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Segregating the Distinct Effects of Sedentary Behavior and Physical Activity on Older Adults' Cardiovascular Profile: Part 2-Isotemporal Substitution Approach.

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1 **Segregating the distinct effects of sedentary behaviour and physical activity on older**
2 **adults' cardiovascular profile: Part 2- Isotemporal substitution approach.**

3

4 **Change in physical behaviour on vascular profile.**

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Abstract

Background – The aim of the study was to provide an isothermal substitution model, to predict how changes in physical behaviour may affect the cardiovascular parameters (CVP) of older adults.

Methods – Participants wore a thigh-mounted accelerometer for seven days. Phenotype of the carotid, brachial, and popliteal artery was conducted using ultrasound. Isothermal substitution was used to simulate the degree to which replacing one hour of physical behaviour with another would affect CVP.

Results – Substitution of sedentary behaviour (SB) with standing and sporadic moderate-vigorous physical activity (sMVPA, MVPA accumulated in bouts < 10 mins) would reduce resting heart rate (-6.20 bpm [-12.1, -0.22], -3.72 bpm [-7.01, -0.44], respectively). Substitution of SB with light intensity physical activity, would reduce carotid artery diameter (-0.54 mm [-1.00, -0.07]). Substitution of standing with sMVPA would increase popliteal artery diameter (1.31 mm [0.11, 2.51]).

Conclusions – Our modelling suggest that an accumulation of MVPA bouts that are shorter than the recommended 10-minute minimum may still improve CVP, with lower intensity PA also influencing CVP. Our findings are a promising avenue for lifestyle interventions in older adults in order to reduce the ageing effects on CVP for those who cannot engage or sustain sufficient MVPA.

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Introduction

It is becoming evident that sedentary behaviour (SB) affects a number of physiological parameters independent of the amount of moderate to vigorous intensity physical activity (MVPA) engagement^{1,2}. With time being finite within a day (i.e. 24 hour endpoint), engagement in one physical behaviour (PB)³ will offset the amount of time that can be spent performing another. Standard regression modelling fails to recognise the time constraints and therefore the use of multiple measures of PB within a regression model will not account for the time that is displaced by engaging in a specific bout of PB.

Isotemporal substitution regression models recognise that time is finite by including a measure of total PB (e.g. sum of waking hours SB and physical activity [PA]), which is kept constant and therefore, provides the opportunity to substitute one PB for another, thereby reflecting the realities of daily life⁴. Rather than prediction, per se, isotemporal substitution reflects the decisions people have made (e.g. prolonged SB) and offers an extrapolation of what would happen should they decided to do something different (e.g. MVPA). Therefore, this analysis may be more advantageous to public health PB action plans, as it clearly illustrates what will happen to markers of health if habitual PB levels and/or patterns are changed. In older adult populations, isotemporal substitution has mainly been used to assess the effect on cardio-metabolic⁵⁻⁷ rather than cardiovascular parameters⁸. However in the one study to date, to the author's knowledge, that cardiovascular parameters have been assessed, it has demonstrated promising results, for instance, suggesting the substitution of SB with light intensity PA (LIPA) would reduce the relative risk of cardiovascular disease (CVD) prevalence within older adult cohorts⁸. Light intensity physical activity is a promising intervention to reduce SB for older adult populations as it can arguably prove to be easier (in

1 comparison to MVPA) to comply with, and be accumulated to consist the greater majority of
2 a 24-hour simplex⁹.

3 Moreover, the ten-minute minimum threshold for an MVPA bout (₁₀MVPA),
4 highlighted in the PA guidelines¹⁰, to show clinically beneficial outcomes, has not been
5 examined using isometeor substitution. If sporadic MVPA (sMVPA, MVPA accumulated
6 in bouts of less than 10 continuous minutes) has beneficial effects on cardiovascular health,
7 this alternative mode of accumulating MVPA would likely allow older adults to improve
8 their health within their physical capacities, and maintain this PB profile in the long term.
9 Therefore, the objective of part 2 of this series was to simulate the degree to which the
10 substitution of SB and lower intensity PA with MVPA would have positive effects on
11 cardiovascular health markers and vice versa, in older adults. The aim was to provide a time-
12 constrained, alternative to bivariate/multivariate regression modelling tool, to predict how
13 changes in PB may affect the cardiovascular health of older adults. It was hypothesised that
14 substituting SB with any intensity of PA would improve cardiovascular parameters and that
15 substituting a PB with a higher intensity would improve cardiovascular profile. It was also
16 hypothesised that substituting SB with ₁₀MVPA would have a greater effect on
17 cardiovascular parameters than seen with sMVPA substitutions.

