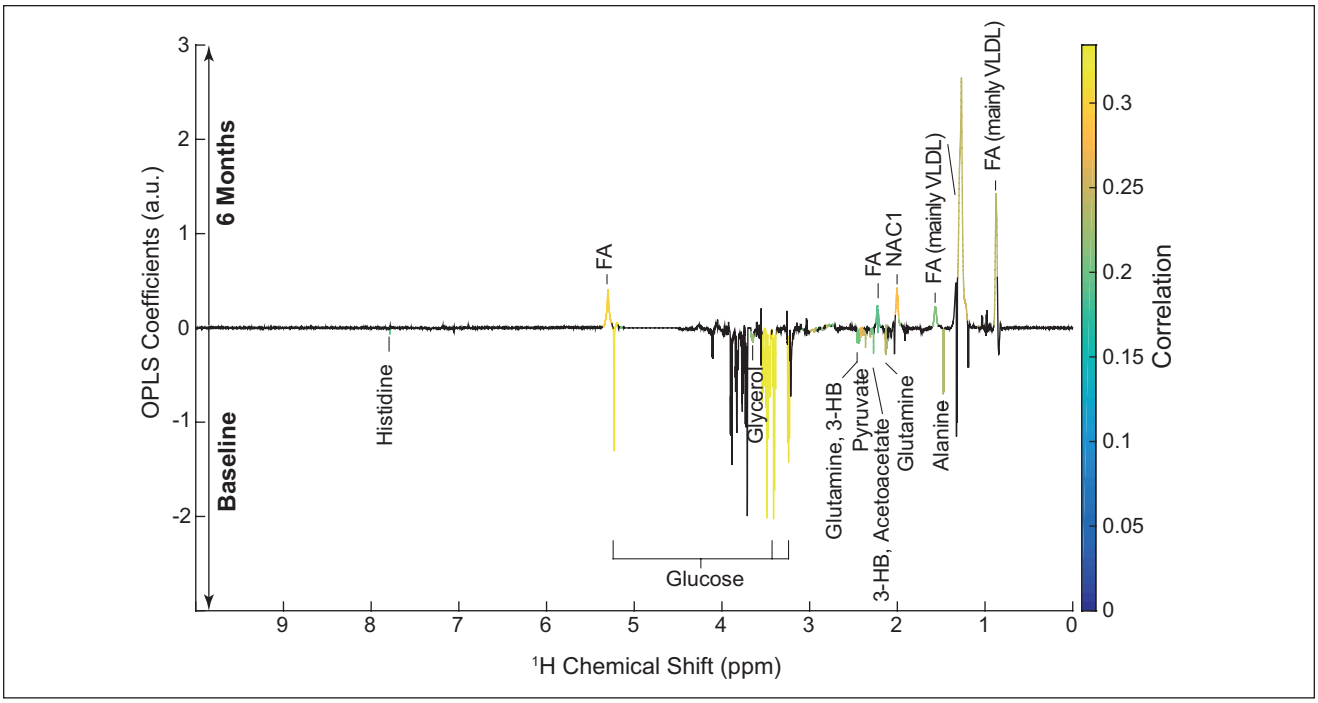


Figure 1. Metabolic signature (O-PLS-DA loadings) from ^{1}H NMR serum profiles discriminating sample at 6 months of intervention with respect to baseline, for the whole cohort of exercisers and control subjects. O-PLS-DA model coefficients are provided in Table 3.
Abbreviations: 3-HB, 3-hydroxybutyrate; FA, Fatty acids; VLDL, Very-low density lipoproteins.

significant. This finding is consistent with the results from a meta-analysis of 6 randomized controlled trials that demonstrated a beneficial effect of exercise combined with reduced caloric intake on breast cancer-related endogenous sex hormones, including total and free estradiol, in healthy, physically inactive postmenopausal women.[36] Moreover, a pilot study assessing the feasibility of an exercise-diet intervention in sedentary, overweight breast cancer patients after treatment, has shown slight, non-significant decrease in serum concentration of total and free estradiol.[37] In the present study, the decrease in estradiol level in the exercise group may be mediated by the decrease in body fat observed at 6 months in this group.[36,37]

Contrarily to the significant decrease in IGF1 observed for post-treatment exercise in breast cancer survivors,[31,32] in this study, an increase in IGF1 level was observed at 6 months in both exercisers and controls. Our results are in line with a previous study evaluating the course of IGF1 during chemotherapy among 151 breast cancer patients receiving anthracycline and taxane in the adjuvant setting that has observed a 29% increase in IGF1 after chemotherapy, while other studies based on smaller samples have shown disparate results.[22] Thus, the capacity of exercise concomitant to chemotherapy to moderate the suggested chemotherapy-induced increase in IGF1 blood levels might be more limited.

