The role of late-differentiated T cells, a proxy for IFN-γ-production, in older adults’ social networks

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A B S T R A C T

Interferon-γ (IFN-γ), an inflammatory biomarker that promotes antiviral immunity, may be a prerequisite for sociability. IFN-γ production in older adulthood is driven by late-differentiated CD8+ T cells, particularly CD28+ and CD57+ subsets, which increase with age, reduce immune response, and increase chronic disease risk. The present study investigated the relationship between late-differentiated T cells (LDTC) and sociability in a longitudinal study of healthy aging. 139 older adults (M_age = 77.95, range 65–93, 58% female, 57% college educated, and 94% Caucasian) provided data at up to 10 occasions (M = 7). Social network size and diversity and cytomegalovirus (CMV) status were collected at every wave. Percentage of LDTC was measured at up to 4 waves and averaged for each participant. There were no significant main effects of LDTC or interactions between LDTC and time on social network size or diversity. Adjustment for baseline age, gender, and sensitivity analyses including CMV and imputed data did not change results. IFN-γ may not play a role in dictating social behavior in older adults. Alternately, LDTC may not have accurately represented circulating levels of IFN-γ. Future work should continue exploring IFN-γ and social behavior, particularly as it relates to age-related changes.

1. Introduction

Cell senescence and higher systemic inflammation characterize immune aging (Franceschi et al., 2007). Subsequent effects are associated with social environments: living in close contact with others typically requires better viral immunity, whereas living alone typically requires better inflammatory responses (for review, see Cole, 2012; Leschak and Eisenberger, 2019). Interferon-γ (IFN-γ), a cytokine that promotes inflammation and antiviral immunity, may influence social behavior as a function of aligning motivation and immune defense associated with different social environments (Farrar and Schreiber, 1993; Filiano et al., 2016). The present study investigated percentages of late-differentiated T cells (LDTC), which produce IFN-γ, and sociability in a longitudinal study of older adults.

IFN-γ production increases with age and is classically recognized as promoting antiviral immunity but also promotes pro-inflammatory processes, such as macrophage activation (Monteiro et al., 2017). One major source of IFN-γ production in older adulthood is late-differentiated CD8+ T cells, particularly CD28+ and CD57+ subsets, which naturally accumulate with age, reduce immune response to challenge, and contribute to chronic disease development (Bandrés et al., 2000; for review, see Strioga et al., 2011). The majority, 68%, of CD28+ cells in older mice produce IFN-γ compared with 9% in younger mice (Ortiz-Suárez and Miller, 2002). In humans aged 17–62 years old, percentage of CD8+ cells producing IFN-γ was positively correlated with percentage of CD8+CD57+ cells (r = 0.58, p < .05) and negatively correlated with CD8+CD57− cells (r = −0.71, p < .01; Bandrés et al., 2000). Further, CD8+CD28− T cells in older adults show a three-fold increase in IFN-γ production compared with young controls (Eylar et al., 2001). Thus, CD8+CD57+ and CD8+CD28− cells in older adults may reflect IFN-γ levels.

LDTC that produce IFN-γ may influence sociability, including qualities of social networks (Bandrés et al., 2000; Filiano et al., 2016; Monteiro et al., 2017). In rodents, IFN-γ-knockout mice did not prefer other mice over inanimate objects, but this preference was restored after one recombinant IFN-γ injection (Filiano et al., 2016). In humans, higher IFN-γ is related to more social support in healthy adults and larger household size in female breast cancer survivors (Leschak et al., 2021; Miyazaki et al., 2005). Thus, normative decreases in older adults’ social network size and diversity over time (Lang and Carstensen, 1994) may
be an element of the behavioral immune system (Ackerman et al., 2018), compensating for decreases in antiviral immunity that occurs with aging. However, to the degree that anti-viral immunity is maintained (e.g., by IFN-γ production), social network size and diversity may also be maintained.

Sociability in older age may be related to IFN-γ via LDTC. The present study tested this hypothesis in older adults, using late-differentiated CD8+ T cells as a proxy for IFN-γ production. The following hypotheses were tested:

1. Higher average percentages of LDTC (CD3+CD8+CD28 and CD3+CD8+CD57) over 5 years will be associated with a larger and more diverse social network.

2. Higher average percentages of LDTC (CD3+CD8+CD28 and CD3+CD8+CD57) over 5 years will be associated with less decrease in size and diversity of social network over time.

