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Intervention and mediation effects of a community-based singing group on older adults' perceived physical and mental health: the Sing4Health randomized controlled trial

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ABSTRACT

Objective: To examine short- and long-term effects of a group singing program on older adults' perceived physical and mental health levels, and also investigate subjective well-being (life satisfaction, positive/negative affect and hedonic balance), body balance and serum biomarkers (C-reactive protein and erythrocyte sedimentation rate) as putative mediating mechanisms, controlling for the cognitive status of the participants. **Design:** The randomized controlled trial included 149 participants (60 to 95 years), allocated to an immediate intervention group (IG) or a wait-list active control group (WLG). The intervention comprised 34 sessions of group singing during 4-months. **Main outcome measures:** Self-report measures of physical health, anxiety, stress, and depression. Blinded assessments were conducted at baseline, post-intervention (4 months) and follow-up (6 months). **Results:** Participants in the IG reported a lower decline in perceived physical health after the intervention, as compared to the WLG. These benefits were maintained at follow-up. Singing-related changes in physical and mental health outcomes were mediated via an increase in positive affect. Moderation results showed that participants with very low cognitive functioning reported more anxiety and depression symptoms after the intervention. **Conclusions:** These findings provide further understanding on the psychological and physical mechanisms and effects of group singing in older adults.

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The last decades have been marked by a global phenomenon of major demographic changes in developed countries, characterized by rapid growth in the older adult population (Creech et al., 2013). Aging is accompanied by gradual but significant declines in biological and cognitive functions, with a profound impact on the physical and psychosocial functioning of older adults (Fu et al., 2018). Consequently, there is an increasing scientific interest devoted to the identification of influencing factors of the elderly's quality of life and the testing of evidence-based interventions that contribute to promoting optimal physical and mental health (Clift et al., 2018; Fu et al., 2018; Johnson et al., 2013). With this in mind, active participation in community-based singing groups received increased interest as a potentially effective intervention with physical and mental health benefits for older people (Clark & Harding, 2012; Daykin et al., 2018; Livesey et al., 2012; Reagon et al., 2016).

Singing is an active form of music participation based on the act of music-making through physical engagement (Kreutz et al., 2004; Reagon et al., 2016). More specifically, group singing is a polyphonic social activity that involves using the body to produce rhythmic and harmonious sounds in a synchronized way with other people (Livesey et al., 2012). Group singing is also cognitively challenging, involving remembering or learning songs, focusing on vocal control, watching the music sheet and the music conductor, and listening to the other singers during the performance, while producing an individual intonation that matches the frequency of the choir (Kreutz et al., 2004). The multimodal aspects of singing in a community group may also include a combination of meaningful opportunities for group work and socialization, positive reminiscing, emotional expression, relaxation and enjoyment (Clark & Harding, 2012; Johnson et al., 2013).

Several studies have identified diverse well-being and health-related benefits associated with choir singing in elderly populations, whereas others have presented inconclusive and mixed findings. Available systematic reviews of the literature indicate that amateur choir singing has a wide range of benefits for well-being and health in community healthy adults (e.g., Daykin et al., 2018), and as a therapeutic intervention concerning specific health problems (Clark & Harding, 2012; Clift et al., 2010; Reagon et al., 2016; Williams et al., 2018). Nevertheless, when compared to a control group or to other interventions, some reviews indicated that group singing participation does not provide additional benefits in some health domains (Clark & Harding, 2012; Clift et al., 2010; Reagon et al., 2016).

Emotional and mental health benefits associated with group singing

Findings from qualitative and cross-sectional survey studies suggest that regular participation in established choirs is linked to emotional and mental health benefits such as higher levels of happiness, enjoyment, sense of purpose, self-confidence, self-worth and self-control, as well as lower levels of depression, anxiety and isolation (Clift et al., 2010; Creech et al., 2013; Livesey et al., 2012). In addition, quasi-experimental and randomized controlled trials (RCT) using amateur chorists (Bullack et al., 2018; Kreutz et al., 2004) or individuals assigned to community singing groups (Cohen et al., 2006; Coulton et al., 2015; Pires et al., 2018) also reported positive effects of choir

singing for older people on measures of affect, morale, and mental health components of quality of life, anxiety, and depression.

On the other hand, some studies have also documented non-significant or contradictory effects of group singing interventions on emotional and mental health dimensions. Two quasi-experimental studies (pre and post-test design) demonstrated that group singing programs specifically developed for older people had no beneficial effect on depression and quality of life scores (Davidson et al., 2014; Fu et al., 2018). Moreover, when compared to an active music therapy intervention (including listening, improvising and dancing to music), the elderly nursing home residents assigned to a recreational group singing reported increased depressive symptoms, while the participants assigned to a music therapy group perceived significant reductions in depression (Werner et al., 2017).

With respect to specific effects of group singing on quality of life, one study with older adults from a community showed higher levels of psychological, social relationships and environmental quality of life, but not physical quality of life, even after adjusting for age and depressive symptoms (Johnson et al., 2013). However, when compared to matched older adults from the general population, amateur choir singers reported higher levels of physical quality of life, but not psychological quality of life (Johnson et al., 2017).

Physical and physiological benefits associated with group singing

Empirical research has documented the physiological health benefits of singing, namely cardiorespiratory, neuroendocrine, and immune systems functioning (Fancourt et al., 2014; Kang et al., 2018). Other studies suggested that amateur choir singing contributes to several physiological benefits, namely physical stress reduction, distraction from pain, more health-seeking behaviors, improved physical fitness, and better respiratory, cardiovascular, endocrine, and immune levels (Lally, 2009; Livesey et al., 2012). Regular participation in group singing may provide additional opportunities to engage in light to moderate physical activity, through commuting to the rehearsal places and back home, physical warm-ups and standing during the rehearsals (Clift et al., 2010; 2018), and contributing to enhance their physical fitness. Conversely, lower levels of physical activity or fitness (for example, balance impairment) are related to fear of falling in community-dwelling older adults (Sawa et al., 2020), and higher stress and anxiety symptoms, that may act as risk factors and actual falls in this population (Hallford et al., 2017). Other studies have also suggested that the physical effects of group singing on older adults' well-being may be mediated by the modulating effects of stress-related and inflammatory responses (e.g., Fancourt et al., 2014), although other studies have not mirrored these effects across all analyzed biomarkers (Bullack et al., 2018; Kreutz et al., 2004). Given these inconsistencies, a specific interest of this study was to simultaneously assess two broad-spectrum inflammatory biomarkers, namely the C-reactive protein (CRP) and the erythrocyte sedimentation rate (ESR), since previous studies have suggested that low-grade systemic elevated inflammation is associated with poorer self-rated health/mental health in older adults (Wium-Andersen et al., 2013).

