

Out-of-home informal support important for medication adherence, diabetes distress, hemoglobin A1c among adults with type 2 diabetes

Lindsay S. Mayberry^{1,2}  · John D. Piette^{3,4} · Aaron A. Lee⁴ · James E. Aikens⁵

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Abstract Adults with type 2 diabetes mellitus (T2DM) often receive self-management support from adult children, siblings or close friends residing outside of their home. However, the role of out-of-home support in patients' self-management and well-being is unclear. Patients (N = 313) with HbA1c > 7.5% were recruited from community primary care clinics for a mobile health intervention trial and identified an out-of-home informal support person, herein called a CarePartner; 38% also had an in-home supporter. We tested cross-sectional adjusted associations between CarePartner relationship characteristics and patients' self-management, diabetes distress, and HbA1c and whether having an in-home supporter modified these associations. Greater CarePartner closeness was associated with a greater odds of perfect medication adherence (AOR =

1.19, $p = .029$), more fruit/vegetable intake ($\beta = 0.14$, $p = .018$), and lower diabetes distress ($\beta = -0.14$, $p = .012$). More frequent CarePartner contact was associated with *better* HbA1c among patients with an in-home supporter but with *worse* HbA1c among patients without an in-home supporter (interaction $\beta = -0.45$, $p = .005$). Emotional closeness with a CarePartner may be important for supporting T2DM self-management and reducing diabetes distress. CarePartners may appropriately engage more frequently when patients with no in-home supporter have poorly controlled diabetes.

Keywords Disease self-management · Informal caregivers · Family · Social support · Type 2 diabetes · Glycemic control

✉ Lindsay S. Mayberry
lindsay.mayberry@vumc.org

John D. Piette
jpiette@med.umich.edu

Aaron A. Lee
aaronlee@med.umich.edu

James E. Aikens
aikensj@med.umich.edu

- ¹ Department of Medicine, Vanderbilt University Medical Center, Nashville, TN, USA
- ² Center for Health Behavior and Health Education, Vanderbilt University Medical Center, Nashville, TN, USA
- ³ Department of Health Behavior and Health Education, University of Michigan, Ann Arbor, MI, USA
- ⁴ VA Center for Clinical Management Research, Ann Arbor, MI, USA
- ⁵ Department of Family Medicine, University of Michigan, Ann Arbor, MI, USA

Introduction

The prevalence of diabetes alongside high rates of poor glycemic control (Centers for Disease Control and Prevention, 2017; Selvin et al., 2014) contribute to overwhelming diabetes-related costs (American Diabetes Association, 2018). For adults with type 2 diabetes mellitus (T2DM), performing recommended self-management (i.e., adherence to medications, maintaining a healthful diet, getting regular physical activity, and self-monitoring blood glucose) improves glycemic control (Jones et al., 2003) and prevents disease-related complications and premature mortality (The Diabetes Care and Complications Trial Research Group, 1993; UK Prospective Diabetes Study Group, 1998). Yet, self-management remains challenging (Bowen et al., 2001; DiMatteo, 2004b) and diabetes related distress is common among adults with diabetes (Fisher et al., 2008, 2012). Daily T2DM self-management often

occurs in social and environmental contexts and is influenced by the availability and involvement of close family and friends (Fisher et al., 1998; Silliman et al., 1996). Among adults with T2DM, family/friend support indirectly affects health through better psychological well-being, including less diabetes distress (Baek et al., 2014; Schiøtz et al., 2012) and better quality of life (Tang et al., 2008), and directly facilitates patients' self-management (DiMatteo, 2004a; Glasgow & Toobert, 1988; Mayberry & Osborn, 2012, 2014; Rosland et al., 2012) leading to reduced HbA1c levels (Mayberry & Osborn, 2014; Nicklett et al., 2013).

