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Research article

Pregnancy reference intervals and exertion and breathlessness ratings for the six minute walk test in healthy nulliparous people

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ABSTRACT

Background: The Six Minute Walk Test (6MWT) is a simple, non-invasive, well-validated test that assesses cardiorespiratory fitness however is rarely used in pregnant people. It may have clinical utilization to assess fitness, breathing and exertion in pregnancy however no reference intervals exist for people 14^{+0} to 35^{+6} weeks gestation. We determined the reference intervals for distance walked for the 6MWT, including exertional and breathlessness ratings for this group.

Method: We conducted a prospective observational cohort study of 196 healthy nulliparous pregnant people in earlier pregnancy (EP) 14^{+0} to 23^{+6} weeks, and middle pregnancy (MP) 24^{+0} to 35^{+6} gestation, who performed a standardized 6MWT protocol including rating exertion and breathlessness (Rating Perceived Exertion (RPE) scale (1 none −15 maximal) and Modified Borg Dyspnea (MBD) scale (0 none – 10 maximal)).

Results: The mean ± SD distance walked was 548 ± 80.9 (EP) versus 547 ± 87.3 (MP) meters (m) P = 0.928. 6MWT reference intervals for the distance walked for the 6MWT were 392–704 m (EP) and 376–718 m (MP). Median (IQR) exertion and breathlessness ratings with exercise for the EP and MP group were 6 (4,7) and 0.5 (0,1) and 6 (4,8) and 0.5 (0,1) respectively. There were no adverse events.

Conclusion: The 6MWT is safe, feasible and acceptable in pregnant people. The reference intervals for the 6MWT are 392–704 m in people 14^{+0} to 23^{+6} weeks gestation and 376–718 m for people 24^{+0} to 35^{+6} weeks gestation. Exertion was light and breathlessness was just noticeable with the 6MWT.

This study provides the reference interval and expected exertional and breathlessness ratings for the Six Minute Walk Test, in people from 14^{+0} to 35^{+6} weeks gestation enabling the test to be used in clinical practice for assessment of exercise capacity and cardiovascular system fitness in pregnant people.

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1. Introduction

Cardiovascular disease, including cardiomyopathy and hypertensive disorders, account for over 30% percent of maternal mortality in the US [1]. Morbidity related to these conditions is also significant and early recognition of these problems may reduce adverse outcomes of pregnancy [2].

Maternal cardiovascular fitness has recently been implicated in the pathogenesis of pre-eclampsia. In 2014, Dennis and Castro proposed that pre-eclampsia arises as an adaptive response to a mismatch between maternal oxygen supply and demand for oxygen by the growing fetus [3]. It was proposed that this mismatch can arise from a combination of pre-placental, placental and post-placental factors. Maternal cardiovascular fitness is a pre-placental factor that may affect adequate oxygen delivery to the placenta, with reduced cardiovascular fitness, leading to reduced efficiency of oxygen delivery to the placenta, and an adaptive physiological response of increased cardiac output and compensatory maternal vascular bed vasoconstriction [3,4]. This is consistent with findings of a maternal hyperdynamic state characterized by increased cardiac output previously reported [5]. The release of placental biomarkers such as soluble fms-like tyrosine kinase-1 (sFLT1) may be related to this relative oxygen deficiency [6].

The quantification of maternal cardiovascular fitness through exercise testing offers a promising method to predict, recognise and monitor cardiovascular disorders, and to enhance the understanding of haemodynamic changes that occur during pregnancy including those associated with hypertensive disorders [7]. Despite the safety of exercise in pregnancy, the use of standardised exercise tests in the pregnant population, including their reference intervals, are notably missing from the literature [8–10].

The Six Minute Walk Test (6MWT) is a safe, non-invasive, inexpensive, reliable and validated exercise test that is often used to predict cardiovascular outcomes in the non-pregnant population [11]. The test itself includes assessment of exertion and breathlessness which in pregnancy is important due to the overlap of these symptoms in pregnancy with those of cardiorespiratory disease. The reference interval for the 6MWT has been established for healthy pregnant people 36 weeks and greater however there are no reference interval studies of healthy people less than 36 weeks gestation [12]. Given that there are significant cardiovascular and respiratory system changes prior to 36 weeks gestation [13,14], and cardiovascular conditions may develop in this period it is important to establish reference intervals for this test in earlier pregnancy. Quantification of perceived and actual exertion and breathlessness with exercise have not been studied in people less than 36 weeks. Therefore, there is no evidence base from which to advise pregnant people about what to expect at these gestations with exercise and with pregnancy itself, or to guide healthcare providers as to what might be reference values.

