



Normative values and reference equation for the six-minute step test to evaluate functional exercise capacity: a multicenter study

Vanessa Salles Albuquerque¹, Simone Dal Corso², Daniel Pereira do Amaral², Túlio Medina Dutra de Oliveira¹, Gerson Fonseca Souza³, Rachel Naara Silva de Souza³, Ana Karolyn Menezes Nogueira³, Pedro Dal Lago⁴, Maria Luísa Rocha Dadalt⁴, Isadora Faraco Correa⁴, Graziella França Bernardelli Cipriano⁵, Fabíola Maria Ferreira Silva⁵, Raquel Rodrigues Britto⁶, Anderson José¹, Carla Malaguti¹

EXPANDED METHOD

Recruitment

The participants were recruited from the five regions of Brazil: north, northeast, south, southeast, and central-west. They were recruited by flyers or social media from the local community and among students and employees of six universities, including their relatives.

Procedures

Team training

Procedures were performed at the university research labs in each location. Previously trained researchers performed all the assessments, and they were knowledgeable about how to perform, contraindicate, and interrupt the tests, as well as having the ability to deal with emergency situations. After the formal acceptance of each eligible research center, a videoconference was held to provide general information. Standardized instructions were provided along with tutorial videos, videoconferences, a material checklist, and text messages about how to conduct the test and collect data.

Anthropometric measurements

For anthropometric measurements, the participants remained standing in the upright position with their arms crossed over their trunk and their feet slightly apart from each other. The measurement of height (cm) and weight (kg) of the volunteer was performed using a stadiometer and a mechanical scale (Welmy, São Paulo, Brazil), respectively. Height was measured with the individual in an orthostatic position after deep inspiration, keeping the feet together and with the bodyweight distributed between them. Body mass was measured with the patient barefoot and wearing light clothing. BMI was determined from height (cm) and body weight (kg) measurements. Abdominal circumference (cm) was assessed using a tape measure around the umbilical scar line. The length of the right lower limb (cm) was measured with the subject barefoot and was determined by the distance between the highest point of the greater trochanter of the femur and the floor. Thigh circumference (midpoint between the greater trochanter and the knee joint line) and the circumference of the largest calf were also measured.⁽¹⁾

Short version of the International Physical Activity Questionnaire

Physical activity (PA) level was determined using the short version of the International Physical Activity Questionnaire (IPAQ).^(2,3) The IPAQ short form asks about three specific types of activity (walking, moderate-intensity activities, and vigorous-intensity activities). It provides separate scores for time (min) and frequency (days) in each type of activity and also includes a sitting question as an additional indicator of sedentary lifestyle. The IPAQ short form identifies the total number of minutes over the last seven days spent on each type of PA. Responses are converted to metabolic equivalent task (MET) according to the IPAQ scoring protocol (8.0 MET for each minute of vigorous activity, 4.0 MET for each minute of moderate activity, and 3.3 MET for each minute of walking). Each type of activity has its MET score calculated, allowing for the following categorization: high level of PA—subjects spending at least 1,500 MET on vigorous activities per week or spending 3,000 MET on moderate plus vigorous PA per week; moderate level of PA (one of the three following criteria)—spending ≥ 3 days on vigorous activity for at least 20 min per day; ≥ 5 days on moderate-intensity activity and/or walking for at least 30 min per day; or ≥ 5 days on any combination of walking, moderate-intensity, or vigorous-intensity activities, achieving at least 600 MET in one week; and low level of PA—under a moderate-activity level of PA.⁽³⁾

Instructions given before the six-minute step test

These are the standardized verbal instructions given before and during the test:

"Mr./Ms. name of the individual:

This is the six-minute step test. The objective of this test is to climb the greatest number of steps you can in six minutes. The more steps you climb, the better your exercise capacity. You need to go up the step and then back down again to the place you started, with your feet on the floor. You can use either of your legs to begin, and you can change to the other whenever you want."

(Demonstrate one step up and down, starting with one leg, and then climb one more starting with the other leg).