Most research on support for adults' T2DM focuses on in-home family members (Burns et al., 2013; Fisher et al., 2000; Stephens et al., 2013; Wen et al., 2004) or does not distinguish between in-home and out-of-home support (Mayberry & Osborn, 2012, 2014; Rosland et al., 2010, 2012; Vongmany et al., 2018). Over one-third of older U.S. adults live alone (US Department of Health and Human Services Administration on Aging, 2017). However, most older adults (78%) have adult children living outside their home with whom they have weekly contact (Piette et al., 2010), and up to 7 million Americans report receiving "long-distance" caregiving (Piette et al., 2010). Even when adults with T2DM live with a spouse or partner, the spouse/partner may be dealing with chronic conditions of their own which may undermine their ability to provide support to the patient (Piette et al., 2010), and increase the need for out-of-home support. Although many patients receive substantial disease-related support from other family members or close friends who reside outside of their household (Lee et al., 2017; Rosland et al., 2013), the role of out-of-home support remains unclear and understudied.

A fuller understanding of the effects of out-of-home support is needed to most effectively leverage existing family structures and relationships to bridge the gap between what clinicians can realistically provide and what patients must do daily to manage chronic illness (Piette et al., 2010). To understand when and for whom out-of-home support may be important, we tested specific hypotheses derived from two theories of social support: the direct (or main) effect model and the deficit (or stress-buffering) model (Cohen & Wills, 1985; Rodriguez & Cohen, 1998). The direct effect model posits that structural and perceived social support improves health outcomes and psychological well-being, whereas the deficit model asserts that the positive effects of support are specifically activated within the context of a psychosocial stressor (Cohen & Wills, 1985). Situations in which an individual is challenged by chronically excessive demands without sufficient resources to meet those demands is one such stressor, and is known as a deficit state (Jacobson, 1986; Weiss, 1976). In the context of T2DM, the required daily self-care

behaviors and potential comorbidities and complications may place patients in or at risk of a deficit state, particularly those without the resource of an in-home supporter.

Applied specifically to in-home versus out-of-home support for T2DM self-management, the direct effect model implies that having an in-home supporter and more out-of-home support will *each* be associated with better self-management, lower diabetes distress, and better glycemic control. If this is true, then it ought to be beneficial to engage out-of-home supporters in self-management regardless of in-home supporter presence. In contrast, the deficit model suggests that out-of-home support is *only or more* beneficial among patients without an in-home supporter, given that these patients have a greater needs deficit than those who have an in-home supporter. In contrast to above, this would imply that engaging out-of-home supporters is beneficial only for patients who have no in-home supporter or a highly strained in-home supporter. Furthermore, we examined frequency of seeing and frequency of talking with the out-of-home supporter separately because differential effects may have implications for the role of long-distance out-of-home supporters.

Methods

As part of a larger evaluation of an intervention designed to support T2DM patients' self-care and strengthen the assistance that they receive from members of their social network (Piette et al., 2014), adults with T2DM and HbA1c > 7.5% were recruited from four community primary care clinics that were geographically distributed throughout the lower peninsula of Michigan (NCT01684709). Recruitment occurred December 2012 through June 2016. Participants were required to identify a close friend or adult relative who lived outside their home and was willing to enroll as a CarePartner in support of the patient's diabetes self-management. Patients were excluded if they were in palliative care, on a transplant waitlist, at high risk for 1-year mortality, or screened positive for either significant cognitive impairment or an unstable psychiatric condition. Eligible participants were identified via electronic health record review and recruited with a letter followed by a screening phone call. During screening, potential participants identified one to four out-of-home supporters who could enroll in the study as their CarePartner. To be eligible, CarePartners had to live within the continental U.S., have a working phone number and access to the internet, be fluent in English, at least 21 years old, and not be psychiatrically unstable or cognitively impaired. For this analysis we used baseline data from patient participants. Participants completed written informed consent and an HbA1c test during a research visit

to their regular clinic. Self-report measures were completed via phone with study staff within 30 days of the baseline HbA1c test.