The primary aims were to determine the reference interval for the 6MWT distance, and to determine levels of exertion and breathlessness with exercise, in healthy nulliparous people between 14^{+0} and 35^{+6} weeks gestation. The secondary aims were to determine the reference interval for resting heart rate in healthy nulliparous people between 14- and 36-weeks' gestation, to determine hemodynamic responses to exercise, and to quantify expected ratings of exertion and breathlessness with exercise and pregnancy.

2. Materials and methods

2.1. Subjects

The study underwent prospective IRB (HREC 15/23 Royal Women's Hospital, Parkville, Australia) and trial registration (Australian and New Zealand Clinical Trials registry (Universal Trial Number: U1111-1173-7990 ACTRN: 12615000964516 (https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=369216). Healthy nulliparous pregnant people were recruited from the antenatal clinic of the tertiary referral obstetric hospital. People were recruited to the study during weekdays and during clinic hours. After initial screening by investigators to determine eligibility, recruitment of participants was achieved both via directly approaching people attending their antenatal visit and via direct telephone contact in those people who investigators were unable to talk to face to face. Written informed consent was obtained from all participants. Two groups of healthy nulliparous pregnant people aged 18–40 years were recruited. Group 1 was between 14^{+0} and 23^{+6} weeks gestation (earlier pregnancy group (EP)) and group 2 was between 24^{+0} and 35^{+6} weeks gestation (middle pregnancy group (MP)). Exclusion criteria were people with any pre-existing condition that limited their ability to exercise, pre-existing or gestational diabetes, pre-existing or pregnancy acquired cardiovascular disease, multiple pregnancy, people who were tobacco users, people with body mass index over 32 kg/m^2, people classified as Society of Anesthesiologists (ASA) Classification III or greater; and any person in labor or post-partum or who was unable to consent to the study.

2.2. Six minute walk test

The study followed previously reported methodology [12]. The study was conducted in a 30 m, indoor, corridor following standardized guidelines used in clinical practice, and published by the American Thoracic Society [15] and the National Heart Foundation of Australia (NHFA), including using specific language, used by each investigator, that advised the participant how to undertake the test [16]. (Supplemental material). Weight was measured in kilograms for each participant using Sanitas SGS06 Glass Scales, and body mass index was calculated using known height in centimetres. The test began with a 5-min seated rest period during which time standardized instructions were read to each participant. Resting heart rate (HR), systolic blood pressure (SBP), diastolic blood pressure (DBP), respiratory rate (RR) and oxygen saturation (SpO_2) were measured with a calibrated automated blood pressure machine (Criticare VitalCare 506N3 Series Monitor). Each person then underwent two 6MWTs separated by a recovery period of at least 5 min. There was no warm up period at the start of the first test. During each recovery period, HR, SBP, DBP, RR and SpO_2 were measured every minute, with the first measurement immediately after exercise then each minute for at least 5 min until heart rate had returned to

the resting value (or below) or within four beats per minute (bpm) from resting measurement. A maximum of 15 min of measurements were made after each test. The 6MWT and recovery period were then repeated. Measurements of the two distances walked by each participant were made using a Crescent NMW1 measuring wheel, which measured distance to the nearest 10 cm.

2.3. Rating scores

After the completion of the second recovery period, participants were asked to complete the Rating of Perceived Exertion Category Scale (15-point scale, 1 = very, very light, 15 = very, very hard) and the Modified Borg Dyspnoea Scale (12-point scale (0 = Nothing at all, 10 = Maximal [also includes 0.5 defined as very, very slight just noticeable breathing difficulty]) (Supplemental material). Participants were also asked six questions to assess their expectations and actual experience of breathlessness and exertion during this stage of pregnancy and during the 6MWT (Supplemental material). The study procedure that the participants underwent is shown in Supplemental Fig. 1S.