Concentrations of the inflammatory cytokines IL6 and TNFα slightly increased in the 2 groups, and no between-group differences were observed. Three studies have assessed the effect of exercise on inflammatory biomarkers during breast cancer treatment.[6,25,34] Van Vulpen et al focused on the effect of exercise on inflammation during adjuvant chemotherapy, by pooling 2 randomized trials including breast cancer patients (n = 130) and evaluating different interventions in terms of type of exercise (ie, resistance or combined resistance and aerobic exercise).[6] Similar to our study, the authors observed an increase in IL6 during chemotherapy in both exercisers and controls and a decrease afterwards. Conversely, in 2 randomized trials evaluating the effects of aerobic and/or resistance exercise during adjuvant radiotherapy, the radiotherapy-induced increase in IL6 was counteracted by exercise.[25,34] We may hypothesize that the chemotherapy-induced increase in IL6 and TNFα might be too important to be effectively offset by exercise, explaining the divergence with the observed benefit of post-treatment exercise on inflammatory cytokines in breast cancer survivors.[6]

The studies evaluating the effects of oncological treatment on adipokine levels are sparse[38] and evidence of the effects of exercise on adipokines remains inconclusive. While the meta-analysis by Kang et al. did not find that exercise can improve adipokine levels (ie, increase

adiponectin and decrease leptin),[31] a randomized controlled trial conducted in sedentary, overweight, or obese breast cancer survivors shortly after treatment completion, reported a significant improvement in adipokine levels in the exercise group who participated in a combined aerobic and resistance training versus the usual care group.[39] In our study, leptin but also adiponectin decreased in both groups with no significant differences between groups. Exercise may have stronger effect on adiponectin in weakly active and overweight women who are susceptible to experience more important changes in body weight and composition associated with exercise.[39,40] We may also hypothesize that the moderate intensity of the intervention was insufficient to affect adiponectin, as more intense exercise might be required to induce an increase in adiponectin levels.[41] Yet, it should be noted that during adjuvant breast cancer treatment, evidence from meta-analyses showed that moderate-intensity physical aerobic exercise is more efficacious than high-intensity exercise in improving fatigue and health-related fitness.[42,43]

The metabolomic analysis of the present study did not identify any difference in serum metabolites between groups. In their review, Daskalaki et al described studies that used a metabolic approach to monitor the effects of exercise mainly in healthy subjects, however, the disparate findings obtained with heterogeneous study designs make it difficult to compare with the results of this study.[13]

Combining aerobic plus resistance training during chemotherapy may be more beneficial on patient-reported outcomes and health-related fitness than aerobic exercise alone.[44] A large cross-sectional study in cancer-free men has shown that engaging in mostly moderate-intensity activity, with preferably combined aerobic and resistance exercises rather than aerobic only, was associated with improved inflammatory biomarkers and insulin response.[45] Yet, the amount of PA and the modalities of practice needed to modify cancer- and treatment related biomarker changes in breast cancer patients undergoing adjuvant therapy have not been investigated.

The strengths of this study include analysis of changes in cancer biomarkers and determination of metabolomic profiles in patients undergoing adjuvant chemo- and radiotherapy. However, several limits should be acknowledged. First, the PASAPAS feasibility study had not been designed to study biomarkers as the primary endpoints. Although the sample size was sufficient to assess the primary objective (ie, feasibility of an exercise intervention),[15,20] the power to examine the secondary objectives including biological effects of exercise may be limited. Second, the relatively short follow-up prevented to study biomarkers evolution after completion of adjuvant treatment. Third, a selection bias toward women highly motivated for PA cannot be ruled out, although there was no recruitment bias with respect to age, BMI, and tumor grade.[15] While a ceiling effect cannot be excluded, a contamination in the control group, which can be expected in exercise oncology trials,[46] may also have further diluted the effect of the intervention. Fourth, the subjective assessment of PA through questionnaires may have induced an over- or under-estimation of actual PA level in each group.[47,48]

Conclusion

The physiological effects of physical activity during adjuvant therapy and the mechanisms mediating beneficial effects in women with breast cancer remain insufficiently explored. Future studies focusing preferably on women with poor physical activity levels should examine biological effects of different doses (frequency, intensity, and duration) of physical activity and types of exercise during cancer treatment.

Declaration of Conflicting Interests

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Ethical Approval

The study was approved by the French Sud-Est IV ethics committee (Comité de Protection des Personnes), the National Security Agency of Medicines and Health Products that applies for biomedical studies (Agence Nationale de Sécurité du Médicament et des produits de santé), and the French National Committee on Informatics and Privacy (Commission Nationale de l'Informatique et des Libertés).

Informed Consent

Informed consent was obtained from all individual participants included in the study.

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Supplemental Material

Supplemental material for this article is available online.

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