Study aims and hypothesis

Despite the diverse health-promoting psychological, social and physical effects associated with group singing for older people documented in the scientific literature, there is a need for randomized controlled trials using non-established community choirs and the follow-up assessment of the group singing effects (e.g., Coulton et al., 2015; Pires et al., 2018), in order to better understand the long-term stability and accumulation of the intervention-related effects (Dingle et al., 2019). Moreover, little experimental research has focused on the potential mechanisms (or mediators of change) underlying the relationship between choir singing and physical or mental health-related outcomes. To the best of our knowledge, the few available studies used a quasi-experimental design with a comparison group to determine possible social mediators of group singing-related improvements in health and well-being (e.g., Pearce et al., 2016) in a wide age range of adult samples. As such, there is a need for further research and a better understanding of the possible mechanisms and linkages among psychological and biological processes associated with group singing benefits (Gick, 2011), especially in older adults.

Therefore, the primary aim of the current study is to examine the short- and long-term effects (at follow-up) of a 4-month community-based singing program on older adults' perceived physical health, anxiety, stress, and depression. A secondary aim of the present study is to investigate the unique and combined mediation effects of psychological (life satisfaction, positive and negative affect, and hedonic balance) and biological/physical (body balance and serum biomarkers: CRP and ESR) variables on choir-singing intervention changes in older adults' perceived physical and mental health.

Based on previous findings and theoretical considerations (e.g., Dingle et al., 2019; Kang et al., 2018; Williams et al., 2018), it was hypothesized that group singing effects on older adults' perceived physical and mental health, compared to a control (waiting list) condition, would be significantly mediated by one or more of the above putative mediator variables. More precisely, different mediating mechanisms were tested, hypothesizing that positive changes in group singing outcomes in older adults are a consequence of enhanced life satisfaction, mood/affect regulation, body balance, and physiological biomarkers (see the hypothesized parallel multiple mediation model in [Figure S1](#) of the supplemental appendix). Taking into consideration the substantial inter- and intra-individual variability in cognitive abilities among elderly individuals (Institute of Medicine, 2015) and how this may influence the perception of the intervention's benefits, both analyses of variance and mediation were also performed, in order to control for the potential moderating effect of cognitive status on the choir singing-outcomes relationships.

Materials and methods

Study design and setting

A two-center, pragmatic, randomized controlled trial (RCT) with two-arms [immediate intervention (IG) and wait-list active control groups (WLG)] and blinded outcome assessment was conducted to assess the effectiveness of the choir singing intervention in older adults. Data was collected at baseline (T0), four months after the baseline assessment (T1) and again at the six-month follow-up (T2). Between T0 and T1,

individuals in the IG participated in the choir singing program, whereas the individuals in the WLG were instructed to maintain their daily/usual social and leisure activities (i.e., active control group). After the second assessment (T1), individuals in the WLG initiated the intervention arm and participated in the choir singing program.

Study participants were recruited from two sites (Lisbon and Almada) of the Santa Casa da Misericórdia (SCM) in Portugal. The SCM is a private, non-profit, centenary institution of public interest that offers various charitable and social support services to senior citizens and whose primary mission is to improve the quality of life of the less privileged.

The study protocol was reviewed and approved by the first author's university research ethics board (ref. n.º 12-09-2018). All procedures performed in this study were in accordance with the ethical guidelines and standards of the research ethics committee, as well as with the European Code of Conduct for Research Integrity and the World Medical Association Declaration of Helsinki.

This study was registered in the ClinicalTrials.gov database (NCT03985917), as part of a larger research project aiming to investigate the psychosocial, cognitive, and health-related benefits of a singing group program for older adults. The current study focuses specifically on potential psychological and physical/biological mechanisms associated with group singing benefits.

Participants

An a priori power analysis (GPower, version 3.1.9) determined that a target sample size of 140 individuals was required to power the study at 90%, with an alpha level of .05 and an estimated large effect size ($\eta_p^2 = 0.14$) for repeated measures analysis of variance. A dropout of 15 participants per group (total of 30 individuals) was estimated to decrease the statistical power of the study to 83%.

Between October 2018 and January 2019, eligible older adults were invited to participate in this research program. To be eligible for inclusion, individuals had to be 60 years of age or older, retired, accepting the invitation to enroll in the study and not having participated in any type of intervention programs in the previous four months. Exclusion criteria included having a severe diagnosed impairment (e.g., auditory, visual or mobility-related) that could impede the participation in the choir-singing activities.

Of the 1624 individuals who were screened for eligibility, 1371 (84.4%) were found to be ineligible and 104 (6.4%) decline to participate for different reasons. The remaining 149 eligible older adults who accepted to participate in the research were randomly assigned to the IG ($n=75$) or the WLG ($n=74$). However, in the first week of the intervention, 12 participants of the IG dropped out of the study without attending any session. In order to maintain the statistical power of this RCT study, the research team decided to randomly select 14 participants from the WLG and allocate them into the IG. These 14 participants started their participation on the second week of the intervention, missing the two initial sessions. After this, the sample distribution included 89 participants in the IG ($n=45$ in Lisbon and $n=44$ in Almada) and 60 in the WLG ($n=30$ in Lisbon and $n=30$ in Almada). The CONSORT flowchart of the participants and the study design is presented in [Figure S2](#) (of the supplemental appendix).

No significant differences were found at baseline between the IG and the WLG concerning the sociodemographic and the cognitive status variables (see [Table S1](#) in the supplemental appendix). The majority of participants were female, aged above 70 years, widowed or lived with their partner (married/cohabitation), and had completed only a few years of formal schooling. A total of 77.9% of all participants (83.1% in the IG and 70.0% in the WLG) had one or more diagnosed medical conditions. No significant interaction (group conditions \times institution sites) or main (institution sites - Lisbon and Almada) effects were observed regarding the sociodemographic and outcome variables (data available upon request). Altogether, results indicate that the sample is characterized by low levels of formal education and household monthly income, as well as low scores on cognitive function as measured by the MoCA.