2.4. Statistical analysis

Data were analysed using SPSS Statistics Version 24 (IBM© SPSS© Statistics Version 24 IBM Corporation 2016, Chicago, Ill). Da Silva demonstrated a mean (SD) of 6MWT distance in a group of healthy pregnant people, between 33 and 38 weeks gestation, of 497 (38) m [7]. Using these data, the sample size to demonstrate a difference of 30 m (a clinically significant difference in older age groups) was 26 in each group. The Type I error probability associated with this test of this null hypothesis is 0.05. As however the primary aim was to determine refence intervals, we used a larger convenience sample of 100 people in each group as the sample size. The distribution of data was assessed using Kolmogorov-Smirnov and Shapiro-Wilk tests. Data are presented as mean, standard deviation (SD), median (quartiles) [lowest and highest value]), or number and percentage as appropriate. Effect sizes for differences, when appropriate, were calculated using the mean difference between the independent groups. The 95% reference interval for both the 6MWT distance and HR were calculated for the EP and MP groups using the mean distance of the two 6MWT walked $\pm 1.96 \times$ SD, or the mean HR $\pm$ 1.96 $\times$ SD. The 95% confidence interval (CI) of the reference interval was calculated using the standard error of the limit of the reference interval such that a 95% CI for the lower and upper limits were determined (https://www-users.york.ac.uk/~mb55/intro/refint.htm) (Supplemental material).

Comparisons between and within groups were performed using paired or unpaired t tests, one-way ANOVA with Tukey's multiple comparison test or Kruskal-Wallis test with Dunn's multiple comparisons test as appropriate. Mean differences, 95% confidence intervals (CI) and two-sided P values are given with a significance level alpha of 0.05. Pearson correlation coefficient with 95% CI was calculated to examine the relationship between resting HR and distance walked in the first 6MWT, and HR change with exercise and the time taken to recover from exercise.

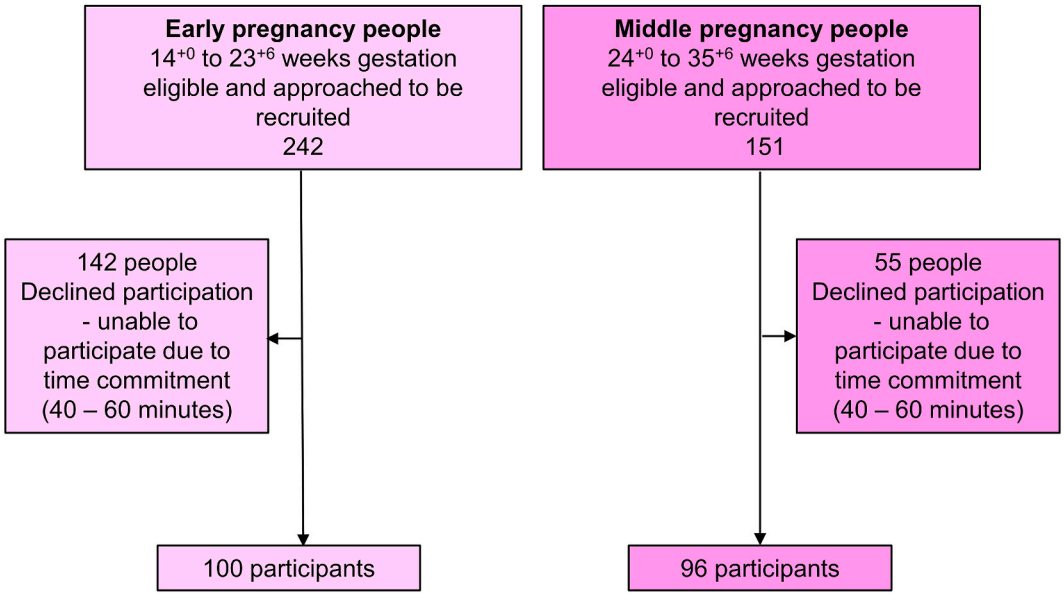

Fig. 1. Participant flow chart
Created with BioRender.com.

3. Results

One hundred and ninety six pregnant people were recruited (100 EP people, and 96 MP people). One participant in the middle pregnancy group did not complete the second 6MWT due to time constraints (Fig. 1). The time for each participant to complete the study was 40–60 min. No person experienced any adverse events.

The participant characteristics are shown in Table 1. There was evidence of a difference in weight and body mass index between the EP and MP groups (mean difference ± SD, 5.1 ± 1.5 kg, 95% CI 2.2–8.0 kg, P = 0.001, and mean difference ± SD, 1.8 ± 0.5 kg/m^2, 95% CI 0.9–2.7 kg/m^2, P < 0.001 respectively).

Six Minute Walk Test data and reference intervals for the EP and MP groups are shown in Table 2. The average walking speed was 3.4 ± 0.50 miles/hr in the EP group 3.4 ± 0.56 miles/hr in the MP group (p = 0.954). The reference interval for the distance walked in the 6MWT was 392–704 m for the EP group and 376–718 m for the MP group (Table 2). The estimation plot of resting heart rates between the early pregnancy and the middle pregnancy groups is shown in Supplemental Fig. S2.

