Biomedicine & Pharmacotherapy 167 (2023) 115453

Contents lists available at ScienceDirect

Biomedicine & Pharmacotherapy

journal homepage: www.elsevier.com/locate/biopha

Two-month period of 500 mg lecithin-based delivery form of quercetin daily dietary supplementation counterbalances chronic fatigue symptoms: A double-blind placebo controlled clinical trial

Mariangela Rondanelli [a,b], Antonella Riva [c], Giovanna Petrangolini [c], Clara Gasparri [d,*], Simone Perna [e]

[a] IRCCS Mondino Foundation, 27100 Pavia, Italy
[b] Department of Public Health, Experimental and Forensic Medicine, Unit of Human and Clinical Nutrition, University of Pavia, 27100 Pavia, Italy
[c] Development Department, Indena SpA, 20139 Milan, Italy
[d] Endocrinology and Nutrition Unit, Azienda di Servizi alla Persona "Istituto Santa Margherita'', University of Pavia, 27100 Pavia, Italy
[e] Division of Human Nutrition, Department of Food, Environmental and Nutritional Sciences (DeFENS), Università degli Studi di Milano, Milano, Italy

ARTICLE INFO

Keywords:
Chronic fatigue
Dietary supplement
Quercetin phytosome
Fatigue impact scale

ABSTRACT

Background: Chronic fatigue (CF) is a complex phenomenon without clear etiology that may require long-term treatment, but to date, no specific therapy has been identified for it. Some botanicals might be helpful in the management of CF. Among these botanicals, quercetin demonstrates its capacity to modulate multiple biological pathways and acknowledged major properties in CF: antioxidant, anti-inflammatory, immunomodulating, improving exercise endurance, enhancing mitochondrial biogenesis, repairing mitochondrial dysfunction.
Purpose: Given this background, the aim of this study was to evaluate if a 2-month period of daily Quercetin Phytosome™ 500 mg supplementation is of benefit for the relief of CF.
Methods: The primary end point has been the evaluation of fatigue, by Fatigue Impact Scale (FIS-40). The secondary end points have been the assessment of sleep, by Pittsburgh Sleep Quality Index (PSQI), evaluation of muscle performance, by short physical performance battery and by wearable armband-shaped sensor in order to evaluate the number of steps, body composition, by DXA and quality of life by Short-Form 12–Item Health Survey (SF-12).
Results: Seventy-eight subjects (42 F; 36 M) (mean age 56 ± 9) reporting CF symptoms, completed the study (placebo/supplement 38/40). The FIS-40 mean difference changes between groups (supplement minus placebo) was − 10.583 points (CI95% −11.985; −9.182) ($p < 0.001$). Also, statistically significant changes between groups have been recorded in Pittsburgh Sleep Quality Index − 2.040 points (CI95%: −2.770; −1.309), $p < 0.01$), number of steps 1443.152 (CI95%: 1199.556; 1686.749), and SPPB (score) 0.248 (CI95%: 0.105; 0.391) ($p < 0.001$).
Conclusion: The quercetin supplementation counterbalances CF symptoms.

1. Introduction

Until today's fast-paced living conditions, phenomena like unreasonable diet, lack of exercise, irregular work and rest, lack of sleep, mental tension, high psychological pressure, and long-term bad mood are widespread [42,43]. As a result, more and more people are in a sub-health state (a special state between health and illness) and facing "unexplained fatigue".

Fatigue was defined as "an overwhelming, debilitating and persistent feeling of burnout that reduces the person's ability to perform activities of daily living, including working effectively and performing customary family and social duties" [29,36]. If the fatigue is persisting for more

Abbreviations: BMI, body mass index; CF, chronic fatigue; CVs, coefficients of variants; DXA, dual-energy X-ray absorptiometry; FFM, fat free mass; FIS-40, fatigue impact scale; HFD, high fat diet; PSQI, Pittsburgh sleep quality index; QG, quercetin group; QoL, quality of life; SF-12, short form- 12 Item health survey; SPPB, short physical performance battery; TUG, time up and go.

* Corresponding author.
E-mail address: clara.gasparri01@universitadipavia.it (C. Gasparri).

https://doi.org/10.1016/j.biopha.2023.115453
Received 27 June 2023; Received in revised form 26 August 2023; Accepted 5 September 2023
Available online 15 September 2023

than 6 months is called chronic fatigue (CF). It was reported that up to a third of adults experience chronic fatigue for six months or more [37]. To date, there was no definitely effective intervention with coherence and reproducibility in treatment of CF [19].

So, fatigue is a complex and comprehensive physiological phenomenon without clear etiology and may require long-term medication [24]. Current CF therapy consists in the use of over-the-counter and prescription drugs to address specific symptoms, coupled with varying levels of physical and psychological support.

An alternative could be the use of natural supplements with proved effectiveness. In particular, a recent review reported the possible efficacy of some natural derivatives in the treatment of fatigue and chronic fatigue, including also the muscle fatigue deriving from intensive training [24]. Among these botanicals reported by Luo and colleagues, quercetin is supported by an extensive scientific literature in animal and in vitro models, that demonstrated its ability to modulate multiple biological pathways and acknowledged the major properties helpful for relief of CF, i.e, the antioxidant, anti-inflammatory and immunomodulating properties, the improvement of exercise endurance, as well as activities of enhancing mitochondrial biogenesis and repairing mitochondrial dysfunction [11,22,33,38,1,44]. Therefore, quercetin fulfills the need for multifactorial action potential that is suitable for counterbalancing the symptoms of CF.

Quercetin is a flavonoid belonging to the flavonol subfamily and it is one of the most studied polyphenols because of its numerous biological activities. It is also the most abundant flavonoid in nature.

Considering improving exercise endurance, numerous studies demonstrated that quercetin has anti-fatigue capacity. The results of a preclinical study showed that dietary supplementation of quercetin significantly prolonged the swimming time to exhaustion and improved anti-fatigue capacity; these findings suggest that quercetin can improve fatigue resistance by reducing muscle damage and increasing fatty acid β-oxidation [8]. Riva and colleagues [31] showed scientific evidence of positive effects due to the high antioxidant capacity of Quercetin Phytosome™: in triathletes, the supplementation of Quercetin Phytosome™ was more effective than placebo in reducing oxidative stress (by measurement plasma free radicals), even if fatigue was not assessed in this study.

. A reduction of both fatigue and tiredness was observed in two trials in early stage Covid-19 patients supplemented for 30 days by 500 mg of Quercetin Phytosome™ twice in a day [9,10].

The aim of the present pilot study was to evaluate if a 2-month period of quercetin supplementation (500 mg Quercetin Phytosome™) is of benefit in the relief of CF symptoms and in the improvement of quality of life.