Models were adjusted for baseline age and gender. Cytomegalovirus (CMV), a latent herpes virus related to immune senescence, is associated with more LDTC and less production of IFN-γ in CMV-specialized T cells, and was included in sensitivity analyses (Derhovanessian et al., 2009; Ouyang et al., 2003; Reed et al., 2019).

2. Materials and methods

2.1. Participants and procedures

Participants included 139 healthy older adults (M_age = 77.95; range 65–93; 58% female; 57% college-educated; 94% Caucasian, 3.7% African American, and 1.8% Asian American/Pacific Islander). Exclusion criteria included chronic diseases or treatments involving the immune system (e.g., autoimmune disease, steroid use) or more than two of the following medication classes: psychotropics, anti-hypertensives, hormone replacements, or thyroid supplement. Study procedures were approved by the Institutional Review Board at the University of Kentucky. Participants were interviewed every six months up to 10 times between July 2011 and April 2018. A phlebotomist drew blood by venipuncture the week following each interview. Participants were compensated US$50 for each visit.

Of 1390 possible observations, 49 participants no longer wanted to participate (186 person-waves), 35 participants did not complete all 10 waves before study end (98 person-waves), participants missed a total of 38 person-waves, 6 participants dropped out due to loss of contact (33 person-waves), and 4 participants dropped out due to death (22 person-waves). Overall, 1013 observations were available for analysis. A sensitivity analysis using the paramtest and bmeta packages in R (Bates et al., 2015; Hughes, 2017) with 1000 iterations indicated the study was powered >.74 for medium effects (|β| = 0.3) and >.97 for large effects (|β| = 0.5).

2.2. Measures

2.2.1. Social Network Index

The 12-item Social Network Index measured social network size and diversity in 12 social relationship categories (Cohen et al., 1997). Network size was calculated as the number of people contacted at least once over two weeks (maximum = 7 or more); network diversity was calculated as the number of social roles (e.g., spouse, children, close friends, etc.; maximum = 7 or more) contacted at least once over two weeks. This index item characterizing social groups was excluded due to inconsistencies across interviewers in how data were recorded.

2.2.2. Late-differentiated T cells

Immunological markers of LDTC were evaluated by flow cytometry (see Reed et al., 2019). T cells were defined as CD3+ cells within the lymphocyte population, and subsets with cell surface markers CD8, CD28, and CD57. Cells were analyzed on a LSR-II flow cytometer (BD, San Jose, CA). Data were analyzed using FlowJo v7.6 software. The T cell composite, representative of IFN-γ-producing T cells, included mean proportions of CD3+CD8+CD28+ and CD3+CD8+CD57+ cells (composite α = .96 across all participants and observations; see Reed et al., 2019, Figure 1, for details on gating strategy used to detect LDTC), expressed as a percentage of total CD8+ cells. The intraclass correlation was 0.93, indicating variability primarily at the between-person level (cf., Reed et al., 2019), thus LDTC were only evaluated at the between-person level.

2.2.3. Covariates

Covariates were baseline age (centered around sample grand mean = 77.95), gender (reference is male), and CMV serostatus (reference is seronegative). CMV serostatus was included to account for differences in T cell aging and IFN-γ production (Reed et al., 2019). CMV IgG titers were collected at each visit and determined by ELISA assay (DRG International, Inc., Springfield, NJ). The intra-assay and inter-assay coefficients of variability were 5.1% and 9.9%. CMV IgG Index values 1.0 IU/mL or greater were considered seropositive (n = 109/149, 73% of the sample). Equivalval values were handled as previously described (Reed et al., 2019).

2.3. Data analysis

Data were analyzed using multi-level models (PROC MIXED) with restricted maximum likelihood estimation in SAS 9.4, with within-person variation at level 1 and between-person variation at level 2. Hypothesis 1 tested main effects of time and the T cell composite; Hypothesis 2 tested an interaction between time and T cell composite. A random intercept and random time slope model was selected based on Akaike’s information criteria and Bayesian information criteria over models with other covariance structures. Kenward-Roger degrees of freedom were used to prevent standard error inflation and protect parameter estimates from the impact of missing data (Chawla et al., 2014). CMV serostatus was included in sensitivity analyses. Fixed effects are reported as unstandardized β weights, analogous to unstandardized b weights, with 95% confidence intervals. In a sensitivity analysis, missing social network data (10%) were imputed (k = 30) using the mice package in R (Van Buuren and Groothuis-Oudshoorn, 2011) and analyzed with PROC MIANALYE in SAS 9.4.