Measures

Covariates

Participants self-reported demographic and diabetes characteristics including age, gender, race/ethnicity, income (5 ordinal categories, see Table 1), diabetes duration, and insulin use (yes/no).

In-home supporter presence

Patients were determined to have an in-home supporter (yes/no) if an adult who lived with them provided help with their diabetes management. This was determined through structured queries (e.g., “is there anyone in your household who helps you manage your diabetes?”).

Table 1 Characteristics of patient sample (N = 313)

Age, years (mean \pm SD)	54.6 \pm 10.5
Female (%)	59.8
Race/ethnicity (%)	
Non-hispanic white	66.4
Non-hispanic black	24.8
Hispanic	3.7
Other race/ethnicity	5.1
Income, USD (%)	
< \$15,000	45.5
\$15,000 to < \$30,000	34.3
\$30,000 to < \$50,000	11.2
\$50,000 to < \$75,000	4.2
\geq \$75,000	4.8
Diabetes duration, years (mean \pm SD)	12.6 \pm 8.1
Prescribed insulin (%)	69.2
Perfect medication adherence, last 7 days (%)	64.5
Frequency of following healthy eating plan, last 7 days (mean \pm SD)	3.7 \pm 2.3
Frequency of fruit/vegetable consumption, last 7 days (mean \pm SD)	3.7 \pm 2.5
Frequency of high-fat food consumption, last 7 days (mean \pm SD)	3.5 \pm 2.0
Frequency of physical activity, last 7 days (mean \pm SD)	1.9 \pm 1.9
Diabetes distress (mean \pm SD)	2.7 \pm 1.00
Hemoglobin A1c, % (mean \pm SD)	9.6 \pm 1.8

CarePartner relationship characteristics

We assessed three aspects of patients’ relationship with their identified CarePartner: emotional closeness, frequency of seeing and frequency of talking with the CarePartner. Participants reported their emotional closeness with their CarePartner on a scale from 1 = “not very close at all” to 10 = “very close.” We mean centered emotional closeness for analyses to aid in interpretability of effects in models including interactions. Frequency of contact was assessed by asking participants “Over the last 6 months, about how often in an average month do you...talk with your CarePartner? ...see your CarePartner in person?” We examined frequency of talking with the CarePartner and seeing the CarePartner separately. Responses were scaled 0 = “once per month or less,” 0.5 = “twice/once every 2 weeks,” 1.0 = “once weekly” and 2.0 = “more than once weekly.”

Patient outcomes: self-management, diabetes distress, and HbA1c

We assessed participants’ diabetes self-management behaviors over the last 7 days with the Brief Medication Questionnaire (Svarstad et al., 1999) and the Summary of Diabetes Self-Care Activity (SDSCA) (Toobert et al., 2000). The Brief Medication Questionnaire is sensitive to both repeat and sporadic dose skipping (Svarstad et al., 1999). We repeated the item “How many times did you miss taking a dose (pill/injection)?” separately for each of up to four prescribed diabetes medications. We calculated a dichotomous variable reflecting perfect medication adherence (i.e. no missed doses the past 7 days) versus less than perfect medication adherence (\geq 1 missed dose the past 7 days). This approach is recommended to help counteract inflated adherence reporting due to social desirability and recall bias and to enhance sensitivity and specificity of self-report measures against objective measures and/or clinical outcomes (Stirratt et al., 2015). Diet items ask about number of days out of the last 7 in which the respondent followed a healthful eating plan, ate five or more servings of fruits or vegetables, and ate high fat foods. The SDSCA items assessing diet were analyzed separately according to Toobert et al.’s (2000) recommendation, due to low inter-item correlations for the subscale ($\alpha = 0.32$ in this sample). We assessed physical activity with the SDSCA subscale ($\alpha = 0.76$ in this sample) which asks about number of days out of the last 7 in which the respondent had 30 min of continuous activity and had a specific exercise session.