"You cannot use your arms to help you climb, but if you feel that you might fall, you can use them to regain

Table S1. Characteristics of the sample, as a whole and by region of the country.^a

Characteristic	Total sample (n = 468)	Region					
		Southeast 1 (n = 163)	Southeast 2 (n = 93)	South (n = 53)	North (n = 52)	Northeast (n = 78)	Central-west (n = 32)
Comorbidity							
Hypertension	47 (10.0)	31 (19.3)	4 (4.3)	0 (0.0)	3 (5.9)	4 (5.1)	0 (0.0)
Diabetes mellitus	6 (3.7)	6 (3.7)	0 (0.0)	5 (9.4)	1 (2.0)	1 (1.3)	0 (0.0)
Smoking	7 (4.3)	7 (4.3)	0 (0.0)	0 (0.0)	1 (2.0)	1 (1.3)	0 (0.0)
Weight, kg	69 ± 13	70.5 ± 13.8	68.8 ± 11.6	69.2 ± 12.4	63.8 ± 10.0 ⁱ	67.6 ± 13.0	68.4 ± 14.6
Height, m	1.66 ± 0.10	1.65 ± 0.10	1.64 ± 0.08	1.68 ± 0.09	1.66 ± 0.10	1.68 ± 0.11	1.65 ± 0.10
BMI classification							
Under weight	11 (2.4)	5 (3.0)	2 (2.2)	0 (0.0)	2 (4.0)	3 (3.8)	2 (6.3)
Normal weight	242 (51.7)	70 (43.5)	45 (48.4)	32 (60.4)	31 (60.8)	46 (59.0)	18 (56.3)
Overweight	215 (45.9)	88 (54.7)	46 (49.5)	21 (39.6)	19 (37.3)	29 (37.2)	12 (37.5)
Age, years	41 ± 17	46.5 ± 16.7*	46.5 ± 19.0*	36.5 ± 17.1	32.8 ± 10.3	35.0 ± 15.3	36.2 ± 13.8
Age range, years							
18-28	135 (28.8)	27 (16.8)	21 (22.6)	21 (39.6)	18 (35.3)	37 (47.4)	11 (34.4)
19-29	110 (23.5)	38 (23.0)	16 (17.2)	14 (26.4)	17 (33.3)	16 (20.5)	10 (31.3)
30-39	65 (13.9)	26 (16.1)	11 (11.8)	3 (5.7)	13 (25.5)	9 (11.5)	3 (9.4)
40-49	65 (13.9)	24 (14.9)	15 (16.1)	9 (17.0)	3 (5.9)	8 (10.3)	6 (18.8)
50-59	60 (12.8)	32 (19.9)	17 (18.3)	3 (5.7)	1 (2.0)	6 (7.7)	2 (6.3)
60-69	33 (7.1)	16 (9.3)	13 (14.0)	3 (5.7)	0 (0.0)	2 (2.6)	0 (0.0)
FVC, % predicted	95 ± 10	91.6 ± 9.8	96.4 ± 11.2	100.0 ± 10.6	97.3 ± 2.5	94.0 ± 10.9 [†]	94.3 ± 9.1 [§]
FEV ₁ , % predicted	98 ± 11	98.1 ± 10.3	96.8 ± 10.0	99.4 ± 11.1	96.5 ± 3.9	101.3 ± 16.3	95.6 ± 7.1
FEV ₁ /FVC	86 ± 6	87.8 ± 6.1	82.3 ± 5.5	84.7 ± 6.1	85.9 ± 3.2	87.6 ± 7.5	85.2 ± 7.9
IPAQ	2.5 ± 1.1	2.6 ± 1.1	2.8 ± 1.0	1.8 ± 0.9 [§]	2.4 ± 0.8	2.5 ± 1.2	2.9 ± 1.2
Physical activity levels							
Very active	58 (12.4)	14 (8.7)	1 (11.0)	19 (35.8)	3 (5.9)	18 (23.1)	3 (9.4)
Active	226 (48.3)	80 (49.7)	49 (52.7)	28 (52.8)	32 (62.7)	27 (34.6)	10 (31.3)
Irregularly active A	74 (15.8)	28 (17.4)	18 (19.4)	1 (1.9)	8 (15.7)	10 (12.8)	9 (28.1)
Irregularly active B	73 (15.6)	25 (15.0)	16 (17.2)	4 (7.5)	7 (13.7)	17 (21.7)	5 (15.6)
Sedentary	37 (7.9)	16 (9.9)	9 (9.7)	1 (1.9)	1 (2.0)	6 (6.4)	5 (15.6)
Six-minute step test							
Final HR, % predicted	83 ± 14	79 ± 14	91 ± 10	82 ± 14	80 ± 13	88 ± 12	76 ± 16
Final HR > 85% predicted ^b	45	41	73*	43	39	64 [†]	41
Steps climbed	179 ± 41	164.0 ± 39.2*	183.8 ± 41.1	208.4 ± 47.5	172.3 ± 36.7	182.4 ± 36.7	195.7 ± 30.4