3. Results

3.1. Descriptive statistics

Participants reported about 20 people in their social network with whom they spoke at least once every two weeks (M_diag = 20.51, SD = 6.7, range = 0–47) and about 6 different types of social relationships (M_varies = 5.82, SD = 1.31, range = 0–16). The average percentage of LDTC across all participants and visits was 52.14% (SD = 21.4%, range = 2.56–91.25).

Social network size was positively correlated with social network diversity (r = 0.69, p < .0001, Table 1). Higher percentage of LDTC was correlated with CMV + serostatus (r = 0.54, p < .0001). Social network size and diversity were modestly, but not significantly, correlated with percentage of LDTC (r = 0.14, p = .10 and r = 0.08, p = .35, respectively).

3.2. Effects of LDTC on social network size and diversity

LDTC percentages were not significantly related to social network parameters, before or after adjusting for baseline age and gender (Table 2, Model 3–4). Although higher percentage of LDTC was associated with larger social network size (effect size r = 0.14–0.15), this relationship was not statistically significant (Model 2, p = .10; Model 3,
Effects of LDTC on social network size and diversity (N = 139).

### Table 1

Correlations among study variables (N = 139).

|            | 1     | 2     | 3     | 4     | 5     | 6     |
|------------|-------|-------|-------|-------|-------|-------|
| 1. SN Size | –     | –     | –     | –     | –     | –     |
| 2. SN Diversity | .694 [.595, .770] | –     | –     | –     | –     | –     |
| 3. Baseline Age | -.006 [-.172, .160] | -.156 [-.313, .012] | –     | –     | –     | –     |
| 4. Education | -.043 [-.208, .124] | .064 [-.103, .228] | .104 [.064, .266] | –     | –     | –     |
| 5. Gender | .011 [-.156, .177] | -.015 [-.181, .152] | -.144 [-.303, .024] | -.210 [-.362, -.043] | –     | –     |
| 6. T cells | .129 [.028, .298] | .081 [.087, .244] | .039 [-.129, .204] | -.046 [-.211, .126] | -.057 [-.221, .111] | –     |
| 7. CMV | .089 [-.079, .252] | -.029 [-.195, .153] | .057 [-.111, .221] | -.101 [-.262, .064] | .065 [.103, .229] | .541 [.410, .648] |

**Note.** Bold font indicates \( p < .05 \). SN Size = Social Network Size subscale of Social Network Index (Cohen et al., 1997); SN Diversity = Social Network Diversity subscale of Social Network Index (Cohen et al., 1997); Gender is coded 1 = female, 0 = male; T cells = Average late-differentiated T cell composite, proxy for IFN-\( \gamma \) production; CMV = Cytomegalovirus serostatus, coded 1 = positive, 0 = negative.

### Table 2

Effects of LDTC on social network size and diversity (N = 139).

#### Social Network Size

|            | Model 1 | Model 2 | Model 3 | Model 4 | Model 5 | Model 6 | Model 7 |
|------------|---------|---------|---------|---------|---------|---------|---------|
| Intercept  | .20.536 [19.423, 21.648] | .20.569 [19.275, 21.863] | .20.569 [19.286, 21.853] | .20.385 [18.551, 22.220] | .20.573 [19.289, 21.858] | .20.383 [18.548, 22.218] | .20.349 [17.665, 23.034] |
| Visit      | -.008 [-.170, .153] | -.005 [-.167, .157] | -.005 [-.167, .157] | -.008 [-.171, .154] | -.008 [-.170, .154] | -.008 [-.171, .154] | -.015 [-.231, .197] |
| T cells    | .045 [-.008, .099] | .046 [-.009, .100] | .056 [-.007, .119] | -.003 [-.011, .005] | -.003 [-.011, .005] | -.003 [-.011, .005] | .056 [-.137, .130] |
| Gender     | .319 [.1957, 2.596] | .319 [.1957, 2.596] | .319 [.1957, 2.596] | .319 [.1957, 2.596] | .319 [.1957, 2.596] | .319 [.1957, 2.596] | .319 [.1957, 2.596] |
| CMV        |        |        |        |        |        |        | .331 [-1.946, 2.609] |