Combined with the work in people from 36 weeks gestation, there are now reference intervals for this test for all gestations from 14 weeks (Table 3).

Resting physiological variables, and response to exercise are shown in Table 2. Heart rate, SBP and DBP during the first 5-min recovery period for the EP and MP groups after the first 6MWT are shown in Fig. 2.

Panels EP1 and MP1 show time variation of heart rate for the early (EP1) and middle (MP1) pregnancy groups commencing at time point −1 (resting measurement). Panels EP2 and MP2 show time variation of systolic blood pressure for the early (EP2) and middle (MP2) pregnancy groups commencing at time point −1 (resting measurement). Panels EP3 and MP3 show time variation of diastolic blood pressure for the early (EP3) and middle (MP3) pregnancy groups commencing at time point −1 (resting measurement).

The earlier pregnancy (EP) 14^{+0} to 23^{+6} weeks gestation group is shown on the left side of figure, and the middle pregnancy (MP) 24^{+0} to 35^{+6} weeks gestation group is shown on the right side of figure. Times are in minutes with −1 representing time at rest, 0 representing time at peak exercise (immediately on cessation of exercise), and 1, 2, 3, 4, 5 representing 1–5 min after the cessation of exercise.

Heart rate, systolic and diastolic blood pressure all demonstrated an initial peak within 1 min of completing exercise, followed by a rapid recovery. HR, SBP and DBP increased by approximately 18 BPM, 11 mmHg and 4 mmHg in the EP group, and 22 BPM, 14 mmHg, and 5 mmHg in the MP group (Table 2 and Supplemental Table S1). Heart rate recovery to resting values occurred within 5 min in 62% of people in the EP group and 47% in the MP group after the first 6MWT, with a longer recovery time for the second test for both groups (Supplemental Fig. 3).

There was evidence of an increase in resting heart rate and a decrease in resting systolic blood pressure in the MP group compared with the EP group (HR mean difference 4 ± 1.5 BPM, 95% CI 0.9 to 6.8 BPM, P = 0.010; SBP mean difference - 8 ± 1.3 mmHg, 95% CI -10.3 to −5.1 mmHg, P < 0.001). The reference interval for resting heart rate was 59–103 BPM for the EP group and 66 to 104 BPM for the MP group Table 2.

In the MP group a lower resting HR was associated with an increased distance walked in the 6MWT in both the first and second 6MWT (r = −0.207, 95% CI -0.391 to −0.006 P = 0.044 test 1; r = −0.217, 95% CI -0.4 to −0.02 test 2 P = 0.035). This relationship was not present in the EP group. There was evidence, in both the EP and MP groups for both test 1 and test 2 that a greater heart rate change with exercise was associated with an increased recovery time from exercise (EP Test 1 r = 0.309 95% CI 0.114 to 0.480 P = 0.002; EP Test 2 r = 0.3499 95% CI 0.328 to 0.638 P < 0.0001; MP Test 1 r = 0.536 95% CI 0.367 to 0.671 P < 0.0001; MP Test 2 r = 0.405 95% CI 0.208 to 0.570 P = 0.0001 respectively).

Table 4 shows expected and actual exertion and breathlessness data for the EP and MP groups. Exertion with the 6MWT, for both groups, was rated at a median value of 6 corresponding with the “fairly light” category on the scale. Breathlessness with exercise, for both groups, rated at a median value of 0,5 corresponding with the “very, very slight (just noticeable) category on the scale. Regarding everyday life while being pregnant, people expected to feel a fairly light level of exertion and a slight amount of breathlessness, and at MP they expected to feel a somewhat hard level of exertion and a moderate amount of breathlessness. For both groups the actual level of exertion, with exercise, and pregnancy, was less than expected.

Table 1
Participant characteristics.

Characteristic	EP group (14^{+0} to 23^{+6} weeks) n = 100	MP group (24^{+0} to 35^{+6} weeks[a]) n = 96	P value (95% CI)
Age (years)	31 ± 5.2	32 ± 3.6	0.520
Gestation (weeks)	21 (19.7, 21.8) [14.6, 23.9]	30 (28.0, 32.7) [24.1, 35.6]	<0.001 (8.8–10.3)
Height (m)	1.7 ± 0.1	1.7 ± 0.1	0.826
Weight (kg)	67 ± 9.7	72 ± 10.4	0.001 (2.2–8.0)
BMI (kg/m^2)	24 ± 3.0	26 ± 3.3	<0.001 (0.9–2.7)

EP = earlier pregnancy; MP = middle pregnancy.
Data are mean ± SD, median (IQR) [lowest, highest value]; CI = confidence interval; SD = standard deviation; BMI = body mass index.
[a] Gestational weeks.