IPAQ: International Physical Activity Questionnaire (short form); and Max: maximum. ^aValues expressed as n (%) or mean ± SD. ^bNumber of participants with HR > 85% of the predicted value at the end of the test. One-way ANOVA and post-hoc results are indicated with the following symbols: *p < 0.05 vs. Southeast; [†]p < 0.05 vs. North; [‡]p < 0.05 vs. Northeast; [§]p < 0.05 vs. Central-west; and; ^{||}p < 0.05 vs. South.

your balance. You need to stop using your arms as soon as possible."

"Six minutes is a long time to climb stairs, so you will be exercising yourself. You can slow down, stop, and even rest in the chair provided, but you should resume climbing as soon as you can. If your heart beats too fast, or the oxygen in your blood becomes too low, I will ask you to stop for a moment, and I will let you know when you can start again. Even if you stop, the countdown timer will not be stopped. Are you read to start? Start now or whenever you are ready."

The verbal feedback at each minute of the test, as adapted from the statement for the six-minute walk test,⁽⁴⁾ was given as follows: After the first minute, tell the patient the following (in same tones): "You are doing well. You have five minutes to continue." When

the timer shows four minutes remaining, the patient is told the following: "Keep up the good work. You have four minutes to continue." When the timer shows three minutes remaining, tell the patient the following: "You are doing well. You are halfway done." When the timer shows two min remaining, tell the patient the following: "Keep up the good work. You have only two minutes left." When the timer shows only one minute remaining, tell the patient: "You are doing well. You have only one minute to continue." Do not use other words of encouragement (or body language to speed up). If the patient stops climbing during the test and needs a rest, say this: "You can lean against the wall if you would like; then, continue climbing whenever you feel able." Do not stop the timer. If the patient stops before the six minutes are up and refuses to continue (or if you decide that they should