Measures

Sociodemographic data was provided at T0 by the staff of the SCM institution and included gender, age, education level (years of educational attainment), marital status, socioeconomic status (monthly household income), and diagnosed medical conditions. This data was missing on marital status (IG, $n=6$; WLG, $n=1$), education level (IG, $n=5$; WLG, $n=1$) and socioeconomic index (IG, $n=4$; WLG, $n=1$).

Psychoemotional and cognitive variables were assessed by questionnaires and administered by trained research assistants using Portuguese-language validated versions of the instruments [see the study protocol (Galinha, Farinha, et al., 2020) for more information]. The body balance was assessed by trained research assistants, whereas the physiological biomarkers were collected by certified laboratory technicians. All measures were completed or collected, in-person, by independent researchers and certified laboratory technicians blinded to group allocation, at the Social Care Institution facilities. The blood samples were analyzed by a certified laboratory. Data collection took place for 10 weekdays, from 10 to 15 participants per day. It started at 8 a.m., while participants were fasting, for: i) biomarkers, respiratory function, and balance data collection, followed by a buffet breakfast; ii) cognitive assessment; and, after 15 minutes break, ii) psychosocial measurement questionnaires and interviews. Every step of the data collection was performed by different researchers.

Outcome variables

Perceived physical health. The self-rated physical health dimension was measured using the physical health component of the World Health Organization Quality of Life: Brief Version (WHOQOL-BREF) instrument (WHO, 1996). Omega values indicated good internal consistency values for this scale, at all assessment points ($T0 = .77$, $T1 = .81$, and $T2 = .77$).

Depression, anxiety and stress. The mental health variables were assessed using the 21-items Depression, Anxiety, and Stress Scales (DASS-21; Lovibond & Lovibond, 1995), which is a short version of the original DASS scales comprising three subscales (depression, anxiety and stress), each with 7-items. Participants were asked to rate the frequency of experienced negative emotions, over the last four

months, on a four-point response scale. In this study, the scales showed good internal consistencies (omega coefficients) at all assessment points: depression ($T0 = .84$, $T1 = .87$, and $T2 = .83$), anxiety ($T0 = .79$, $T1 = .84$, and $T2 = .78$), and stress ($T0 = .82$, $T1 = .88$, and $T2 = .86$).

Mediating variables

Life satisfaction. The Satisfaction with Life Scale (SWLS; Diener et al., 1985) was used to assess life satisfaction as a global cognitive appraisal of one's life. This scale includes 5 items which were answered using a five-point Likert scale. The SWLS showed good reliability, in terms of omega coefficients, at all assessment time points ($T0 = .84$, $T1 = .88$, and $T2 = .84$).

Positive and negative affect. The Positive Affect and Negative Affect Schedule (PANAS; Watson et al., 1988) was used to assess the positive and negative affect, as well as the balance between the two dimensions. This scale comprises a total of 10 adjective words for positive and 10 words for negative affect. Participants were asked to indicate how they generally felt, over the last four months, on a five-point Likert scale. Preliminary analyses (correlation and principal components) indicated that the item "excited" showed higher inter-item correlations and factor loadings on the negative affect dimensions at all assessment points, irrespective of the participants group. Thus, this item was removed from further analysis. The PANAS' subscales showed good internal consistencies (omega coefficients) at all assessment points, namely: positive affect ($T0 = .80$, $T1 = .76$, and $T2 = .85$) and negative affect ($T0 = .85$, $T1 = .85$, and $T2 = .88$). The affect balance score was calculated by subtracting the negative affect scores from the positive affect scores (PANAS' subscales), with total scores range from -4 to 4 and with higher scores representing higher levels of overall affective well-being.

Body balance. The timed unipedal stance test was performed to assess the static balance. Participants were instructed to stand barefoot on their preferred leg, with their eyes open, keeping their legs from touching and maintaining an unipedal stance for up to 20seconds. The duration of the test performed correctly was measured using a stopwatch chronometer and the best score of two trials was registered and used for analysis (Springer et al., 2007).

Serum biomarkers. The C-reactive protein and erythrocyte sedimentation rate tests are commonly used laboratory tests to monitor and identify inflammatory diseases. The C-reactive protein concentrations were determined using the latex-enhanced immunoturbidimetric method. The erythrocyte sedimentation rate levels were determined in glass capillaries by microphotometry.

In this study, blood samples were collected early in the morning after an overnight fasting period. The collection, handling, and processing of these serum samples were performed by certified technicians of a licensed laboratory, using standard operating and quality control procedures.

Moderator variable

Cognitive function. The Montreal Cognitive Assessment (MoCA; Nasreddine et al., 2005; Simões et al., 2008) is a brief, single-page screening instrument, administered to assess the cognitive status of the participants. In the present study, only the MoCA total scores were used, with possible scores varying from 0 (worst performance) to 30 (best performance). Omega values indicated good internal consistency values for this scale, at all assessment points ($T0 = .83$, $T1 = .81$, and $T2 = .79$).

Procedures

Detailed information regarding the study procedures is available in the published protocol (see Galinha, Farinha, et al., 2020), and is briefly summarized here.

After obtaining the SCM institution's written agreement, eligible individuals were approached and invited by the institution to participate in the research. Interested individuals were informed about the study's terms and conditions and provided signed informed consent. A briefing session was provided by the intervention and the research teams to inform eligible older adults about the intervention program and research study participation. Eligible participants were then randomly assigned, with a 1:1 (IG: WLG) allocation ratio and blocking within sites (Lisbon and Almada), using the randomization function in IBM SPSS Statistics software.

The group singing program (Sing4Health) was a 4-months intervention consisting of twice-weekly, 34 sessions of 2 hours of duration, and a final public performance show. Sessions were conducted by a team of professional choir directors, one of which is a professional singer/artist and a public figure. Sessions took place in local theatres, intending to promote the mobility of the older adults and a greater contact with the local community. The group singing program included several components, namely: relation exercises and vocal warm-up; vocal technique exercises; rehearsals of the repertoire; breaks of nearly 20 minutes for interaction and socialization purposes; and, creation and presentation of a performance show. However, due to the intervention team limitations it was not possible to fully implement the group dynamics' exercises as initially planned. The singing repertoire included Portuguese traditional songs (folk and *fado*) suggested and selected by the intervention team and the participants, after discussion.