2. Materials and methods

2.1. Population

A randomized, double-blind, placebo-controlled trial was conducted in subjects who complain CF symptoms. The subjects were recruited from the Dietetic and Metabolic Unit of the "Santa Margherita" Institute, University of Pavia, Italy.

Inclusion criteria were subjects aged 18 years or older, of both genders, provided signed written informed consent and complain symptoms of fatigue from different etiologies, as reported in Table 1. Symptoms accepted for inclusion were recorded using a self-reported fatigue score assessed using the validated Fatigue Impact Scale (FIS-40) that comprised chronic tiredness impacts related to physical, cognitive and psychosocial functions.

Exclusion criteria were: any clinical condition for chronic fatigue (untreated hypothyroidism, sleep apnea, narcolepsy, medication side effects, and iron deficiency anemia and others); previous diagnosis not unequivocally resolved (chronic hepatitis, malignancy); past or current neuropsychiatric disorders (major depressive disorder with psychotic or melancholic features, bipolar disorder, schizophrenia, delusional disorder, dementias, anorexia nervosa, bulimia nervosa); and concomitant participation in all kinds of other clinical trials within 30 days prior to study inclusion; inability to follow the instructions or to complete the supplementation satisfactorily; failure to provide signed informed consent; use of certain drugs/supplements that might influence outcome in the last 90 days or whose withdrawal might be a relevant problem; anticoagulant treatment; pregnancy or breast-feeding; smoking, alcohol intake or substance abuse; obesity (BMI > 35 kg/m2).

Table 1
Baseline demographic characteristics of study participants (n = 78) who completed the final assessment.

Variable	Placebo (n = 38)	QG (n = 40)	Total sample (n = 78)	P-value between Groups at baseline*
Age (years)	55.92 ± 8.67	56.28 ± 10.00	56.10 ± 9.32	0.868
Female/males	20/18	22/18	42/36	
Height (m)	1.68 ± 0.10	1.65 ± 0.07	1.67 ± 0.09	0.210
Weight (kg)	84.90 ± 12.74	81.52 ± 9.40	83.17 ± 11.21	0.185
BMI (kg/m^2)	30.07 ± 2.01	29.76 ± 1.80	29.91 ± 1.90	0.475
Waist circumference (cm)	103.37 ± 10.54	100.80 ± 9.42	102.05 ± 10.00	0.260
VAT (g)	1343.84 ± 723.98	1116.90 ± 581.90	1227.46 ± 660.61	0.130
Fat Mass (g)	35,059.00 ± 9757.69	34,323.60 ± 6524.43	34,681.87 ± 8212.72	0.695
Fat Free Mass (g)	47,091.71 ± 7773.73	44,599.20 ± 6364.53	45,813.50 ± 7150.33	0.125
Fatigue Etiology				
- Long Covid	11	10	21	
- Post traumatic stress	1	3	4	
- Sine causa	26	27	53	

Data are expressed as means ± standard deviation (SD).
QG: Quercetin Phytosome™ supplemented group; BMI: body mass index; VAT: Visceral Adipose Tissue.

The experimental protocol was approved by the Ethics Committee of the University of Pavia (ethical code Number: 0912/01072022) and was registered at ClinicalTrials.gov under registration number: NCT05730660. All the volunteers gave their written informed consent to participate.

2.2. Primary endpoint

The prospectively defined main outcome measure was the mean difference changes in self-reported fatigue scores assessed using the validated Fatigue Impact Scale (FIS-40) from baseline to the final study visit held two months later. The study was powered to detect a 3-point difference between active treatment group and placebo. The FIS-40 comprises 40 items divided into three domains that describe how perceived fatigue impacts upon cognitive (10 items), physical (10 items), and psychosocial functioning (20 items) over the previous four weeks. Each item is scored from 0 (no fatigue) to 4 (severe fatigue). The total FIS-40 score is calculated by adding together responses from the 40 questions (score range 0–160). Higher scores indicate more functional limitations due to severe fatigue [12,15].

2.3. Secondary endpoints

2.3.1. Pittsburgh Sleep Quality Index

Sleep disturbances were assessed through the self-administered 19-item Pittsburgh Sleep Quality Index (PSQI) questionnaire. Scores are obtained on each of seven domains of sleep quality: subjective sleep

quality, sleep latency, sleep duration, habitual sleep efficiency, sleep perturbations, use of sleeping medication, and daytime dysfunction. Each component is scored from 0 to 3 (0 = no sleep problems and 3 = severe sleep problems). The global PSQI score ranges from 0 to 21 points, with scores of ≥ 5 indicating poorer sleep quality [5].

2.4. Body composition

Body composition (FFM, fat mass, and gynoid and android fat distribution) was measured by dual-energy X-ray absorptiometry (DXA) with the use of a Lunar Prodigy DXA (GE Medical Systems). The in vivo coefficients of variants (CVs) were 0.89% and 0.48% for whole body fat (fat mass) and FFM, respectively. Visceral adipose tissue volume was estimated using a constant correction factor (0.94 g/cm^3). The software automatically places a quadrilateral box, which represents the android region, outlined by the iliac crest and with a superior height equivalent to 20% of the distance from the top of the iliac crest to the base of the skull [26].

2.5. Nutritional assessment

Body weight (to the nearest 0.1 kg) and height (to the nearest 0.5 cm) were measured according to standard procedures, and body mass index (BMI) was derived accordingly [13]. A trained dietitian was responsible for the evaluation of calories and protein intake. At inclusion visit a 24 h dietary recall (with the aid of the caregiver) was performed using a food atlas, while at the end of study a calibrated dietetic spring scale was used to weigh all foods served and returned on consecutive days. A computer program (DR3 v3.1.0; Sintesi Informatica Srl, Milano, Italy) was used to estimate the energy and the macronutrient content of consumed food, including nutritional supplementation.

2.6. Quality of life (QoL) assessment

Quality of life was assessed for each participants: by the Short-Form 12–Item Health Survey (SF-12) questionnaire, consisting of a short, generic health-status measure reproducing the physical and the mental summary scores of the SF-36 by addressing eight health domains (physical functioning, role physical, bodily pain, general health, vitality, social functioning, role emotional, and mental health) [39].