Table 2
Resting physiological variables, Six Minute Walk Test data and reference intervals for the two pregnancy groups.

Variable	EP group (14^{+0} to 23^{+6} weeks) n = 100	MP group (24^{+0} to 35^{+6} weeks) n = 96	P value (95% CI)
Distance walked test 1 (m)	538 ± 78.9 [374, 718]	539 ± 86.2 [343, 809]	0.951
Distance walked test 2 (m)	557 ± 82.1 [379, 749]	556 ± 89.2 [370, 827]	0.911
Average distance walked (m)	548 ± 80.9	547 ± 87.3	0.928
6MWT reference interval (m)	392 to 704	376 to 718	NA
95% CI lower limit, upper limit (m)	365 to 419, 677 to 731	346 to 406, 688 to 748	NA
Resting systolic blood pressure (mmHg)	114 ± 10.1 [94,138]	107 ± 8.4 [87,132]	<0.001 (−10.3 to −5.1)
Resting diastolic blood pressure (mmHg)	71 ± 8.8 [52, 93]	70 ± 6.5 [55, 84]	0.429
Resting oxygen saturation (%)	98 ± 0.8 [96, 99]	98 ± 1.5 [94, 100]	0.338
Resting respiratory rate (breaths/min)	16 ± 2.1 [12, 24]	15 ± 2.6 [12, 24]	0.003 (−1.7 to −0.4)
Resting heart rate (beats/min)	81 ± 11.1 [55, 108]	85 ± 9.5 [60, 105]	0.010 (0.9–6.8)*
Maximal heart rate (beats/min) test 1	102 ± 19.4 [58, 160]	108 ± 17.5 [76,160]	0.018 (1.1–11.6)
Maximal heart rate (beats/min) test 2	105 ± 19.0 [66, 162]	111 ± 19.0 [80, 163]	0.034 (0.4–11.2)
Heart rate change with exercise (beats/min) test 1	18 (8.25, 28.75) [-17, 67]	22 (11.0, 30.0) [-5, 67]	0.107
Heart rate change with exercise (beats/min) test 2	21 (11,33.75) [-9, 67]	21 (13.0, 37.0) [-3, 78]	0.327
Heart rate recovery time (min)	1 [(1,2) [1, 13][a]	1 [(1,3) [1,11][b]	0.081
Resting heart rate reference interval (beats/min)	59–103	66–104	NA
95% CI lower limit, upper limit (beats/min)	55 to 63, 99 to 107	63 to 69, 101 to 107	NA

EP = earlier pregnancy; MP = middle pregnancy. *Gestational weeks. Data are mean ± SD, median (quartiles) [lowest, highest value]. CI = confidence interval. CI presented when $P < 0.05$. Recovery time was defined as the time taken in minutes for the heart rate (HR) to return to resting level (baseline ± 4 BPM) once the first Six Minute Walk Test (6MWT) was completed. The HR change with exercise was defined as the first HR recorded after the completion of the first 6MWT minus the resting HR. m = meters; min = minutes. One MP person did not do second 6MWT. *mean difference (effect size) 3.8 ± 1.5 beats/min.

[a] For the 95 people recovered in EP group.

[b] For the 88 people who recovered by 15 min. Calculations for the determination of the lower and upper reference interval confidence intervals are shown in Supplementary material.

Table 3
Reference intervals for the Six Minute Walk Test in pregnancy.

	Gestation range (weeks)	**Sample size (n)**	**Distance (mean ± SD)** (m)	**Reference interval (m)**
Earlier Pregnancy	14^{+0} to 23^{+6}	100	548 ± 80.9	392 to 704
Middle Pregnancy	24^{+0} to 35^{+6}	96	547 ± 87.3	376 to 718
Late Pregnancy [12]	$\geq 36^{+0}$	300	488 ± 94.9	302 to 674

SD = standard deviation.

4. Discussion

We confirmed that the Six Minute Walk Test is safe, feasible and acceptable to pregnant people. We determined for the first time the reference interval for the 6MWT in these gestational periods. Low levels of exertion and breathlessness with the 6MWT were reported confirming that this test is an assessment of submaximal exercise capacity.

Expected exertion with exercise and pregnancy were less than levels actually experienced by people in pregnancy.