Statistical analyses

A preliminary inspection of the dataset was conducted to screen the data for accuracy, normality of the distributions and missing values.

All variables, at all assessment points, showed reasonable univariate normality (i.e., skewness and kurtosis values between ± 2), except for the CRP values. A square root transformation was applied, only resulting in a normal distribution for $T0$ CRP values. With respect to CRP values, two (IG, $n=1$; WLG, $n=1$) severe outliers (i.e., CRP values > 6.0) at $T1$ and one (WLG, $n=1$) at $T2$ were replaced by the winsorizing procedure. A square root transformation resulted in acceptable distributions of $T1$ and $T2$ CRP data.

The missing values analysis indicated 0.3% of missing data at T0, 29.5% at T1, and 44.3% at T2. However, results of the Little's missing completely at random (MCAR) test showed that missing values appeared to be at least missing at random at T1, $\chi^2(281) = 288.46$, $p = .367$, and at T2, $\chi^2(204) = 211.94$, $p = .337$. Hence, missing values at all assessment points were imputed using the expectation-maximization algorithm (Dempster et al., 1977), using all available data at each assessment time point.

The internal consistency (reliability) of the composite (latent) variables were evaluated using McDonald's omega coefficients, which were estimated using the free software JASP, version 0.11.1 (JASP Team, 2019, Amsterdam, The Netherlands).

Descriptive statistics included mean (M), standard deviation (SD), mean score difference (ΔM) between time points (ex. $M_{T0} - M_{T1}$) and respective 95% confidence intervals (CI), frequency and percentage (%).

One-way analysis of variance (ANOVA) and Chi-Square (χ^2) tests were used to compare group differences (IG vs. WLG) at baseline. In order to investigate the effects of the group singing intervention on the outcomes, a series of separate two-way (groups \times time points) mixed-design ANOVAs were performed. A second series of two-way ANOVAs were performed in order to examine the moderating effect of cognitive status (as a continuous variable) on the previous interaction effects. When a significant moderation was identified, the conditional effect of group singing intervention on the outcome variables, as a function of the moderator (continuous variable), was probed using the Johnson-Neyman (J-N) technique (Hayes, 2018). Dependent t -tests were used to estimate the follow-up effects only in the IG.

Effect sizes after the first intervention (T1) and at follow-up (T2) were estimated using omega squared (ω^2) for the ANOVAs and Cohen's d_z for the dependent t -tests, and interpreted according to Cohen's (1988) suggestion: small ($\omega^2 \geq .01$ and $d_z \geq 0.2$), medium ($\omega^2 \geq .06$ and $d_z \geq 0.5$) and large ($\omega^2 \geq .14$ and $d_z \geq 0.8$).

Four parallel multiple mediation models (one for each outcome) were tested to examine the direct effects of the group singing intervention on the outcomes (physical and mental health indicators), the indirect effects through several mediators and the related moderated mediation effects. Based on modern mediation analysis (Hayes, 2009), indirect effects were further tested, even in the absence of statistically significant total and/or direct effects, since the indirect effects tend to be estimated with higher relative power than the total and the direct effects (Kenny & Judd, 2014). With this in mind, models 4 (mediation only) and 59 (moderated mediation) of the PROCESS macro, version 3.4 (Hayes, 2018) for the SPSS, were used to compute and estimate coefficients for the direct, indirect and conditional effects, as well as, to provide bootstrapped CI for the indirect effects. The estimates of the 95% percentile CI of the indirect effects were derived from 10000 bootstrap samples. The indirect effect was considered significant when zero was not included in the 95% CI (Hayes, 2018). The percentile bootstrap method was selected given that this procedure shows a good balance between type I errors and statistical power (Yzerbyt et al., 2018, p. 941).

All analyses, except reliability and effect sizes (JASP), were conducted using the IBM SPSS Statistics for Windows, version 23 (IBM Corp., Armonk, N.Y., USA). The level of statistical significance was set at $p \leq .05$.

Results

Participants flow and randomization

The recruitment process and participants' flow throughout the study is presented in Figure S2. As presented in Table S1 (in the supplementary appendix), homogeneity between IG and WLG was guaranteed at baseline for the participants' demographic characteristics and the outcome variables, all $F_s(1, 147) < 1.33$, $p_s > .25$ (see Table 1 for the descriptive statistics).

Of the participants allocated to the IG, 12 dropped out before S1 starting the study, 23 participants were considered non-compliers (i.e., attended less than 75% of the sessions), and the remaining 54 participants were compliers. With respect to the second group (WLG) singing intervention (T1 to T2), 33 older adults agreed to participate, with 10 attending less than 75% of the sessions and 23 participating in 75% or more sessions.

The main reasons for the participants dropping out included: health issues (personal and/or family illnesses, scheduled treatments or operations, recovery from operations, etc.); participants had enrolled with friends or spouses that were allocated to a different group; and, scheduling incompatibility with the group singing sessions. Moreover, during the six months waiting list period, participants from the WLG enrolled in other activities that included singing and dancing in group (popular marches), which led to the withdrawal of this intervention.

The attrition rate for the IG was 13.4% at T1 and 34.8% at T2, whereas for the WLG it was 20.0% at T1 and 50.0% at T2.

In order to reduce the potential bias related to stemming from missing data, the following analyses were performed according to the intention-to-treat (ITT) principle, which includes all randomized individuals (IG, $n=89$; WLG, $n=60$). In addition, per-protocol (PP) analyses were also performed, including the IG compliers ($n=54$) and the WLG individuals assessed at T1 ($n=48$), or the WLG compliers of the second group singing intervention ($n=23$). Since the results of the ITT and PP analyses were very similar, the results presented next are based on the ITT analysis, whereas the PP results are available in an online supplemental appendix (see Tables S2 to S6).

Table 1. Descriptive statistics, effect sizes and 2x2 ANOVA results on the outcomes between T0 and T1 (ITT, $N=149$).