2.7. Evaluation of physical performance

It comprised multiple tests. Gait speed was evaluated by the 4 m walking test, asking the patients to walk at their usual pace and taking into account the best time of two attempts. Patients could use an assistive device, if needed. Specifically, the patient was asked to walk: down a hallway through a 1 m zone for acceleration, a central 4 m "testing" zone and a 1 m zone for deceleration (the patient should not start to slow down before the 4 m mark), starting and stopping the timer with the first footfall after the 0 m line and the 4 m line, respectively [2]. Lower body leg strength and endurance were investigated through the chair-stand test (time required to rise five consecutive times from a chair without arm rests) [17]. Composite evaluation of mobility, balance, walking ability, and fall risk was performed using the timed up and go (TUG) test, which assesses the time taken to rise from an arm chair, walk 3 m, turn, walk back, and sit down again [27]. Finally, we considered the Short Physical Performance Battery (SPPB), which consists of three components: gait speed, chair-stand test, TUG, and balance (three different tests assessing ability to stand with the feet together in the side-by-side, semi-tandem, and tandem positions). Accordingly, each component was scored from 0 (*not possible*) to 4 (*best performance*); the scores add up to a total score ranging from 0 to 12 [16].

2.8. Wearable armband-shaped sensor

Total steps were evaluated using a wearable armband-shaped sensor, SenseWear Pro2 Armband, dressed for 72 h, during the 2 weeks before the start of the study and during the last 2 week of the study.

2.9. Dietary supplement

In the supplemented group, named Quercetin Group (QG), 40 subjects received the supplementation of Quercetin Phytosome™ 500 mg/day in 2 divided doses or, whereas the 38 subjects received placebo (placebo group). Quercetin Phytosome™ was formulated as (white) film-coated tablets, each containing 250 mg of Quercetin Phytosome™ and food-grade ingredients. In order to guarantee the fast bioaccessibility of quercetin, thus improving its bioavailability [30], Quercetin Phytosome™ film-coated tablets were characterized by fast disaggregation time (<30 min). Placebo was formulated as (white) film-coated tablets indistinguishable in appearance from Quercetin Phytosome™ tablets.

2.10. Statistical analysis

Baseline data have been presented as the mean values ± standard deviation of the mean (SEMs) unless otherwise indicated. The normal distribution of the variables was checked using the Shapiro Wills test and using Q-Q graphs. Baseline differences in demographic and clinical characteristics between the groups (placebo and supplementation) were examined using independent t tests. The homogeneity of the variances was estimated by using Levene's test. One-factor covariance ANCOVA test was used for continuous variables to determine differences among the two treatments. The same model was also used to compare changes of the variables (supplementation minus placebo) adjusting for age and gender. Correlations between changes pre-post in supplemented group have been estimated by the Pearson's correlation coefficient when the assumptions of normality were met and by the Spearman's correlation coefficient when the assumptions of normality were not met. We performed all analysis on an intention-to-treat basis. A $p < 0.05$ value was considered significant. Statistical Package for the Social Sciences version 28 software was used to perform the statistical analysis (SPSS Inc, Chicago, IL, USA).

3. Results

A total of 78 adults (42 F; 36 M) (mean age 56 years old ± 9) were randomly assigned to a supplementation group and completed the study (38 placebo and 40 QP). The dropout rates did not significantly differ between the two group and placebo group. Briefly, dropouts were 2 in the placebo group, lost to follow-up visit (Fig. 1).

Baseline demographic parameters are reported in Table 1, together with the description of the different etiologies of fatigue symptoms. As for baseline demographic features, also clinical characteristics (Table 2) were similar in both groups.

Table 3 and Fig. 2 show the mean difference changes in the primary and secondary outcomes. The changes differ significantly between the groups (supplement minus placebo effect) for almost all FIS-40 TEST items with chance in total score of − 10.583 (CI95% −11.985; −9.182) ($p < 0.001$). Statistically significant changes in Pittsburgh Sleep Quality Index have been recorded, specifically for Efficiency, Sleeping pills and Total score − 2.040 (CI95%: −2.770; −1.309) ($p < 0.01$). Adjusted between-group difference was significant also for Steps (number) 1443.152 (CI95%: 1199.556; 1686.749), and SPPB (score) 0.248 (CI95%: 0.105; 0.391) ($p< 0.001$).

Considering body composition parameters (visceral adipose tissue, fat free mass, fat mass), no significant differences were observed in the supplemented group vs placebo. However, a reduction of fat mass was statistically significant only intragroup in the supplemented group.

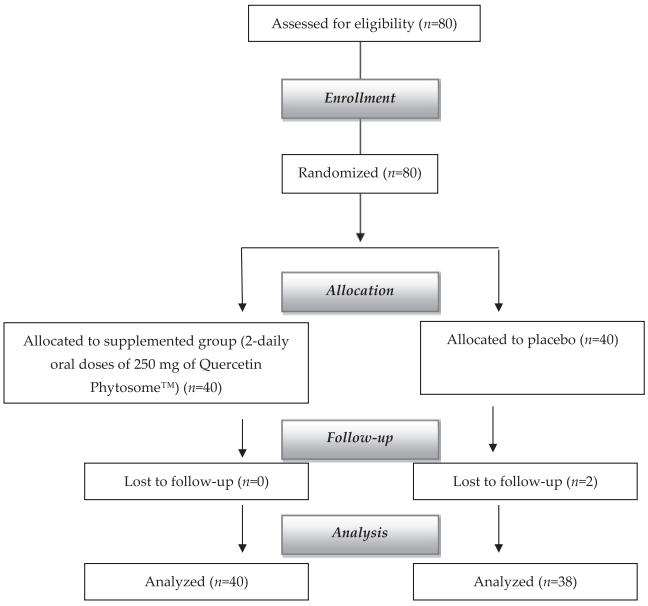

Fig. 1. Flow diagram of the study.

Table 2
Baseline test scores of study participants who completed the final assessment.

Test scores	Placebo (n = 38)	QG (n = 40)	Total sample (n = 78)	P-value between Groups at baseline
FIS-40 TEST				
Cognitive 10	29.95 ± 4.04	30.48 ± 3.33	30.22 ± 3.68	0.530
Physical 10	31.79 ± 3.79	32.7 ± 3.03	32.26 ± 3.43	0.243
Psychosocial 20	63.29 ± 5.36	64.15 ± 4.43	63.73 ± 4.89	0.441
Total	125.03 ± 7.09	127.33 ± 6.51	126.21 ± 6.85	0.140
Pittsburgh Sleep Quality Index				
Sleep quality	1.76 ± 0.71	1.75 ± 0.74	1.76 ± 0.72	0.937
Sleep latency	1.55 ± 0.64	1.58 ± 0.59	1.56 ± 0.62	0.874
Sleep duration	1.61 ± 0.72	1.65 ± 0.70	1.63 ± 0.70	0.781
Efficiency	1.61 ± 0.72	1.63 ± 0.74	1.62 ± 0.72	0.905
Disturbance	1.82 ± 0.69	1.78 ± 0.66	1.79 ± 0.67	0.791
Sleeping pills	1.53 ± 0.69	1.63 ± 0.67	1.58 ± 0.68	0.522
Daytime dysfunction	1.08 ± 0.27	1.15 ± 0.43	1.12 ± 0.36	0.387
Total	12.66 ± 1.92	12.75 ± 1.89	12.71 ± 1.89	0.832
Other outcomes				
Steps (number)	2597.42 ± 722.66	2684.55 ± 861.96	2642.10 ± 793.21	0.631
SPPB	11.84 ± 0.37	11.75 ± 0.44	11.79 ± 0.41	0.320
Physical-SF 12	37.68 ± 11.45	35.34 ± 8.88	36.48 ± 10.22	0.315
Mental-SF 12	44.58 ± 11.88	43.18 ± 9.47	43.86 ± 10.67	0.564