18 **Methods**

19 Ninety-three older participants (73.8 ± 6.22 years, 60 – 89 years, 55% female, table 1)
20 who did not suffer from an untreated cardiovascular disease (CVD), had not sustained a PB
21 limiting injury within the last three months, who were independently mobile, and deemed
22 generally healthy were recruited for the study. Participant approval for study inclusion was
23 provided with a written informed consent and the study was granted approval by the
24 Manchester Metropolitan University ethics sub-committee. Participants visited the laboratory
25 on two occasions separated by at least seven days.

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2 **Table 1 Participant demographics.**

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Variable	Mean (Standard Deviation)
Age (yrs)	73.8 (6.22)
Height (m)	1.65 (0.08)
Mass (kg)	75.9 (13.1)
BMI (kg·m ²)	27.9 (4.71)
Primary CVD Meds (%)†	48.0
(in)direct CVD Meds (%)‡	59.0
Hydration (%)	50.6 (7.15)
SB (hrs·day ⁻¹)	9.68 (1.30)
Standing (hrs·day ⁻¹)	1.10 (0.40)
LIPA (hrs·day ⁻¹)	1.95 (0.60)
sMVPA (hrs·day ⁻¹)	2.58 (0.66)
₁₀ MVPA (hrs·day ⁻¹)	0.08 (0.18) _m
Total PB (hrs·day ⁻¹)	15.4 (4.77) _m

m Median (Interquartile Range). † Participants are currently prescribed an amount of medication that reduces the risk or treats CVD (i.e. statins, warfarin). ‡ Participants are currently prescribed a medication that may affect the cardiovascular system either directly or as a side effect. SB – sedentary behaviour, LIPA – light intensity physical activity, sMVPA – sporadic moderate to vigorous intensity physical activity (accumulated in bouts < 10 mins), ₁₀MVPA – 10 minute moderate to vigorous intensity physical activity (accumulated in bouts ≥ 10 mins), Total PB – total physical behaviour, BMI – body mass index.

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First Laboratory Visit

The methods follow that of Part 1 of the current series of papers. In brief, participant demographics (table 1) were collected during the first visit. Medication use was provided through hard copies of current prescriptions, with hard copy medication later categorised as primarily used to target CVD or could indirectly target CVD. This information is reported as Primary CVD Meds (number of drugs primarily targeting CVD) or (in)direct CVD Meds (sum of primary CVD meds and drugs that may indirectly affect CVD). Participants were fitted with a commercially available, dominant leg, thigh mounted (anterior aspect, at 50% of greater trochanter to femoral condyle distance) triaxial accelerometer (GENEA, GENEActiv Original, Activinsights Ltd, Kimbolton, UK) using a waterproof adhesive patch (3M Tegaderm Film, North Ryde, Australia), for seven consecutive free-living days. Residual G ($\text{Residual G} = \sqrt{[\text{standard deviation } x]^2 + [\text{standard deviation } y]^2 + [\text{standard deviation } z]^2}$), adapted from our previous work on older adults total movement¹¹, was used to analyse the 10.0 s epoch (60.0 Hz) GENEActiv data and termed The Cheshire Algorithm for Sedentarism (CAS). CAS was developed using cut-off points developed in our laboratory calibrated against the expired gas samples of a sub sample of 20 older adults for ten PBs. SB was recognised as any seated or reclined posture, using the GENEActiv axes orientation, similar to that of the ‘Sedentary Sphere’¹², whilst standing was any standing posture that had a Residual G¹¹ value below the SB-LIPA cut-off point of 0.057 G (representing 1.50 Metabolic Equivalent Task [METs]). Remaining standing postures were then classified into LIPA or MVPA dependent on whether they met the LIPA-MVPA cut-off point of 0.216 G (representing 3.00 METs). MVPA was categorised as sMVPA if bouts were less than 10 continuous minutes in duration or ₁₀MVPA if bouts were greater than or equal to 10 continuous minutes in duration. One MET was equal to the Resting Metabolic Rate (RMR)