		Baseline (T0)	Post-intervention (T1)	Within-group		Group x time interaction		Time effect	
Variables	Condition	<i>M</i> (<i>SD</i>)	<i>M</i> (<i>SD</i>)	<i>p</i> ^a	<i>Dz</i>	<i>F</i> (<i>p</i>)	ω ²	<i>F</i> (<i>p</i>)	ω ²
<i>Outcomes</i>									
Physical health	IG (<i>n</i> = 89)	3.62 (0.73)	3.48 (0.66)	< .001	0.32	4.07	.01	30.88	.02
	WLG (<i>n</i> = 60)	3.76 (0.67)	3.46 (0.66)	< .001	.058	(.045)		(< .001)	
Anxiety	IG (<i>n</i> = 89)	12.37 (3.86)	12.87 (4.73)	.328	−0.10	1.20	.00	0.06	.00
	WLG (<i>n</i> = 60)	12.97 (4.75)	12.65 (4.44)	.536	0.08	(.276)		(.811)	
Stress	IG (<i>n</i> = 89)	11.38 (3.80)	12.12 (4.58)	.049	−0.21	0.46	.00	3.57	.00
	WLG (<i>n</i> = 60)	11.75 (4.25)	12.10 (4.02)	.427	−0.10	(.498)		(.061)	
Depression	IG (<i>n</i> = 89)	11.62 (4.36)	11.43 (4.70)	.638	0.05	0.00	.00	0.25	.00
	WLG (<i>n</i> = 60)	11.58 (4.73)	11.44 (3.84)	.795	0.03	(.950)		(.618)	

^a*p* value for the paired *t*-test.

Intervention effects on perceived physical health and mental health outcomes

Results of the first series of the two-way ANOVAs (see Table 1) showed a significant group \times time interaction for perceived physical health levels ($p = .045$, $\omega^2 = .01$, observed power = .52), with WLG participants showing a greater decline ($p < .001$, $dz = 0.58$) than IG participants ($p < .001$, $dz = 0.32$). No significant group \times time interactions were found for anxiety, stress, and depression ($ps > .27$). When controlling for the potential moderating effect of cognitive status on the choir singing-outcomes relationships, results of a second series of two-way ANOVAs (see Table 2) showed that the MoCA total scores did not moderate the group \times time interaction for perceived physical health and stress, but significantly moderated the group \times time interactions for anxiety ($p = .039$, $\omega^2 = .01$, observed power = .55) and depression ($p = .020$, $\omega^2 = .01$, observed power = .65). The J-N technique was used to identify the cognitive function levels where these conditional effects were significant (see Figures S3 and S4). Results showed that the intervention had a detrimental effect on anxiety and depression levels, but only among IG participants with very low cognitive functioning (MoCA total scores of ≤ 12.97 and of ≤ 7.92 , respectively).

Follow-up effects on perceived physical health and mental health outcomes

Given that only the IG participants were assessed at 6-months after the intervention, the follow-up effects were examined using the IG within-subjects' comparisons from T1 to T2. Results show that all outcome variables at T1 remained unchanged at follow-up (see Table 3), except anxiety ($p < .001$, $dz = 0.49$) that decreased from the post-intervention to the follow-up period.

Table 2. Moderation effects of cognitive function levels (as measured by the MoCA) on the group singing-outcomes between T0 and T1 (ITT, N=149).

Outcomes	Effects	<i>F</i>	<i>p</i>	ω^2
Physical health	Time x cognitive function	0.77	.382	.00
	Group x cognitive function	0.06	.808	.00
	Time x group x cognitive function	0.04	.838	.00
Anxiety	Time x cognitive function	2.70	.103	.00
	Group x cognitive function	0.08	.783	.00
	Time x group x cognitive function	4.36	.039	.01
Stress	Time x cognitive function	3.84	.052	.00
	Group x cognitive function	0.08	.780	.00
	Time x group x cognitive function	0.93	.336	.00
Depression	Time x cognitive function	0.20	.656	.00
	Group x cognitive function	0.04	.833	.00
	Time x group x cognitive function	5.54	.020	.01

Table 3. Follow-up effects for the outcomes in the IG (ITT, N=89).

Outcomes	Follow-up (T1 to T2)		
	ΔM (95% CI)	<i>p</i> ^a	<i>dz</i>
Physical health	−0.07 (−0.18, 0.05)	.239	−0.13
Anxiety	2.05 (1.17, 2.92)	< .001	0.49
Stress	−0.27 (−1.15, 0.60)	.535	−0.07
Depression	0.39 (−0.47, 1.24)	.374	0.10

Note: ^a*p* value for the paired *t*-test.

Mediation effects of group singing intervention on perceived physical and mental health outcomes

The direct and indirect effects of the group singing intervention on the physical and mental health outcomes, after controlling for baseline values, are presented in Table 4. Separate parallel multiple mediation analyses were performed for affect balance, after removing positive and negative affect variables from these models, and the pattern of results maintained between models.

No significant direct effects (c' -paths) were observed between the group singing intervention and the outcomes. The mediation results showed significant effects (a -paths) of group singing intervention on the mediating variables positive affect and affect balance, indicating that these variables increased in the IG participants significantly more than in the WLG participants. In addition, several significant effects (b -paths) were observed linking the subjective well-being variables (life satisfaction and positive/negative affect) and the outcomes, although not as a result of the intervention. More precisely, perceived physical health was significantly predicted by all subjective well-being variables, anxiety and stress were predicted by negative affect and affect balance, and depression was significantly predicted by all affective dimensions. Furthermore, higher body balance predicted lower scores of anxiety and stress, whereas higher CRP levels predicted higher stress scores. Significant indirect effects (ab -paths) between group singing intervention and the outcomes were only observed via positive affect. More precisely, the group singing intervention had significant indirect effects on perceived physical health, anxiety, stress, and depression levels, through preserving a better hedonic (affect) balance, as well as on perceived physical health and depression levels through higher positive affect scores. The four tested models explained 64% to 74% of the variance of the outcome variables, namely: perceived physical health, $R^2 = .74$, $F(14, 134) = 27.34$, $p < .001$; anxiety, $R^2 = .64$, $F(14, 134) = 16.86$, $p < .001$; stress, $R^2 = .73$, $F(14, 134) = 25.22$, $p < .001$; and, depression, $R^2 = .68$, $F(14, 134) = 20.38$, $p < .001$.