Data are expressed as means ± standard deviation (SD).
QG: Quercetin Phytosome™ supplemented group; SPPB, short physical performance battery; BMI, body mass index; VAT, visceral adipose tissue.

Regarding quality of life, an increase of Physical-SF 12 of + 3.131 (CI95%: 0.992; 5.271) was observed after supplementation.

Table 4 reports the results of Pearson correlation analysis between the mean difference changes (t1-t0) in QG group, which reported a significant change in Table 3.

In QG the overtime decreases of sleeping pills ($r = -0.314$ *; $p < 0.01$) was associated simultaneously to the decrease of total Pittsburgh Sleep Quality Index ($r = -0.384$; $p < 0.01$).

A positive statistically significant association was also recorded between the changes in Pittsburgh Sleep Quality Index and changes in Physical-SF 12 ($r = 0.353$; $p < 0.01$).

4. Discussion

This is the first study in literature that evaluated the use of a lecithin-based delivery form of quercetin (Quercetin Phytosome™) at 500 mg/day for 2 months as of benefit for CF. The main finding of this double-blind placebo-controlled clinical study was a statistically significant relief in fatigue symptoms in supplemented group in respect of placebo administration. All three domains of related to fatigue, cognitive, physical and psychosocial functioning were positively involved.

Comparing our results with the results of previous studies that evaluated the efficacy of dietary supplements of different compositions on chronic fatigue, we find that the effect size of quercetin supplementation is about 10% and is greater than the effect size of 3 g Korean red ginseng supplementation which turns out to be equal to 7% in the Won-Suk study [7] in a middle-aged and moderate level of CF patients, even if that another fatigue assessment scale (The Chalder fatigue severity questionnaire) has been used compared to our study [35].

Also considering the effect of 400 mg/day of dry ethanolic extract of *R. rosea* which was studied in subjects with prolonged or chronic fatigue symptoms, assessed by Multidimensional Fatigue Inventory 20, the effect size is 8% [23], therefore lower than the result of the present study.

There is evidence that quercetin in vitro and in animal studies has an anti-inflammatory and anti-fatigue effectiveness (Chen 2018). This is the first clinical evidence specifically related to fatigue symptoms, only partially suggested by previous pilot studies in Covid-19 subjects [9,10].

The improvement of the cognitive functioning, could be explained by the neuroprotective effect that is exerted by quercetin, as demonstrated in animal models [28]. Moreover, in vitro quercetin demonstrated the increase of gene expression of adenosine A1 receptors in the brain; specifically, quercetin had the highest affinity for this receptor [4]. Quercetin exhibits α7nAChR/Nrf2/HO-1-mediated neuroprotection against STZ-induced mitochondrial toxicity and cognitive impairments in experimental rodents [34].

As secondary endpoint, the sleep quality recorded with PSQI was significantly improved by quercetin supplementation in respect to placebo.

Regarding quality of life, an increase of Physical-SF 12 of + 3.131 (CI95%: 0.992; 5.271) was observed after supplementation, even if, with surprise, the intragroup difference is not statistically significant.

We also noted a discrepancy between the average values of the fatigue test at baseline (which are in line with the values presented in other studies, such as in the study by Castro-Marrero [6] and the values of the SF-12 test. We then considered some explanations. The main one concerns the overestimation of fatigue by patients with chronic fatigue. Already in 1996 it was reported that fatigue can be overestimated in patients with chronic fatigue [14]. This situation is confirmed in numerous subsequent studies [25]. Moreover, we also considered that the shortened version of the SF 12 test is not adequate for assessing quality of life in patients with chronic fatigue, but that the SF 36 test probably needs to be used, although other studies, albeit conducted in different settings, such as obese patients [40,41], have demonstrated that SF-12 and the SF-36 are highly correlated, and both summary measures of the physical and mental components in the SF-12 explained about 90% of the variation in the same summary measures in the SF-36. However, no studies have demonstrated this correlation in patients with CF.

Finally, as a secondary end point, the evaluation of body composition, using DXA, and of muscle function, using SPPB and the step count by wearable arm band, were also included in order to evaluate both the direct efficacy of quercetin on these functions and the efficacy mediated

Table 3
Within group (pre post) and between group (treatment minus placebo effect) mean difference changes from baseline (from day 0 to the end of the supplementation) for all investigated variables during the supplementation period * in bold: value with $p < 0.05$.