1 (whilst seated) of the participants to account for individual differences in physical fitness.
2 There was a strong association between Residual G and METs ($r^2 = 0.89$, $p < 0.001$). Postural
3 identification showed a perfect agreement with known time spent performing SB and PA
4 (Cohen's kappa = 1.00 [95% CI 1.0, 1.0], $p < 0.001$). Residual G cut-off points and MET
5 thresholds had a strong agreement for PB intensity classification (Cohen's kappa = 0.81 [95%
6 CI 0.49, 1.31], $p < 0.001$). Sleeping hours data was collected through a self-reported Sleep
7 Diary (wake-up time, lights-off go to sleep time, naps not included) throughout the
8 monitoring week.

9 Second Laboratory Visit

10 Upon arrival of the second laboratory visit in a fasted and hydrated state, a
11 standardised meal (30.0 g carbohydrate, 24.0 g protein, 8.0 g fat) was provided before
12 continuation with the testing session.

13 Participants were fitted with a three lead electrocardiogram (ECG), as described in
14 part 1 of the current series, and rested in the supine position for 15.0 minutes to minimize the
15 impact of orthostatic change¹³. Room temperature (22.0 °C) and light intensity (20.0 lm·ft²)
16 were kept constant throughout testing. Hydration status, represented as a percentage of total
17 body mass was determined using right wrist to right ankle bioelectrical impedance (BodyStat
18 1500, BodyStat, Douglas, UK).

19 Echo Doppler ultrasound (model AU5; Esaote, Genova, Italy) using a 7.50 MHz
20 broadband linear array transducer was used to perform vascular assessments (angle of
21 insonation: 60.0°, B gain: 75.0, Doppler gain: 49.0, CFM gain: 47.0, depth of penetration:
22 49.3 mm, depth of focus: 27.0 – 31.0). Live streamings were collected on a Hewlett-Pickard
23 computer running video capture software through an analogue to digital converter (Pinnacle,
24 Corel Inc., Ottawa, Canada) at 25.0 Hz. Left common carotid artery and right brachial artery
25 assessments were performed in the supine position whilst left popliteal artery assessments

1 were in the prone position. Baseline systemic peak blood velocity, intima-media thickness
2 (IMT), artery diameter, calculation of shear rate, and resistance index (RI, carotid artery only)
3 measures were collected over ten cardiac cycles for all three arteries (definitions provided in
4 Part 1 of this series). All measurements occurred within a 10 mm region of interest (ROI), 10
5 mm distal of the carotid bulb in the anterior longitudinal (AL) and posterior longitudinal (PL)
6 plane, 10 mm distal of the superior medial genicular bifurcation of the popliteal artery, and
7 65.0% of upper-arm length (acromion process to lateral radial head) distal of the
8 glenohumeral joint for the brachial artery¹³⁻¹⁷. These cardiovascular parameters were selected
9 due to the exploratory nature of this study in an attempt to distinguish any limb specific
10 associations between PB and cardiovascular parameters.

11 Offline analyses of diameter measures for all arteries was performed using Brachial
12 Analyzer (Medical Imaging Application LLC, Iowa, USA) and IMT measures of all arteries
13 was performed with Carotid Analyzer (Medical Imaging Application LLC, Iowa, USA). Data
14 was R-gated to ensure artery diameter and IMT were measured during the diastolic phase
15 only. Frame-to-frame measurements were filtered from final analysis if they did not use
16 70.0% of the ROI and/or were more than one standard deviation (SD) from the mean artery
17 diameter or IMT. All automated processes were assessed for error by one researcher. Intra-
18 day coefficients for variation (CV) ranged from 2.34% - 4.97% whilst inter-day CV ranged
19 from 1.57% - 5.33% for artery diameter. Intra-day coefficients for variation (CV) ranged
20 from 3.04% - 7.04% whilst inter-day CV ranged from 1.45% - 11.3% for IMT. Blood
21 velocity inter and intra-day CV was below 20.0% for all arteries. Shear rate inter and intra-
22 day CV was below 16.0% for all arteries. Carotid RI inter and intra-day CV was below
23 12.0%. All CV measures indicated that there was sufficient sensitivity to detect changes in
24 cardiovascular health based on observed changes in these variables following PB
25 interventions¹⁸⁻²⁰.