Moderated mediation effects of group singing intervention on perceived physical and mental health outcomes

Conditional process results showed no significant interaction effects of cognitive status on any of the a -, b - and ab -path relationships. Moreover, the other significant effects observed in the parallel multiple mediation analyses were unmoderated on levels of cognitive status. However, when considered the analyses of the direct (c' -paths) effects, results showed that the elderly's cognitive status moderated the relationship between group singing and depression levels, $c' = -0.183$, $SE = 0.078$, $p = .020$, 95% CI $[-0.338, -0.029]$. Using the J-N technique, results indicated that the intervention had a detrimental effect on depression scores, but only among those with very low cognitive functioning levels (MoCA total scores ≤ 12.68).

Discussion

The main objectives of this study were to examine the intervention effects of a non-established community-based singing program on older adults' perceived physical

Table 4. Direct and indirect effects of the group singing intervention on the outcome variables (ITT, N = 149).

Antecedent	→	Consequent	Path	SC	UC	SE	p	95% CI
<i>Direct effects (X → Y)</i>								
Intervention	→	Physical health	c'1	0.015	0.010	0.062	.871	−0.112 to 0.132
Intervention	→	Anxiety	c'2	0.075	0.345	0.512	.501	−0.667 to 1.357
Intervention	→	Stress	c'3	0.059	0.256	0.420	.593	−0.575 to 1.087
Intervention	→	Depression	c'4	0.080	0.348	0.451	.442	−0.544 to 1.240
<i>Direct effects (X → M)</i>								
Intervention	→	Life satisfaction	a1	0.212	0.811	0.529	.127	−0.235 to 1.857
Intervention	→	Positive affect	a2	0.381	0.275	0.093	.004	0.087 to 0.463
Intervention	→	Negative affect	a3	−0.025	−0.019	0.102	.855	−0.220 to 0.183
Intervention	→	Affect balance	a4	0.294	0.296	0.128	.022	0.043 to 0.549
Intervention	→	Body balance	a5	−0.062	−0.427	0.803	.596	−2.015 to 1.161
Intervention	→	CRP	a6	−0.192	−0.062	0.051	.231	−0.163 to 0.040
Intervention	→	ESR	a7	0.092	1.606	2.557	.531	−3.450 to 6.662
<i>Direct effects (M → Y)</i>								
Life satisfaction	→	Physical health	b1	0.202	0.034	0.010	.001	0.014 to 0.055
Life satisfaction	→	Anxiety	b2	−0.034	−0.041	0.085	.630	−0.210 to 0.128
Life satisfaction	→	Stress	b3	−0.039	−0.044	0.070	.530	−0.184 to 0.095
Life satisfaction	→	Depression	b4	−0.112	−0.127	0.076	.096	−0.278 to 0.023
Positive affect	→	Physical health	b5	0.207	0.188	0.054	.001	0.081 to 0.295
Positive affect	→	Anxiety	b6	−0.024	−0.153	0.444	.732	−1.032 to 0.726
Positive affect	→	Stress	b7	0.007	0.040	0.366	.912	−0.683 to 0.764
Positive affect	→	Depression	b8	−0.165	−0.999	0.395	.013	−1.781 to −0.217
Negative affect	→	Physical health	b9	−0.170	−0.150	0.055	.007	−0.258 to −0.041
Negative affect	→	Anxiety	b10	0.634	3.914	0.456	< .001	3.012 to 4.816
Negative affect	→	Stress	b11	0.508	2.966	0.377	< .001	2.220 to 3.712
Negative affect	→	Depression	b12	0.468	2.740	0.406	< .001	1.936 to 3.543
Affect balance	→	Physical health	b13	0.277	0.180	0.041	< .001	0.099 to 0.262
Affect balance	→	Anxiety	b14	−0.414	−1.889	0.390	< .001	−2.662 to −1.117
Affect balance	→	Stress	b15	−0.323	−1.394	0.316	< .001	−2.019 to −0.769
Affect balance	→	Depression	b16	−0.442	−1.913	0.315	< .001	−2.535 to −1.291
Body balance	→	Physical health	b17	−0.013	−0.001	0.006	.845	−0.014 to 0.011
Body balance	→	Anxiety	b18	−0.182	−0.122	0.052	.020	−0.225 to −0.020
Body balance	→	Stress	b19	−0.142	−0.090	0.043	.037	−0.175 to −0.005
Body balance	→	Depression	b20	−0.136	−0.087	0.046	.064	−0.178 to 0.005
CRP	→	Physical health	b21	−0.031	−0.064	0.109	.560	−0.279 to 0.152
CRP	→	Anxiety	b22	0.074	1.059	0.905	.244	−0.732 to 2.850
CRP	→	Stress	b23	0.120	1.630	0.758	.033	0.131 to 3.129
CRP	→	Depression	b24	0.003	0.041	0.807	.960	−1.555 to 1.636
ESR	→	Physical health	b25	−0.068	−0.003	0.002	.248	−0.007 to 0.002
ESR	→	Anxiety	b26	−0.015	−0.004	0.018	.829	−0.040 to 0.032
ESR	→	Stress	b27	0.023	0.006	0.015	.697	−0.024 to 0.036
ESR	→	Depression	b28	−0.061	−0.015	0.016	.351	−0.047 to 0.017
<i>Indirect effects through life satisfaction (X → M → Y) ^a</i>								
Intervention	→	M → Physical health	ab	0.043	0.028	0.022	—	−0.007 to 0.078
Intervention	→	M → Anxiety	ab	−0.006	−0.027	0.091	—	−0.256 to 0.120
Intervention	→	M → Stress	ab	−0.007	−0.032	0.071	—	−0.189 to 0.111
Intervention	→	M → Depression	ab	−0.021	−0.090	0.093	—	−0.303 to 0.070
<i>Indirect effects through positive affect (X → M → Y) ^a</i>								
Intervention	→	M → Physical health	ab	0.079	0.052	0.021	—	0.015 to 0.097
Intervention	→	M → Anxiety	ab	−0.009	−0.041	0.126	—	−0.266 to 0.254
Intervention	→	M → Stress	ab	0.002	0.010	0.112	—	−0.177 to 0.283
Intervention	→	M → Depression	ab	−0.058	−0.255	0.135	—	−0.555 to −0.034
<i>Indirect effects through negative affect (X → M → Y) ^a</i>								
Intervention	→	M → Physical health	ab	0.004	0.003	0.016	—	−0.031 to 0.036
Intervention	→	M → Anxiety	ab	0.003	0.014	0.412	—	−0.782 to 0.836
Intervention	→	M → Stress	ab	−0.003	−0.014	0.316	—	−0.684 to 0.563
Intervention	→	M → Depression	ab	−0.011	−0.047	0.289	—	−0.678 to 0.463
<i>Indirect effects through affect balance (X → M → Y) ^a</i>								
Intervention	→	M → Physical health	ab	0.081	0.053	0.025	—	0.009 to 0.106
Intervention	→	M → Anxiety	ab	−0.114	−0.523	0.272	—	−1.100 to −0.043
Intervention	→	M → Stress	ab	−0.087	−0.378	0.215	—	−0.850 to −0.017

(Continued)

Table 4. Continued.