Variable	PLACEBO Intra-Group Δ Change (CI 95%)	PLACEBO Intra-Group Δ Change in Percentage (%)	QG Intra-Group Δ Change (CI 95%)	QC Intra-Group Δ Change in Percentage (%)	Effects between Groups (QG minus placebo) Δ Change (CI 95%)	P-value between group
FIS-40 TEST						
Cognitive 10 (score)	0.87 (0.09; 1.65)	+3	-2.95 (−3.71; −2.19)	-10	-3.82 (−4.91; −2.74)	0.001
Physical 10 (score)	0.76 (0.12; 1.39)	+2	-3.12 (−3.74; −2.50)	-9.5	-3.87 (−4.77; −2.89)	0.001
Psychosocial 20 (score)	0.16 (−0.46; 0.78)	+0.2	-2.73 (−3.33; −2.13)	-4.2	-2.89 (−3.75; −2.03)	0.001
Total (score)	1.79 (0.78; 2.79)	+1.4	-8.80 (−9.78; −7.82)	-7	-10.58 (−11.99; −9.18)	0.001
Pittsburgh Sleep Quality Index						
Sleep quality (score)	-0.05 (−0.28; 0.17)	-3	-0.15 (−0.37; 0.07)	-8.5	-0.10 (−0.41; 0.12)	0.537
Sleep latency (score)	0.05 (−0.14; 0.25)	3	-0.18 (−0.37; 0.02)	-11.5	-0.23 (−0.50; 0.05)	0.101
Sleep duration (score)	0.21 (−0.07; 0.50)	+13	-0.30 (−0.58; −0.03)	-18	-0.52 (−0.91; −0.12)	0.011
Efficiency (score)	0.26 (0.01; 0.51)	+16	-0.20 (−0.44; 0.04)	-12	-0.46 (−0.80; −0.11)	0.010
Disturbance (score)	0.05 (−0.20; 0.31)	+3	-0.28 (−0.53; −0.03)	-16	-0.33 (−0.69; 0.03)	0.071
Sleeping pills (score)	0.37 (0.15; 0.59)	+24	-0.08 (−0.29; 0.14)	-5	-0.45 (−0.76; −0.14)	0.005
Daytime dysfunction (score)	0.00 (−0.08; 0.08)	0	-0.08 (−0.16; 0.01)	-7	-0.08 (−019; 0.04)	0.196
Total (score)	-0.81 (−1.34; −0.29)	-6	-2.85 (−3.36; −2.34)	-22	-2.04(−2.77; −1.31)	0.001
Other outcomes						
Steps (number)	-198.28 (−372.69; −23.88)	-8	1244.87 (1074.88; 1414.86)	+47	1443.15 (1199.56; 1686.75)	0.001
SPPB (score)	0.00 (−0.10; 0.10)	0	0.25 (0.15; 0.35)	+2	0.25 (0.11; 0.39)	0.001
Weight (kg)	-0.98 (−1.60; −0.37)	-1	-1.59 (−2.19; −0.99)	-2	-0.61 (−1.47; 0.25)	0.165
BMI (kg/m$^{2)}$	0.38 (−1.46; 0.71)	+1	-1.28 (−2.33; −0.22)	-4	-0.90 (−2.42; 0.62)	0.241
Waist circumference (cm)	-1.20 (−2.08; −0.32)	-1	-1.25 (−2.10; −0.39)	-1	-0.04 (−1.27; 1.19)	0.946
VAT (g)	770.24 (−431.24; 1971.72)	+57	-49.91 (−1220.95; 1121.14)	-4	-820.15 (−2498.30; 858.01)	0.333
Fat Mass (g)	-547.03 (−1805.59; 711.53)	-1.5	-2158.40 (−3385.07; −931.72)	-6	-1611.37 (−3369.25; 146.51)	0.072
Fat Free Mass (g)	-666.70 (−1077.97; −255.43)	-1	-187.34 (−588.19; 213.51)	-0.4	479.36 (−95.07; 1053.80)	0.101
Physical-SF 12	2.60 (0.40; 4.79)	+7	3.13 (0.99; 5.27)	+9	0.535 (−2.532; 3.60)	0.729
Mental-SF 12	-3.19 (−4.90; −1.47)	-7	-2.01 (−3.69; −0.34)	-5	1.17 (−1.23; 3.57)	0.333

QG: Quercetin Phytosome™ supplemented group; SPPB, short physical performance battery; BMI, body mass index; VAT, visceral adipose tissue; CI, confidence interval; 95% Confidence of interval.

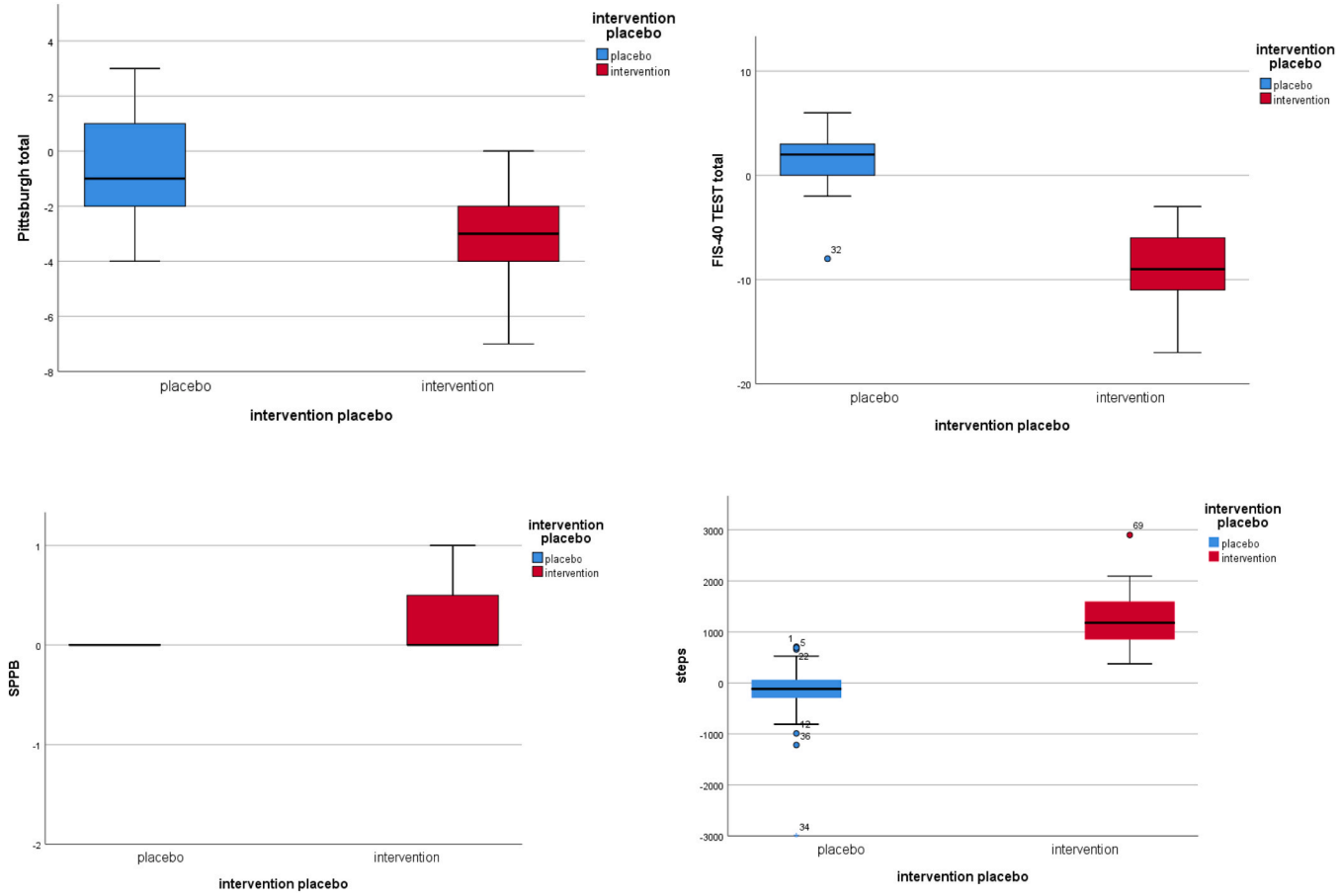

Fig. 2. Within-group mean changes from baseline (from day 0 to the end of the supplementation).