Statistical Analyses

1
2 SPSS version 22 (IBM, New York, USA) was used for statistical analyses. Pearson
3 correlation was used to assess multicollinearity between PB parameters and total PB, no
4 adjustment was made to the data if multicollinearity was present. Isotemporal substitution
5 regression modelling (forced entry) was implemented to examine the impact of one hour of
6 PB substitution⁴. Isotemporal substitution modelling is performed by removing one PB
7 (hereafter referred to as the substituted PB) from the regression model (i.e. substitute SB
8 model: Intercept + (β_1 x Standing) + (β_2 x LIPA) + (β_3 x sMVPA) + (β_4 x 10MVPA) + (β_5 x
9 Total PB) + Covariates + Error). Significant PB predictors within the isotemporal substitution
10 model illustrate that replacing one hour of the substituted PB (as data is measured in hrs·day⁻¹)
11 ¹) with the significant PB would have an effect on the respective cardiovascular parameter
12 (magnitude of unit change illustrated by beta coefficient and 95% CI[s]). Including Total PB
13 at the end of the isotemporal substitution model represents the time-constrained hours within
14 a waking hours day, which standard linear regression modelling does not account for.
15 Isotemporal substitution models were conducted without (Model 1) and with (Model 2)
16 adjustment for covariates to determine how hydration status and medication affect the
17 relationship between PB and cardiovascular profile. Hydration status was used as a covariate
18 as it has been shown to affect artery diameter²¹ whilst medication use was used as a covariate
19 as it has been shown to effect cardiovascular parameters²²⁻²⁴. Hydration, primary CVD meds,
20 and (in)direct CVD meds were used for covariate adjustment where preceding bivariate linear
21 regressions had shown that they were significantly associated with specific cardiovascular
22 parameters. Cardiovascular data were natural LOG transformed if they violated normal
23 distribution. Data are presented as beta coefficient (95% confidence interval [95%CI]) unless
24 otherwise stated.

Results

1 Isotemporal Substitution

2 Isotemporal substitution showed that changes in PB levels would significantly affect
3 three out of the 19 assessed cardiovascular parameters (Supplemental Material), these being
4 resting heart rate, carotid AL artery diameter, and popliteal artery diameter. The significant
5 substitutions are shown in figure 1.

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-Insert Figure 1 here-

9 **Figure 1 Significant physical behaviour isotemporal substitutions and their impact on**
10 **cardiovascular parameters. Markers indicate (left to right) -95%CI, beta coefficient,**
11 **and +95%CI. * Normalised for Primary CVD Medication. SB – sedentary behaviour,**
12 **LIPA – light intensity physical activity, sMVPA – sporadic moderate to vigorous**
13 **intensity physical activity (accumulated in bouts < 10 mins), 10MVPA – 10 minute**
14 **moderate to vigorous intensity physical activity (accumulated in bouts ≥ 10 mins),**
15 **Carotid AL artery diameter – carotid anterior longitudinal plane artery diameter.**

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18 Substitution of SB with Standing and sMVPA was suggested to reduce resting heart
19 rate (figure 1, -6.20 bpm [-12.1, -0.22], -3.72 bpm [-7.01, -0.44], respectively) which, is
20 clinically relevant as a 5 bpm increase in resting heart rate increases the risk of cardiovascular
21 mortality by 3% (2.0, 4.0%)²⁵. After the substitution of SB with LIPA, carotid AL artery
22 diameter was predicted to reduce (figure 1, -0.54 mm [-1.00, -0.07]) and vice versa (figure 1,
23 0.54 mm [0.08, 1.00]), which is clinically relevant as a 0.78 mm increase is associated with a
24 2.1 (1.3, 3.3) hazard ratio risk of all-cause mortality²⁶. Substitution of Standing with sMVPA
25 (figure 1, 1.31 mm [0.11, 2.51]) would increase popliteal artery diameter and vice versa