We have provided novel reference data for heart rate, systolic blood pressure and diastolic blood pressure responses in healthy pregnant people, of gestations 14^{+0} to 35^{+6}, with exercise.

Many studies across the world have created reference ranges for the 6MWT differing in age, sex, and population [17–25]. However these studies only had a limited number of people aged less than 40 years included in the "normal" population. Additionally, despite the extensive clinical use of the 6MWT in non-pregnant adults, there is little published work reporting the reference intervals for distance walked in pregnant people using the 6MWT. One study with a small sample size of participants is of importance as it is the first study using the 6MWT in people with preeclampsia [7]. Da Silva and colleagues found that the distance walked in 37 people with preeclampsia compared with 37 healthy pregnant control subjects, at an average gestational age of 37 weeks, was significantly lower (421 ± 59 m versus 497 ± 38 m $P = 0.001$) [7]. Within the context of da Silva's work, we have shown that the mean distance walked by people in EP and MP were similar to each other but shorter than distances walked in late pregnancy in the healthy cohort [7,12]. Together, these results suggest that exercise capacity may be reduced in healthy people in late pregnancy. The issue of a reduced exercise capacity in people with preeclampsia requires further investigation. Interestingly, da Silva and colleagues also reported higher minute ventilation in people with preeclampsia compared to healthy gestationally matched pregnant controls. da Silva and colleagues speculated that this may be due to higher leptin levels (not measured in their study) in people with preeclampsia compared with healthy pregnant people, however the finding also supports the hypothesis that there is a need for increased oxygen delivery, through both increased cardiac output and increase minute ventilation, in people with preeclampsia contributing to the development of

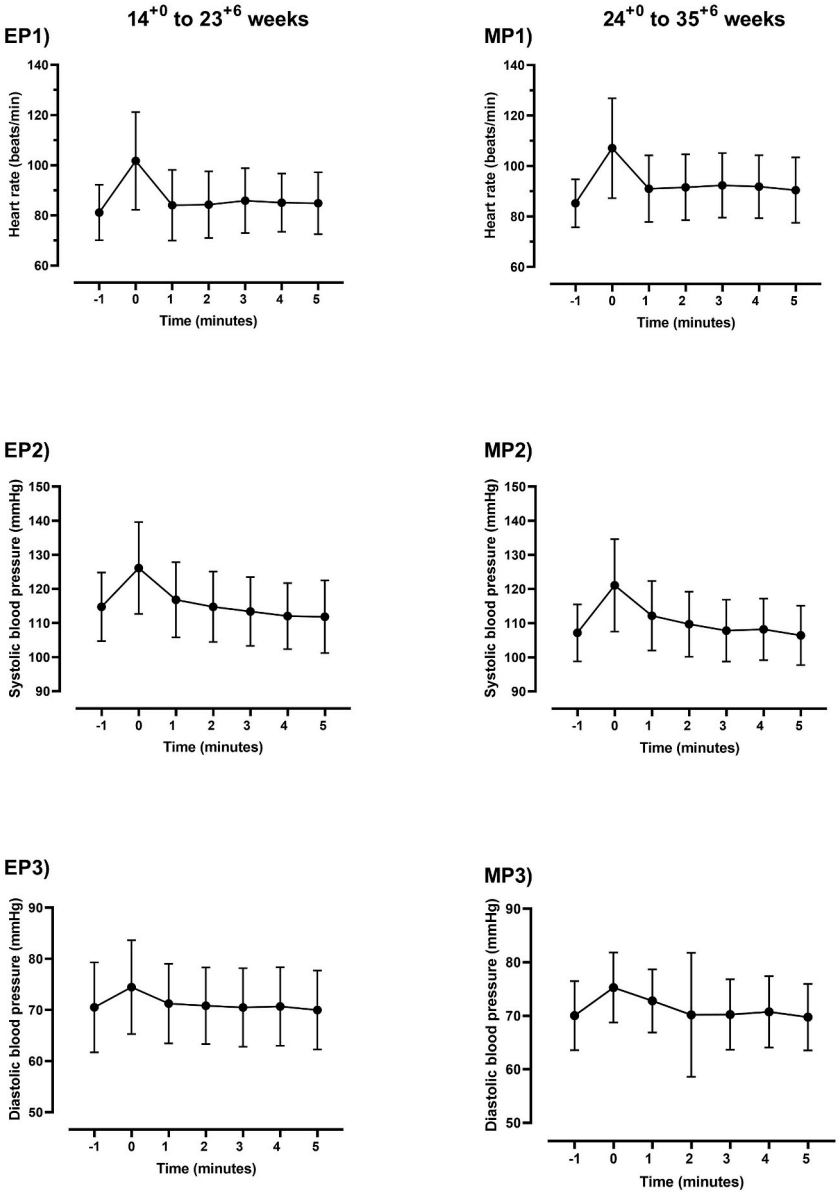

Fig. 2. Heart rate, systolic and diastolic blood pressure changes from resting to 5 min after exercise for the two pregnancy groups.

hyperntesion [3].