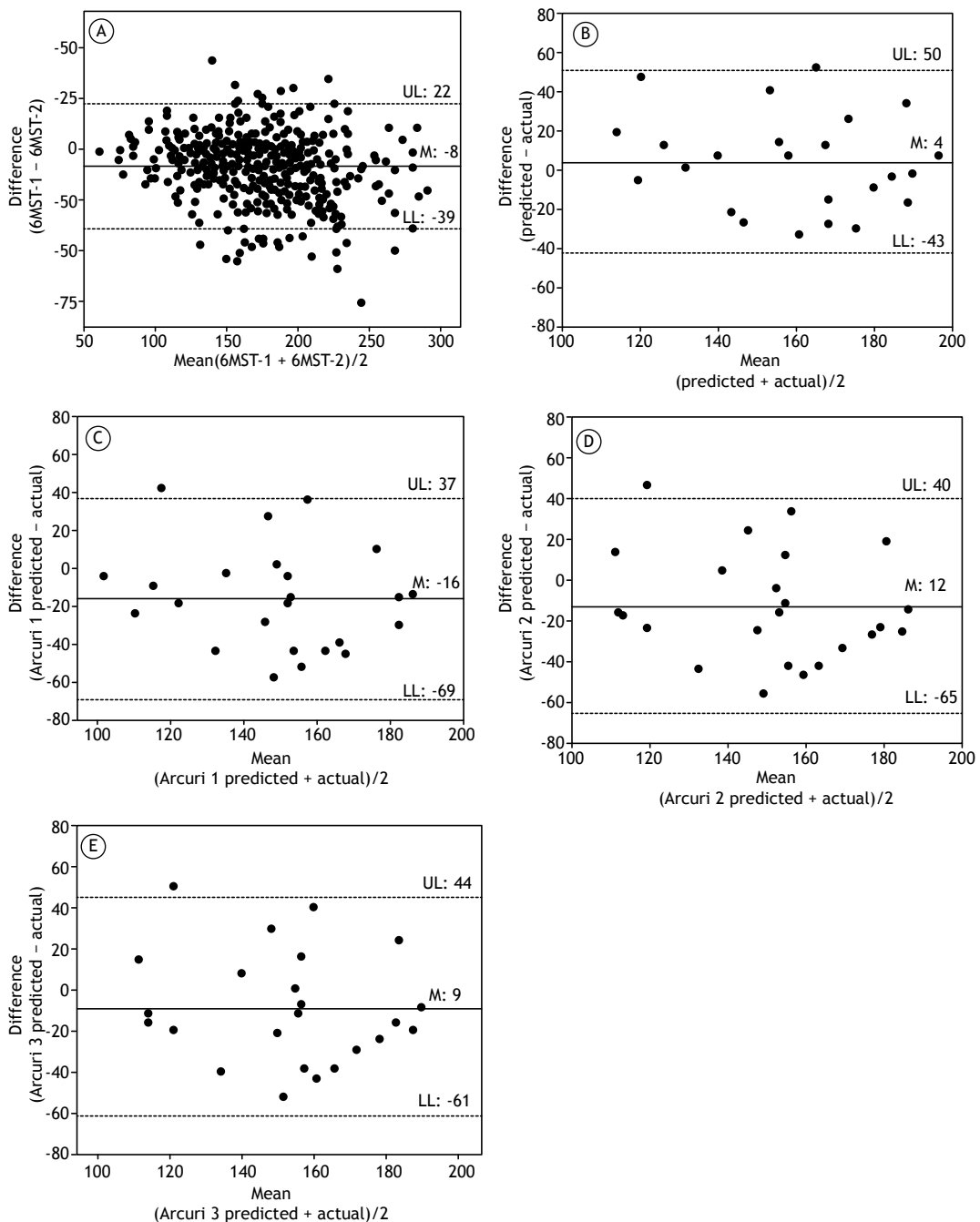


Figure S1. Bland-Altman plots. In A, plot of the difference against test and retest of the six-minute step test (6MST; N = 468). In B, plot of the difference against the predicted and actual values of the 6MST in the independent sample (N = 24). In C, plot of the difference against the predicted values using Arcuri 1 equations^a and actual values in the independent sample (N = 24). In D, plot of the difference against the predicted values using Arcuri 2 equations^a and actual values in the independent sample (N = 24). In E, plot of the difference against the predicted values using Arcuri 3 equations^a and actual values in the independent sample (N = 24). UL: upper limit of agreement; M: mean difference; and LL: lower limit of agreement. ^aEquations by Arcuri et al.⁽¹³⁾ Arcuri 1, 2, and 3, respectively, represent the equations for the first 6MST, the second 6MST, and the better of the two tests (different equations for male and female).

not continue), wheel the chair over for the patient to sit on, discontinue the test, and note on the worksheet the number of steps climbed, the time stopped, and the reason for stopping prematurely. When the timer is 15 s from completion, say this: "In a moment, I'm going to

tell you to stop. When I do, just stop climbing, and I will check you." When the timer rings (or buzzes), say this: "Stop!" and check the patient. Consider picking up the chair if they look exhausted. Record the total number of steps climbed on the worksheet.

Table S2. Physiological responses and symptoms induced by the first and second six-minute step test.^a

Variable	First test	Second test	p
HRRi, bpm	78 ± 12	83 ± 15	< 0.01
HRRf, bpm	141 ± 26	147 ± 26	< 0.01
HRR1, bpm	112 ± 22	116 ± 24	< 0.01
SBPi, mmHg	116 ± 12	116 ± 12	0.34
SBPf, mmHg	151 ± 21	151 ± 22	0.97
DBPi, mmHg	76 ± 9	76 ± 10	0.15
DBPf, mmHg	78 ± 11	77 ± 13	0.37
SpO ₂ i, %	97 ± 1.5	97 ± 1.3	0.22
SpO ₂ f, %	97 ± 1.8	96 ± 4.0	0.07
Dyspnea i,	0 (0-4)	0 (0-4)	0.02
Dyspnea f	4 (0-10)	4 (0-10)	0.09
Fatigue i	0 (0-5)	0 (0-9)	< 0.01
Fatigue f	4 (0-10)	4 (0-10)	0.05

i: initial; f: final. HRR1: HR at one minute of rest; SBP: systolic blood pressure; and DBP: diastolic blood pressure.
^aValues expressed as mean ± SD or median (minimum-maximum).

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