1 (figure 1, -1.52 mm [-2.83, -0.22]). This result is clinically relevant as an 8-week interval
2 training program increased popliteal artery diameter by 0.14 mm per hour of training²⁷ as
3 well as the popliteal artery diameter of healthy controls being 0.6 mm ($p = 0.11$) larger than
4 those with coronary artery disease (males aged 40 – 70 years)²⁸.

5 Within model 2, the results for all cardiovascular variables remained the same after
6 covariate adjustment suggesting that co-variables had no effect on the relationship between PB
7 and cardiovascular profile.

8 Multicollinearity

9 The largest correlation coefficient within the multicollinearity matrix was between SB
10 and LIPA, sMVPA (both $r = -0.69$) whilst the remaining variables only had weak correlations
11 suggesting low influence of collinearity on the results (table 2).

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Table 2 Collinearity statistics for PB parameters.

	SB	Standing	LIPA	sMVPA	10MVPA	Total PB
SB	-	-0.58***	-0.69***	-0.69***	-0.23*	0.32**
Standing		-	0.64***	0.35**	0.01	0.24*
LIPA			-	0.45***	-0.02	0.13
sMVPA				-	0.19	0.23*
10MVPA					-	0.05

Pearson Correlations.

*** $p \leq 0.001$, ** $p \leq 0.01$, * $p \leq 0.05$.

SB – sedentary behaviour, LIPA – light intensity physical activity, sMVPA – sporadic moderate to vigorous intensity physical activity (accumulated in bouts < 10 mins), 10MVPA – 10 minute moderate to vigorous intensity physical activity (accumulated in bouts \geq 10 mins), Total PB – total physical behaviour.

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Discussion

4 The objective of this study was to determine whether the substitution of SB and lower
5 intensity PA with MVPA would have positive effects on cardiovascular health and vice versa,
6 in older adults. The aim was to provide a time-constrained, alternative to
7 bivariate/multivariate regression modelling, to simulate how changes in PB would affect the
8 cardiovascular profile of older adults. It was hypothesised that substituting SB with any
9 intensity of PA would improve cardiovascular parameters and that substituting a PB with a
10 higher intensity would improve cardiovascular profile. It was also hypothesised that
11 substituting SB with 10MVPA would have a greater effect on cardiovascular parameters than
12 seen with sMVPA substitutions.

13 Heart rate is controlled by the central nervous system, which is compromised of the
14 sympathetic and parasympathetic pathways. The simulation of the replacement of SB with
15 Standing or sMVPA suggested it would reduce resting heart rate. Physiologically, this could

1 be achieved through improved baroreceptor function, which naturally declines with age²⁹.
2 Given that six weeks of yoga (consisting mainly of static postures [and breathing exercises])
3 has been reported to improve high frequency baroreceptor sensitivity, and to reduce resting
4 heart rate in older adults (whereas prolonged aerobic training did not)³⁰, a similar effect may
5 be at play in the Standing PB within our current modelling. High frequency baroreceptors
6 represent the sympathetic nervous system, suggesting that vasoconstriction response was
7 improved to counteract the natural fall in blood pressure with standing activities³¹.
8 Subsequently, increased vasoconstriction would increase venous return and stroke volume,
9 which would result in the need for a lower heart rate to maintain resting cardiac output. On
10 the other hand, the modelling of reduction in heart rate through increased sMVPA may be
11 achieved via improvements in the parasympathetic pathway. Interval training consisting of
12 nine, 5-minute repeated bouts at 65% of maximum heart rate (MVPA) over 14 weeks
13 improved markers of parasympathetic activity (PNN50 (percentage of successive normal
14 sinus RR intervals > 50.0 ms) and RMSSD (root mean square of the successive normal sinus
15 RR interval difference)) and subsequently decreased 24-hour mean heart rate within older
16 adults³². Therefore, the simulations from real data in our current study suggest that reducing
17 SB with PA, such as Standing (arguably easy to accumulate, due to limiting the common
18 socio-economic-volition barriers to structured exercise normally reported in older persons³³),
19 could yield health benefits. However, engagement in MVPA is also important, as it would
20 appear that different pathways are targeted by the two distinct PA intensities.