Heart rate changes and recovery times, as well as systolic and diastolic blood pressure changes, with submaximal exercise with the 6MWT in the EP and MP groups have also been reported for the first time.

4.1. Clinical implications

The establishment of reference intervals in a standardized healthy population enables the 6MWT to be used in clinical practice, including in the emerging subspecialty of cardio-obstetrics, and in research studies investigating cardiovascular fitness and hemodynamic responses in pregnant people.

We reported the reference interval for HR for people 14^{+0} to 23^{+6} and 24^{+0} to 35^{+6} weeks gestation. We showed that no person in the cohort had a resting HR more than 108 BPM. The knowledge of a reference interval and maximal HR at gestations from 14^{+0} to 35^{+6} weeks may assist with refinement of early warning charts used to detect clinical deterioration of obstetric patients, and for identifying patients in whom further cardiovascular investigation is required. Heart rates above 110 BPM at rest are likely to be pathological.

The data quantifying expected and actual exertion and breathlessness with exercise and pregnancy has shown that there was an

Table 4
Expected and actual exertion and breathlessness data for the two pregnancy group.

Variable	EP group[a] (14^{+0} to 23^{+6} weeks) n = 100			n/total n (%) overestimating exertion[c]	n/total n (%) overestimating breathlessness[c]
	Expected	Actual	P value		
Exertion with 6MWT	6 (5,8) [1,19]	6 (4,7) [1,13][b]	<0.001	43/79 (54%)	NA
Exertion with pregnancy	6 (4,8) [1,15]	6 (3,7) [1,10]	0.014	39/70 (56%)	NA
Breathlessness with 6MWT	3 (2,3) [0,8]	0.5 (0,1) [0,3][b]	<0.001	NA	66/79 (84%)
Breathlessness with pregnancy	2 (0,5,3) [0,9]	2 (0.5,3) [0,10]	0.966	NA	34/67 (51%)
	MP group[a] (24^{+0} to 35^{+6} weeks) n = 96				
Exertion with 6MWT	7 (5.75,8.0) [1,12]	6 (4,8) [1,13][b]	0.004	40/76 (53%)	NA
Exertion with pregnancy	8 (6,8) [1,14]	6 (4,7) [1,12]	<0.001	48/71 (66%)	NA
Breathlessness with 6MWT	3 (2,3) [0,8]	0.5 (0,1) [0,3][b]	<0.001	NA	70/79 (87%)
Breathlessness with pregnancy	3 (2,3.75) [0,8]	2 (1,3) [0,8]	0.028	NA	37/56 (66%)

EP = earlier pregnancy; MP = middle pregnancy.
Additional questions were added during recruitment for the EP group which occurred before the MP group. Of the EP group 79 people were asked. Of the MP group every person was asked additional questions.
Data are median (quartiles), [range lowest and highest value]. NA = not applicable.
6MWT = Six Minute Walk Test.
[a] Gestational weeks.
[b] recorded for 196 people as part of 6MWT.
[c] only people who had expectations.

overestimation by the participants of exertion and breathlessness with exercise and pregnancy. This may lead to pregnant people not exercising at all or not exercising to the exertional levels that they are capable of, likely due to inaccurate expectations. This may have the clinical consequence of pregnant people not undertaking exercise to assist with the maintenance of healthy body mass, or to pregnant people not undertaking activities that may keep them socially engaged for their positive mental health.

The data can assist with the education of pregnant people by providing clarity about the actual exertional and breathlessness ranges that occur during submaximal exercise, and during pregnancy. These results importantly also may help to inform clinicians about expected levels of exertion and breathlessness in people during exercise and with pregnancy and help to emphasise the findings of the MBBRACE-UK [2] report that any significant or persistent breathlessness in pregnant people should be considered abnormal. These results could therefore allow better identification of those people experiencing abnormal levels of breathlessness or exertion who should have further investigations to exclude any underlying pathological cause such as emerging cardiovascular compromise.