Table 4
Pearson correlations between Δ-changes (t1-t0) in supplementation group based on the statistically significant makers in Table 3.

	Δ sppb	Δ steps	Δ FIS-40	Δ Efficiency	Δ Sleeping pills	Δ Pittsburgh tot
Δ Sppb	1	0.183	0.274	0.000	0.166	0.099
Δ Steps		1	0.242	-0.115	**-0.314***	**-0.384***
Δ FIS-40			1	0.058	-0.041	0.213
Δ Efficiency				1	-0.150	0.245
Δ Sleeping pills					1	0.295
Δ Pittsburgh						1

* Pearson correlations are in the upper triangular matrix with P values (in **bold**: $P < 0.05$)

by the improvement in fatigue.

A statistically significant change has been demonstrated only in the supplemented group for short physical performance battery and for number of steps. We can therefore hypothesize that a reduction in fatigue in the supplemented group resulted in an improvement in daily physical activities.

It is very attractive to see how closely these results are linked to each other; we can speculate that the reduction of fatigue symptoms reported in supplemented group allows the amelioration of quality of life with an increase in daily movement, as demonstrated by the significant increase in the number of steps, with a consequent improvement in physical performance, as demonstrated by the results of the SPPB, resulting also in improved sleep. Even the reduction of adipose tissue, although not statistically significant between groups, but only intragroup in the supplemented group, could be a consequence of this increase in physical activity.

Regarding the reduction of adipose tissue, our study is in agreement with a previous placebo-controlled double-blind parallel-group comparison study by Saito and colleagues. In that trial for subjects with BMI of ≥25 kg/m2, daily intake of tea containing 110 mg quercetin glycoside for 12 weeks significantly decreased visceral fat area [32]. The mechanism underlying suppression of fat accumulation by quercetin was indicated to involve suppression of the expression of peroxisome proliferator-activated receptor γ, which is related to fat accumulation, and sterol regulatory element-binding protein 1 and fatty acid synthase which are related to fatty acid synthesis. Also the increase of the expression of cAMP which is related to lipolysis, could be associated to the reduction of fat accumulation, as reported in animal models [3,20,21]. Moreover, preclinical studies have demonstrated that dietary supplementation with quercetin attenuated high fat diet (HFD)-induced obesity and insulin resistance by increasing energy expenditure through a mechanism involving skeletal muscle mitochondrial adaptations [18].

Despite the significant findings obtained in the present investigation, there are some limitations. Thus, future studies using larger sample sizes and including a wide spectrum of biomarkers are required. In particular, it should be noted that we did not measure antioxidant and inflammatory markers and inflammation could have been present as related to the observed general fatigue. In addition, our sampling was based on

individuals suffering from chronic fatigue from different etiologies and different timelines.

Although these preliminary results are promising, they need further confirmation from large cohort studies.

Funding

The authors thank the whole partnership on the Project MIND FoodS Hub: *Milano Innovation District Food System Hub: Innovative concept for the eco-intensification of agricultural production and for the promotion of dietary patterns for human health and longevity through the creation in MIND of a digital Food System Hub* (P.O.R. 2014-2020_BANDO Call HUB Ricerca e Innovazione _D.G.R. NR 727 del 5/11/2018).

CRediT authorship contribution statement

Mariangela Rondanelli: Conceptualization, Project administration, Writing – original draft, Writing – review & editing. **Antonella Riva:** Conceptualization, Data curation, Investigation, Supervision, Writing – original draft. **Giovanna Petrangolini:** Data curation, Investigation, Supervision, Writing – review & editing. **Clara Gasparri:** Data curation, Writing – review & editing. **Simone Perna:** Formal analysis, Methodology, Writing – review & editing.

Declaration of Competing Interest

AR and GP are employed by Indena SpA. The remaining authors declare no conflict of interest.

Acknowledgments

None.

References

[1] M. Ay, J. Luo, M. Langley, H. Jin, V. Anantharam, A. Kanthasamy, A. G. Kanthasamy, Molecular mechanisms underlying protective effects of quercetin against mitochondrial dysfunction and progressive dopaminergic neurodegeneration in cell culture and MitoPark transgenic mouse models of Parkinson's Disease, J. Neurochem. 141 (5) (2017) 766–782, https://doi.org/10.1111/JNC.14033.

[2] G. Abellan Van Kan, Y. Rolland, S. Andrieu, J. Bauer, O. Beauchet, M. Bonnefoy, M. Cesari, L.M. Donini, S. Gillette-Guyonnet, M. Inzitari, F. Nourhashemi, G. Onder, P. Ritz, A. Salva, M. Visser, B. Vellas, Gait speed at usual pace as a predictor of adverse outcomes in community-dwelling older people an International Academy on Nutrition and Aging (IANA) Task Force, J. Nutr. Health Aging 13 (10) (2009) 881–889, https://doi.org/10.1007/S12603-009-0246-Z.

[3] J. Ahn, H. Lee, S. Kim, J. Park, T. Ha, The anti-obesity effect of quercetin is mediated by the AMPK and MAPK signaling pathways, Biochem. Biophys. Res. Commun. 373 (4) (2008) 545–549, https://doi.org/10.1016/J.BBRC.2008.06.077.

[4] S.P.H. Alexander, Flavonoids as antagonists at A1 adenosine receptors, Phytother. Res. 20 (11) (2006) 1009–1012, https://doi.org/10.1002/PTR.1975.

[5] D.J. Buysse, C.F. Reynolds, T.H. Monk, S.R. Berman, D.J. Kupfer, The Pittsburgh sleep quality index: a new instrument for psychiatric practice and research, Psychiatry Res. 28 (2) (1989) 193–213, https://doi.org/10.1016/0165-1781(89)90047-4.

[6] J. Castro-Marrero, M.C. Zaragozá, I. López-Vílchez, J.L. Galmés, B. Cordobilla, S. Maurel, J.C. Domingo, J. Alegre-Martín, Effect of melatonin plus zinc supplementation on fatigue perception in myalgic encephalomyelitis/chronic fatigue syndrome: a randomized, double-blind, placebo-controlled trial, Antioxidants 10 (7) (2021) 1–17, https://doi.org/10.3390/antiox10071010.