21 The reduction in resting heart may also be a result of vascular remodelling within
22 compliant blood vessels such as the carotid and popliteal arteries, but not the stiffer brachial
23 artery. With ageing, artery diameter increases as elastin stiffness decreases causing the load
24 bearing to shift to collagen fibres within the vascular smooth muscle³⁴. This structural change
25 may not be due solely to ageing but also due to increased SB, as the substitution of LIPA

1 with SB suggested it would increase carotid AL artery diameter in our modelling. The
2 opposite association was shown when the reverse substitution between SB and LIPA was
3 made. These inferences are in line with previous older adult research which found an increase
4 and decrease in carotid-femoral pulse wave velocity with increased engagement in LIPA and
5 SB, respectively³⁵.

6 The increase in arterial stiffness with ageing is also a determinant for the fall in
7 orthostatic blood pressure, which begins before baroreceptor mediated reflexes³⁶. Orthostatic
8 posture increases lower limb blood pressure, which subsequently leads to an increase in total
9 peripheral resistance and declined cardiac output. With the substitution of standing with
10 sMVPA, it was suggested that popliteal artery diameter would increase. This, in line with
11 Poiseuille's Law of flow, would decrease local blood pressure and thus total peripheral
12 resistance. However, sMVPA engagement would also acutely increase blood flow³⁷. Blood
13 flow declines with age in the legs due to increased sympathetic activity³⁸, the latter which
14 could increase total peripheral resistance. Training interventions within physically inactive
15 have shown that the acute vascular responses to interval training (MVPA bouts < 10 mins,
16 representative of sMVPA) stimulates baroreceptor activity³² and increases artery diameter²⁷,
17 subsequently leading to improved popliteal endothelial function and distensibility³⁹. Overall,
18 our results suggest a potential for older adults who cannot/choose not to sustain MVPA for 10
19 continuous minutes to still attain positive vascular adaptations (reduced resting heart rate and
20 increased popliteal artery diameter). This is relevant given the sample population averaged
21 less than one ₁₀MVPA bout per day (0.28 ± 0.71 n·day⁻¹) and only 34.2 ± 81.6 mins·week⁻¹ of
22 ₁₀MVPA, suggesting the majority of the study population could not/chose not to sustain
23 MVPA for 10 continuous minutes (see Part 1 of this series).

24 **Conclusion**

1 Our isothermal substitution modelling suggest that an accumulation of MVPA bouts
2 that are shorter than the recommended 10-minute minimum would improve cardiovascular
3 parameters (including resting heart rate and popliteal artery diameter), with lower intensity
4 PA also influencing cardiovascular parameters. Our findings are therefore a promising
5 avenue for lifestyle interventions in older adults in order to reduce the ageing effects on
6 cardiovascular health, especially those end-users who cannot engage or sustain sufficient
7 MVPA to be classed as physically active. The replacement of SB with PA influenced two of
8 the 19 (resting heart rate, and carotid AL artery diameter) whilst the replacement of sMVPA
9 with a lower intensity PB influenced one (popliteal artery diameter) cardiovascular
10 parameter(s). Our findings suggest that the reduction of SB is just as important as the need to
11 be physically active for older adults.

12 Finally, the current study illustrates the usefulness of isothermal substitution
13 modelling in simulating the different effects (and/or physiological pathways) that a PB
14 outcome of interest, may have on a unique (or a set of) cardiovascular parameter(s),
15 dependent on the PB it is displacing. This is the first study, to the authors' knowledge, to
16 demonstrate changes in cardiovascular phenotype within an isothermal substitution model
17 for an older adult cohort using objective measures of physical behaviour and cardiovascular
18 parameters.

19 Intervention studies are needed to determine the time course of the suggested
20 temporal changes shown in isothermal substitution modelling in older adult populations.

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