#### Social Network Diversity

|            | Model 1 | Model 2 | Model 3 | Model 4 | Model 5 | Model 6 | Model 7 |
|------------|---------|---------|---------|---------|---------|---------|---------|
| Intercept  | .5.856 [5.636, 6.075] | .5.966 [5.730, 6.263] | .5.966 [5.729, 6.263] | .5.636 [5.666, 6.263] | .5.995 [5.728, 6.262] | .5.636 [5.666, 6.262] | .5.636 [5.666, 6.262] |
| Visit      | -.033 [-.070, .003] | -.033 [-.069, .004] | -.033 [-.069, .003] | -.032 [.069, -.004] | -.032 [.069, -.004] | -.032 [.069, -.004] | -.032 [.069, -.004] |
| T cells    | .006 [.005, .017] | .006 [.005, .017] | .004 [.009, .017] | .000 [.001, .002] | .000 [.001, .002] | .000 [.001, .002] | .007 [-.008, .022] |
| Gender     | -.072 [-.052, .376] | -.072 [-.052, .376] | -.073 [-.052, .376] | -.073 [-.052, .376] | -.073 [-.052, .376] | -.073 [-.052, .376] | -.073 [-.052, .376] |
| CMV        |        |        |        |        |        |        | .375 [.404, .466] |

**Note.** Bold font indicates \( p < .05 \). T cells = Average (Level 2) late-differentiated T cell composite, proxy for IFN-\( \gamma \) production; Gender is coded 1 = female, 0 = male; CMV = Cytomegalovirus serostatus, coded 1 = positive, 0 = negative.

\( p = .10 \). There were no statistically significant interactions between time and percentage of LDTC on social network size or diversity (ps > .50; **Table 2**, Models 5–7). A 1-year increase in baseline age was associated with a \(-0.044\) decrease in social network diversity (\( p = .039 \)). Inclusion of CMV serostatus and imputation of missing data did not change these results. Post-hoc, we repeated all analyses using the absolute LDTC counts instead of percentages (see Reed et al., 2019 for details regarding the calculation of counts). LDTC counts were significantly positively correlated with CMV serostatus (\( r = 0.439, p < .0001 \)) but not other study variables. LDTC count did not significantly predict social network size (\( \gamma = -0.0007, SE = 0.006, p = .91, 95\% CI [-.012, 0.011] \)) or diversity (\( \gamma = -0.001, SE = 0.001, p = .34, 95\% CI [-.003, 0.0012] \)) and adding covariates to these models did not change the null results (\( \gamma = -0.0002, SE = 0.007, p = .98, 95\% CI [-.015, 0.015] \)) and \( \gamma = -0.001, SE = 0.002, p = .33, 95\% CI [-0.004, 0.002], \) respectively).

### 4. Discussion

IFN-\( \gamma \) is a cytokine with proinflammatory and antiviral properties that may influence sociability in older age (Moieni and Eisenberger, 2022).
In this study, LDTC, which produce IFN-γ, was not related to social network size or diversity. CMV seropositivity correlated with a higher proportion of LDTC, but including CMV serostatus in the models did not change results (Bandrés et al., 2000; Reed et al., 2019). An exploratory analysis testing the interaction between CMV and LDTC was also not significant. A positive, though not statistically significant, relationship between LDTC and social network size merits future investigation. This study was sufficiently powered to statistically significant, relationship between CMV and LDTC was also not significant. A positive, though not selectively induced by IFN-γ or other socially relevant IFN-γ target tissues in representative samples of older adults.

Strengths of this study include longitudinal study design and adjustment for CMV serostatus, a major immunological confounder. The sample of healthy older adults was a strength in contrast with previous investigations in rodent models, younger, and clinical populations, but its relative unrepresentativeness may have contributed to null findings. Further, since strict exclusion criteria, it is always possible that chronic diseases, developed after the initial study visit, could have confounded results. Lastly, low within-person variability in LDTC suggests that a longer period of data collection is required to investigate within-person changes. Studies conducted over longer periods may have different findings, as within- and between-person results do not always align (Kievit et al., 2013). Although preclinical evidence suggests that IFN-γ plays a causal role in social behavior (Filiano et al., 2016), the present study design does not allow for causal inference. Future work should build on the present, correlational work with experimental investigations exploring the potential causal role of IFN-γ in age-related changes and decline, subjective and objective changes in sociability that occur with age, the inclusion of other determinants of social changes and decline, subjective and objective changes in sociability which may occur with age, the inclusion of other determinants of social interaction, and what may mitigate these effects.

Declaration of competing interest
The authors declare no conflicts of interest.

Data availability
The data that has been used is confidential.

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