[7] J. Castro-Marrero, M.D. Cordero, M.J. Segundo, N. Sáez-Francàs, N. Calvo, L. Román-Malo, L. Aliste, T. Fernández De Sevilla, J. Alegre, Does oral coenzyme Q10 plus NADH supplementation improve fatigue and biochemical parameters in chronic fatigue syndrome? Antioxid. Redox Signal. 22 (8) (2015) 679–685, https://doi.org/10.1089/ars.2014.6181.

[8] X. Chen, D. Liang, Z. Huang, G. Jia, H. Zhao, G. Liu, Anti-fatigue effect of quercetin on enhancing muscle function and antioxidant capacity, J. Food Biochem. 45 (11) (2021), https://doi.org/10.1111/JFBC.13968.

[9] F. Di Pierro, S. Iqtadar, A. Khan, S. Ullah Mumtaz, M. Masud Chaudhry, A. Bertuccioli, G. Derosa, P. Maffioli, S. Togni, A. Riva, P. Allegrini, S. Khan, Potential clinical benefits of quercetin in the early stage of COVID-19: results of a second, pilot, randomized, controlled and open-label clinical trial, Int. J. Gen. Med. 14 (2021) 2807–2816, https://doi.org/10.2147/IJGM.S318949.

[10] F. Di Pierro, G. Derosa, P. Maffioli, A. Bertuccioli, S. Togni, A. Riva, P. Allegrini, A. Khan, S. Khan, B.A. Khan, N. Altaf, M. Zahid, I.D. Ujjan, R. Nigar, M.I. Khushk, M. Phulpoto, A. Lail, B.R. Devrajani, S. Ahmed, Possible therapeutic effects of adjuvant quercetin supplementation against early-stage COVID-19 infection: a prospective, randomized, controlled, and open-label study, Int. J. Gen. Med. 14 (2021) 2359–2366, https://doi.org/10.2147/IJGM.S318720.

[11] M. Endale, S.C. Park, S. Kim, S.H. Kim, Y. Yang, J.Y. Cho, M.H. Rhee, Quercetin disrupts tyrosine-phosphorylated phosphatidylinositol 3-kinase and myeloid differentiation factor-88 association, and inhibits MAPK/AP-1 and IKK/NF-κB-induced inflammatory mediators production in RAW 264.7 cells, Immunobiology 218 (12) (2013) 1452–1467, https://doi.org/10.1016/J.IMBIO.2013.04.019.

[12] J.D. Fisk, P.G. Ritvo, L. Ross, D.A. Haase, T.J. Marrie, W.F. Schlech, Measuring the functional impact of fatigue: initial validation of the fatigue impact scale, Clin. Infect. Dis. (1994) S79–S83, https://doi.org/10.1093/CLINIDS/18.SUPPLEMENT_1.S79.

[13] A.R. Frisancho, New standards of weight and body composition by frame size and height for assessment of nutritional status of adults and the elderly, Am. J. Clin. Nutr. 40 (4) (1984) 808–819, https://doi.org/10.1093/ajcn/40.4.808.

[14] A.M. Fry, M. Martin, Fatigue in the chronic fatigue syndrome: a cognitive phenomenon? J. Psychosom. Res. 41 (5) (1996) 415–426, https://doi.org/10.1016/S0022-3999(96)00190-0.

[15] G.S. Gorman, J.L. Elson, J. Newman, B. Payne, R. McFarland, J.L. Newton, D. M. Turnbull, Perceived fatigue is highly prevalent and debilitating in patients with mitochondrial disease, Neuromuscul. Disord. 25 (7) (2015) 563–566, https://doi.org/10.1016/j.nmd.2015.03.001.

[16] J.M. Guralnik, E.M. Simonsick, L. Ferrucci, R.J. Glynn, L.F. Berkman, D.G. Blazer, P.A. Scherr, R.B. Wallace, A short physical performance battery assessing lower extremity function: association with self-reported disability and prediction of mortality and nursing home admission, J. Gerontol. 49 (2) (1994) M85–M94.

[17] R. Hardy, R. Cooper, I. Shah, S. Harridge, J. Guralnik, D. Kuh, Is chair rise performance a useful measure of leg power? Aging Clin. Exp. Res. 22 (5–6) (2010) 412–418, https://doi.org/10.1007/BF03324942.

[18] T.M. Henagan, W.T. Cefalu, D.M. Ribnicky, R.C. Noland, K. Dunville, W. W. Campbell, L.K. Stewart, L.A. Forney, T.W. Gettys, J.S. Chang, C.D. Morrison, In vivo effects of dietary quercetin and quercetin-rich red onion extract on skeletal muscle mitochondria, metabolism, and insulin sensitivity, Genes Nutr. 10 (1) (2015), https://doi.org/10.1007/S12263-014-0451-1.

[19] D.Y. Kim, J.S. Lee, S.Y. Park, S.J. Kim, C.G. Son, Systematic review of randomized controlled trials for chronic fatigue syndrome/myalgic encephalomyelitis (CFS/ME), J. Transl. Med. 18 (1) (2020) 1–12, https://doi.org/10.1186/S12967-019-02196-9/FIGURES/2.

[20] M. Kobori, S. Masumoto, Y. Akimoto, H. Oike, Chronic dietary intake of quercetin alleviates hepatic fat accumulation associated with consumption of a Western-style diet in C57/BL6J mice, Mol. Nutr. Food Res. 55 (4) (2011) 530–540, https://doi.org/10.1002/MNFR.201000392.

[21] U.R. Kuppusamy, N.P. Das, Effects of flavonoids on cyclic AMP phosphodiesterase and lipid mobilization in rat adipocytes, Biochem. Pharmacol. 44 (7) (1992) 1307–1315, https://doi.org/10.1016/0006-2952(92)90531-M.

[22] K.M. Lee, M.K. Hwang, D.E. Lee, K.W. Lee, H.J. Lee, Protective effect of quercetin against arsenite-induced COX-2 expression by targeting PI3K in rat liver epithelial cells, J. Agric. Food Chem. 58 (9) (2010) 5815–5820, https://doi.org/10.1021/JF903698S.

[23] Y. Lekomtseva, I. Zhukova, A. Wacker, Rhodiola rosea in subjects with prolonged or chronic fatigue symptoms: results of an open-label clinical trial, Complement. Med. Res. 24 (1) (2017) 46–52, https://doi.org/10.1159/000457918.

[24] C. Luo, X. Xu, X. Wei, W. Feng, H. Huang, H. Liu, R. Xu, J. Lin, L. Han, D. Zhang, Natural medicines for the treatment of fatigue: bioactive components, pharmacology, and mechanisms, Pharmacol. Res. 148 (2019), https://doi.org/10.1016/J.PHRS.2019.104409.