4.2. Research implications – future studies

Future studies should determine what is the minimally clinically important difference (MCID) in distance walked for the pregnant population. In chronically ill older adults, for example, the MCIC is approximately 30 m, and a decrease in the 6MWT distance of ≥30 m compared with the lower limit of the reference interval signifies increased mortality in the next 12 months [26,27]. Future studies should also define reference intervals for other exercise tests in pregnancy such has the incremental shuttle walk test, a non-volitional externally paced test, as it is known that motivation may influence distance walked [28]. Future studies should also examine the use of software and the application of artificial intelligence in this area. Future studies should also collect participants daily and weekly activity levels to determine pre-existing fitness levels.

4.3. Limitations and strengths

The main limitation of the study was that there was no assessment of the participants' level of fitness, the exercise they had already done that day, or their food intake in relation to the timing of the test. This may limit the study's internal and external validity. Secondly, the study recruited voluntary participants who were motivated and interested in exercise. As such they were a very healthy group of people who enjoyed exercising and introduced selection bias to the study. The restrictive inclusion criteria, necessary to determine reference intervals, limit generalizability to other groups of pregnant patients such as those with hypertension or respiratory disease and may assist with determine what is abnormal exercise capacity in these other groups The differences in weight and BMI of the EP and MP groups mean that their data cannot be pooled and need to be examined separately. We did not follow these people

longitudinally or perform the 6MWT serially on them. Lastly, the interpretation of the results of breathlessness and exertion during exercise and pregnancy is limited by the scoring systems themselves. The perceptions of "breathlessness" and "exertion" are subjective and are determined by the individual's interpretation of the scoring systems and their own perception of the sensations. Assessing participants exercise levels and determining participants' inclusion based on a wider exercise capacity, with subgroups based on pre-existing fitness levels, may enhance future studies.

5. Conclusions

This study has determined the reference intervals for the distance walked in using the 6MWT in people from 14^{+0} weeks to 35^{+6} weeks gestation, and ratings of exertion and breathlessness with exercise. Heart rate responses to exercise confirmed the submaximal nature of the test, and exertional and breathlessness questioning determined that both exertion and breathlessness were overestimated by the majority of pregnant people during exercise and in pregnancy. The results provide a reference interval for the 6MWT that can easily and inexpensively be used in clinical practice to assess and monitor symptoms of pregnancy and cardiorespiratory pathology through pregnancy. Results of this study provide evidence to inform exercise program development, and educational programs for both clinicians and pregnant people. Integrating this simple test into clinical practice may enable the prediction of adverse pregnancy outcomes based on distances walked, and exertion, breathlessness, heart rate and blood pressure responses to exercise. Responses beyond the reference intervals described in this study may predict the development of cardiovascular complications in later pregnancy including the development of hypertension.

Declaration and disclosure statement

The authors report no conflict of interest.

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Trial registration

The study was prospectively registered with the Australian and New Zealand Clinical.

Date of registration – 15/9/2015; Date of initial participant enrolment - 4/11/2015; Clinical trial identification number; ACTRN12615000964516; URL of the registration site - https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=369216.

This work was presented at the Obstetric Anesthesia Special Interest Group meeting, Australian and New Zealand College of Anaesthetists, Australian Society of Anaesthetists, New Zealand Society of Anaesthetists Annual Scientific Meeting, Sydney, Australia, 5–6 May 2018 and the Obstetric Anaesthetists' Association of and published in abstract form: Dennis AT, Francis A, Leeton L. Assessment of exercise capacity and oxygen utilization, and the prediction of pre-eclampsia in women in early pregnancy using the Six Minute Walk Test International Journal of Obstetric Anesthesia 2018; 35; (S1):O.10: S11.

Tweetable statement

The reference interval for the Six Minute Walk Test for people 14^{+0} to 23^{+6} weeks gestation is 392–704 m. For people 24^{+0} to 35^{+6} weeks gestation, it is 376–718 m. Exertion is light and breathlessness is just noticeable when doing the test.

Ethics declarations

This study was reviewed and approved by the Human Research Ethics Committee (HREC) of the Royal Women's Hospital Parkville Australia HREC number 15/23 and was prospectively registered with the Australian and New Zealand Clinical Trials registry (Universal Trial Number: U1111-1173-7990 ACTRN: 12615000964516 (https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=369216. All participants provided informed consent to participate in the study.

Data availability statement

Data associated with this study has not been deposited into a publicly available repository. Data included in article/supplemental material/referenced in article.

CRediT authorship contribution statement

Alaina R. Francis: Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Tahila J. Sugrue:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Alicia T. Dennis:** Writing – review & editing,

Supervision, Software, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e25863.

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