Antecedent	→	Consequent	Path	SC	UC	SE	<i>p</i>	95% CI
Intervention →	<i>M</i>	→ Depression	<i>ab</i>	−0.121	−0.529	0.276	—	−1.128 to −0.056
<i>Indirect effects through body balance (X → M → Y) ^a</i>								
Intervention →	<i>M</i>	→ Physical health	<i>ab</i>	0.001	0.001	0.007	—	−0.013 to 0.017
Intervention →	<i>M</i>	→ Anxiety	<i>ab</i>	0.016	0.073	0.119	—	−0.152 to 0.342
Intervention →	<i>M</i>	→ Stress	<i>ab</i>	0.014	0.062	0.093	—	−0.091 to 0.285
Intervention →	<i>M</i>	→ Depression	<i>ab</i>	0.013	0.056	0.086	—	−0.093 to 0.255
<i>Indirect effects through CRP (X → M → Y) ^a</i>								
Intervention →	<i>M</i>	→ Physical health	<i>ab</i>	0.006	0.004	0.009	—	−0.015 to 0.025
Intervention →	<i>M</i>	→ Anxiety	<i>ab</i>	−0.015	−0.067	0.103	—	−0.295 to 0.134
Intervention →	<i>M</i>	→ Stress	<i>ab</i>	−0.025	−0.108	0.104	—	−0.345 to 0.066
Intervention →	<i>M</i>	→ Depression	<i>ab</i>	−0.001	−0.002	0.067	—	−0.141 to 0.151
<i>Indirect effects through ESR (X → M → Y) ^a</i>								
Intervention →	<i>M</i>	→ Physical health	<i>ab</i>	−0.006	−0.004	0.008	—	−0.023 to 0.012
Intervention →	<i>M</i>	→ Anxiety	<i>ab</i>	−0.001	−0.006	0.055	—	−0.126 to 0.110
Intervention →	<i>M</i>	→ Stress	<i>ab</i>	0.002	0.010	0.042	—	−0.078 to 0.107
Intervention →	<i>M</i>	→ Depression	<i>ab</i>	−0.006	−0.026	0.059	—	−0.135 to 0.116

Note: ^a partially standardized effects, percentile estimates and 95%CI values based on 10000 bootstrap samples; SC = standardized regression coefficient; UC = unstandardized regression coefficient; SE = standard error of the unstandardized regression coefficient; CRP = C-reactive protein; ESR = erythrocyte sedimentation rate.

health, anxiety, stress and depression levels, and also to investigate whether subjective well-being (life satisfaction, positive/negative affect and hedonic balance) and/or physical health variables (body balance, CRP and ESR) mediated the choir singing-related changes in the outcome measures. The invited older adults that participated in the current study were mostly women, aged above 70 years, with lower education levels and with one or more diagnosed diseases.

Group singing intervention effects

With regards to the intervention effects on the physical and mental health outcomes, a key finding of the current study shows that the group singing program contributed to mitigate the declines in perceived physical health (in comparison to the control group), but not to decrease anxiety, stress and depression. This evidence supports previous research on the perceived physical health benefits of group singing (Cohen et al., 2006, 2007; Johnson et al., 2017; Lally, 2009; Livesey et al., 2012; Pearce et al., 2016) and corroborates previous studies that did not report benefits in anxiety, stress and/or depression levels related with group singing activity (Davidson et al., 2014; Werner et al., 2017). On the other hand, these results do not concur with other findings that documented better mental health (e.g. increased psychological well-being and lower levels of distress, anxiety and depression) due to group singing (Clift et al., 2010; Reagon et al., 2016; Williams et al., 2018), including those of a RCT study (Coulton et al., 2015) that indicated lower scores of anxiety and depression, but no significant effects on the perceived physical health, after 3 months of choral singing participation. Possible explanations for these differences include different study designs, intervention programs, assessment instruments and sample characteristics.

The high retention rate (86.6%) and the percentage of participants who attended to 75% or more sessions of the first intervention (70.1%) suggests that the older adults in the IG showed interest and willingness to engage in the community choir, which may be understood as an indication of the feasibility and acceptability of this

singing group program (Coulton et al., 2015). On the other hand, the current study also showed that the randomization procedure and four months of waiting period for the WLG participants has caused some dropouts. For example, some participants had enrolled with friends or spouses that were allocated to a different group, were not willing to be randomized to a waiting-list control condition or enrolled in other singing groups. With this in mind, the use of a stepped wedge cluster design may provide a more viable alternative to the WLG condition (Dingle et al., 2019), especially for older adults from the same institution site and that intended to share the program participation experience with friends and/or spouses.

A limited number of studies have also addressed and examined possible negative experiences and effects associated with amateur choral singing (e.g., Kreutz & Br nger, 2012). In line with this issue, moderated results in this study indicated that, alongside the previously mentioned benefits, participation in the group singing program was also found to induce higher levels of anxiety and depression, but only among older adults with diminished cognitive functioning. Studies that only included older adults with cognitive impairment/dementia showed that previous choir singing interventions had mixed effects, with some indicating benefits in terms of social relationships, sense of purpose and personal growth (e.g., Tamplin et al., 2018), whereas other studies reported a deterioration in depressive symptoms (Werner et al., 2017). Given previous findings and the results of this study, it is suggested that choir singing interventions for cognitively impaired elderly individuals should occur within more individualized (smaller groups) and flexible settings, as in the case of group music therapy. This framework is expected to help address individualized needs and abilities of the participants, as opposed to larger choir settings which pose higher demands and more potential stressors in singing instructed songs in a coordinated way with people with different cognitive abilities (Dingle et al., 2019; Werner et al., 2017).