[25] F.A. Metzger, D.R. Denney, Perception of cognitive performance in patients with chronic fatigue syndrome, Ann. Behav. Med. 24 (2) (2002) 106–112, https://doi.org/10.1207/S15324796ABM2402_07.

[26] A. Mohammad, E. De Lucia Rolfe, A. Sleigh, T. Kivisild, K. Behbehani, N. J. Wareham, S. Brage, T. Mohammad, Validity of visceral adiposity estimates from DXA against MRI in Kuwaiti men and women, Nutr. Diabetes 7 (1) (2017), https://doi.org/10.1038/nutd.2016.38.

[27] D. Podsiadlo, S. Richardson, The timed Up Go: a test of basic functional mobility for frail elderly persons, J. Am. Geriatr. Soc. 39 (2) (1991) 142–148.

[28] Y. Qi, L. Guo, Y. Jiang, Y. Shi, H. Sui, L. Zhao, Brain delivery of quercetin-loaded exosomes improved cognitive function in AD mice by inhibiting phosphorylated tau-mediated neurofibrillary tangles, Drug Deliv. 27 (1) (2020) 745–755, https://doi.org/10.1080/10717544.2020.1762262.

[29] W.T. Riley, N. Rothrock, B. Bruce, C. Christodolou, K. Cook, E.A. Hahn, D. Cella, Patient-reported outcomes measurement information system (PROMIS) domain names and definitions revisions: further evaluation of content validity in IRT-derived item banks, Qual. Life Res. Int. J. Qual. Life Asp. Treat. Care Rehabil. 19 (9) (2010) 1311–1321, https://doi.org/10.1007/S11136-010-9694-5.

[30] A. Riva, M. Ronchi, G. Petrangolini, S. Bosisio, P. Allegrini, Improved Oral Absorption of Quercetin from Quercetin Phytosome, a New Delivery System Based on Food Grade Lecithin, Eur. J. Drug Metab. Pharmacokinet 44 (2) (2019) 169–177, https://doi.org/10.1007/s13318-018-0517-3.

[31] A. Riva, J.A. Vitale, G. Belcaro, S. Hu, B. Feragalli, G. Vinciguerra, M. Cacchio, E. Bonanni, L. Giacomelli, R. Eggenhöffner, S. Togni, Quercetin phytosome® in triathlon athletes: a pilot registry study, Minerva Med. 109 (4) (2018) 285–289, https://doi.org/10.23736/S0026-4806.18.05681-1.

[32] K. Saito, T. Tanaka, H. Obata, J. Nakamura, N. Fukui, N. Tonozuka, Body fat reducing effect and safety evaluation of long-term consumption of tea containing quercetin glucosides in obese subjects (in Japanese), Jpn. Pharmacol. Ther. 43 (2015) 181–194.
[33] A. Septembre-Malaterre, A. Boumendjel, A.L.S. Seteyen, C. Boina, P. Gasque, P. Guiraud, J. Sélambarom, Focus on the high therapeutic potentials of quercetin and its derivatives, Phytomed. Plus Int. J. Phytother. Phytopharmacol. 2 (1) (2022), https://doi.org/10.1016/J.PHYPLU.2022.100220.
[34] N.K. Singh, D. Garabadu, Quercetin exhibits α7nAChR/Nrf2/HO-1-mediated neuroprotection against STZ-induced mitochondrial toxicity and cognitive impairments in experimental rodents, Neurotox. Res. 39 (6) (2021) 1859–1879, https://doi.org/10.1007/S12640-021-00410-5.
[35] W.-S. Sung, H.-R. Kang, C.-Y. Jung, S.-S. Park, S.-H. Lee, E.-J. Kim, Efficacy of Korean red ginseng (Panax ginseng) for middle-aged and moderate level of chronic fatigue patients: a randomized, double-blind, placebo-controlled trial, Complement. Ther. Med. 48 (2020), 102246, https://doi.org/10.1016/j.ctim.2019.102246.
[36] V. Suzan, B.B. Kanat, H. Yavuzer, Fatigue and primary sarcopenia in geriatric patients, Rev. Assoc. Med. Bras. 68 (11) (2022) 1565–1570, https://doi.org/10.1590/1806-9282.20220662, 1992.
[37] M. Van'T Leven, G.A. Zielhuis, J.W. Van Der Meer, A.L. Verbeek, G. Bleijenberg, Fatigue and chronic fatigue syndrome-like complaints in the general population, Eur. J. Public Health 20 (3) (2010) 251–257, https://doi.org/10.1093/eurpub/ckp113.
[38] F.T.M.C. Vicentini, T.R.M. Simi, J.O. Del Ciampo, N.O. Wolga, D.L. Pitol, M. M. Iyomasa, M.V.L.B. Bentley, M.J.V. Fonseca, Quercetin in w/o microemulsion: in vitro and in vivo skin penetration and efficacy against UVB-induced skin damages evaluated in vivo, Eur. J. Pharm. Biopharm. 69 (3) (2008) 948–957, https://doi.org/10.1016/J.EJPB.2008.01.012.
[39] J.E. Ware, M. Kosinski, S.D. Keller, A 12-item short-form health survey: construction of scales and preliminary tests of reliability and validity, Med. Care 34 (3) (1996) 220–233, https://doi.org/10.1097/00005650-199603000-00003.
[40] J.E. Ware, M. Kosinski, S.D. Keller, A 12-item short-form health survey: construction of scales and preliminary tests of reliability and validity, Med. Care 34 (3) (1996) 220–233, https://doi.org/10.1097/00005650-199603000-00003.
[41] C.C. Wee, R.B. Davis, M.B. Hamel, Comparing the SF-12 and SF-36 health status questionnaires in patients with and without obesity, Health Qual. Life Outcomes 6 (1) (2008) 11, https://doi.org/10.1186/1477-7525-6-11.
[42] World Health Organization, A Healthy Lifestyle, 2019. https://www.euro.who.int/en/health-topics/disease-prevention/nutrition/a-healthy-lifestyle.
[43] World Health Organization, World Health Day 2019: Key Messages, 2019. https://www.who.int/campaigns/world-health-day/2019/key-messages.
[44] D. Yang, X. Liu, M. Liu, H. Chi, J. Liu, H. Han, Protective effects of quercetin and taraxasterol against H2O2-induced human umbilical vein endothelial cell injury in vitro, Exp. Ther. Med. 10 (4) (2015) 1253–1260, https://doi.org/10.3892/ETM.2015.2713.