Follow-up effects of the group singing intervention

Taking into consideration the results of the long-term effects, it was also found in this study that the group singing intervention benefits were sustained at 6-month follow-up. Moreover, between post-intervention and follow-up, significant reductions in anxiety were also observed in the IG. Given that some variability in the follow-up outcomes should be expected (Cohen et al., 2006, 2007), it is not possible to determine if these changes are the direct result of potential delayed intervention effects or if they are specifically associated with the aging process. However, both the maintenance of the short-term intervention-related effects and the follow-up improvements on the outcomes suggest accumulated health-related benefits for the IG individuals, after participating in the group singing program, especially when considering the characteristics of the current sample. Nevertheless, more research is needed regarding the psychological and physiological dynamics associated with group singing for older adults over time.

Mediation effects of group singing

After identifying the direct effects of the choir singing intervention on the outcomes, another aim of this study was to test and understand the potential 'generative'

mechanisms and linkages between group singing and the older adults' perceived levels of physical health, anxiety, stress and depression. Results of the parallel (multiple) mediation analyses showed that the singing intervention had significant indirect effects on the perceived physical and mental health outcomes, but only through some psychological mediators, namely affect balance and positive affect. The remaining psychological (life satisfaction and negative affect) and physical/physiological mediators (body balance, CRP and ESR) showed no significant mediating effects on the choir singing-outcomes relationships.¹ Additionally, when controlling for these indirect pathways, the significant effect of group singing on the perceived physical health was no longer significant, thus indicating indirect-only mediation. As such, these results provide new insights into the identification and understanding of the particular importance of the mechanisms responsible for the transmission of group singing benefits on physical and mental health outcomes.

Taken together, these findings suggest that an increase in positive affect experienced by the participants during the group singing intervention leads to increased hedonic well-being, which in turn leads to higher levels of perceived physical health and reduced anxiety, stress and depression symptoms. These findings confirm previous studies suggesting that choral singing activities and experiences are associated with better affective regulation (Bullack et al., 2018; Kreutz et al., 2004; Pires et al., 2018), and extend it by demonstrating that positive affect balance is a primary mechanism through which group singing benefits operate on health perception and depression in older adults. As such, the current study shows that improved mood regulation due to choral singing promotes better perceived physical and mental health in elderly individuals, and that this mediating mechanism was not conditional on the cognitive status of the participants. A possible explanation for the absence of significant direct effects in the mediation models tested for this sample of older adults, may lie on the duration of the intervention, which may have been insufficient for the beneficial effects of the group singing program to be transmitted to the physical and mental health outcomes. Therefore, future studies should attempt to investigate potential dose-response relationships between the number/duration of the group sessions, the mediating effect of (positive) hedonic balance and the related outcomes.

On the other hand, and contrary to expectations (Fancourt et al., 2014; Kang et al., 2018; Lally, 2009; Livesey et al., 2012), the singing program had no mediation effect through the physical and physiological variables (for more details on the direct effects on physiological variables, see Galinha, Pinal, et al., 2021). Possible explanations for these findings may include the diverse and heterogenous characteristics of our sample in terms of age (young and very old), education and socioeconomic levels (low), cognitive functioning (low to high), physical fitness (low) and the presence of several diagnosed diseases. Moreover, but contrary to initially planned in the research protocol, it was not possible to fully implement the group dynamics' exercises, which were expected to further promote both social and physical/physiological benefits. More precisely, the intervention team (maestro and pianist) tended to shorten this part of the sessions and focused more in the singing activity. Furthermore, the fact that this intervention was based on a non-established singing group may suggest the need for a longer intervention period, given that the short-term related effects tend to be more associated with social and/or emotional outcomes (Bullack et al.,

2018). Noteworthy, but not as direct result of the group singing intervention, this study also showed beneficial effects of life satisfaction on physical health and depression scores, a positive relationship between CRP and stress, as well as inverse associations of body balance with anxiety and stress levels. These findings are consistent with prior research on the same-aged population (Hallford et al., 2017; Sawa et al., 2020; Wium-Andersen et al., 2013), and emphasize the pertinence and relevance of studying these variables as potential explanatory mechanisms of the group singing-induced changes in the older adults' physical and mental health (Dingle et al., 2019).

Study strengths and limitations

Study strengths include the recruitment of a diverse community-based elderly sample, the use of an RCT design, the evaluation of an intervention based on a non-established choir, the simultaneous assessment and analysis of self-report and biological measures, and the analysis of follow-up data. Study limitations include the weaker implementation of the group dynamics' exercises as initially defined in the research protocol and the loss of participants at follow-up. Dropouts in studies and RCTs with older adults is a well-documented issue, particularly of waiting-list participants (Dingle et al., 2019). The sample of our study was drawn from a particular population, from a social care institution, different from the usually studied samples of younger older adults whose enrollment in the singing group was their own initiative. This sample includes younger and advanced aged older adults with concomitant health and cognitive function issues. It includes a majority of low educative and socioeconomic status participants, typically less interested in cultural programs. Moreover, this sample's participants did not take the initiative to enroll the program, but were invited. Nevertheless, results from general population samples are needed to test the efficacy of singing group programs in a wider spectrum of participants.

Conclusion

In summary, the results from this study suggest that group singing for older adults contribute to mitigate the declines in perceived physical health levels in comparison to the control group (WLG). These benefits were maintained at 6-month follow-up. In addition, results of the mediation analyses showed that the positive influences of choir singing on physical health, anxiety, stress and depression levels were better explained by indirect-only effects through increase of positive affect. On the other hand, moderation results also indicated that participation in the group singing program was associated with increased anxiety and depression symptoms, but only among participants with diminished cognitive functioning.

Taken as whole, these findings highlight the psychological and physical effects, and mediational mechanisms of choral singing in a large, demographically and cognitively diverse sample of older adults, invited to a singing group. Nevertheless, future research should further test these findings on the interplay of psychological, social

and physiological effects of group singing for older adults, taking into consideration the participants' characteristics, as cognitive function, education and physical health status.

Note

1. For detailed results on the direct effects of the Sing4Health on these variables, see Galinha, Pinal, et al. (2021) and Galinha, García-Martín, et al. (In press).

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No potential conflict of interest was reported by the authors.

RCT registration number

The Sing4Health study was pre-registered in June 14, 2019, with the trial registration number NCT03985917.

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