Participants completed the Diabetes Distress Scale (Polonsky et al., 2005), a 17-item questionnaire ($\alpha = 0.89$ in this sample) which assesses patients’ emotional burden, physician-related distress, regimen-related distress, and

diabetes-related interpersonal distress. Respondents rate the degree to which each item had been a problem over the last month from 1 = “not a problem” to 6 = “a very serious problem.” Responses were averaged to generate scale scores ranging 1–6, with higher scores indicating more diabetes distress. Glycemic control was assessed with the hemoglobin A1c (HbA1c) test, which was completed during enrollment using a Siemens DCA Vantage Analyzer point-of-care test which analyzes a drop of fingertip blood. Higher HbA1c values indicate worse glycemic control.

Analysis

Stata version 14.2 was used for all analyses. Descriptive statistics were used to describe the sample and types of relationships represented by their CarePartners. Spearman correlation coefficients were used to explore pairwise associations between the predictors and outcomes of interest. Next, we used a series of logistic or linear ordinary least squares multivariate regression models to test specific hypotheses. To evaluate the direct effect model hypotheses, we examined cross-sectional associations between predictors (in-home supporter presence and CarePartner relationship characteristics) and outcomes (medication adherence, diet, exercise, diabetes distress, and HbA1c). To evaluate the deficit model hypotheses, we then tested whether these associations were modified by in-home supporter presence by adding an interaction term (in-home supporter presence X CarePartner relationship characteristic) to each of the direct effect models. Interaction effects significant at $p < .05$ were stratified and graphically depicted for interpretation, and marginal effects were used to calculate adjusted estimates and their 95% confidence intervals.

All regression models were adjusted for age, gender, minority race/ethnicity, annual income < \$15,000, diabetes duration, and insulin use. Covariates were selected a priori on the basis of their documented associations with adherence and HbA1c. We dichotomized race as minority versus non-Hispanic White and income as < \$15,000 USD versus \geq \$15,000 USD. These dichotomizations were selected for statistical reasons (i.e., to achieve similar cell sizes) and substantive differences [i.e., racial/ethnic minorities with T2DM have worse adherence and poorer outcomes than non-Hispanic Whites (Herman et al., 2009; Kirk et al., 2006; Mayberry, Bergner, et al., 2016; Weinstock et al., 2011), and \$15,000 USD was roughly the poverty level for a two-person household during the study enrollment period (United States Census Bureau)]. We used robust standard errors (type HC3) for efficient estimates with conservative standard errors regardless of heteroscedasticity (Hayes & Cai, 2007).

Data were incomplete for seven variables used in multivariate analyses, ranging from 0.03 to 7.8% missing values. Casewise deletion would have eliminated 15.0% of the sample thereby reducing our statistical power and potentially introducing non-response bias (Little et al., 2012). Therefore, we imputed 10 datasets with multiple imputation using chained equations. The imputation model including all variables to be used in the multivariate models and the effect modifier of in-home supporter presence (Graham, 2009; Little et al., 2012). All regression models were conducted with imputed data.

Results

Table 1 shows descriptive statistics for $N = 313$ patient participants. Patients had a mean age of 54.6 years old ($SD = 10.5$) and were predominantly non-Hispanic White (66.4%) or non-Hispanic Black (24.8%), 59.8% were female, and 46.2% had an annual household income less than \$15,000 USD. Most participants (86.9%) had completed high school or a GED. A quarter (25.8%) of participants lived alone, while 37.9% had an in-home supporter. Notably, 50.3% were married/partnered but not all of patients' spouses/partners met in-home supporter criteria according to their responses to the structured query (e.g., they lived separately or the spouse/partner did not provide any help managing T2DM). Participants were prescribed an average of 2.1 ($SD = 0.8$) diabetes medications and 69.2% were prescribed insulin. Average HbA1c was 9.6% ($SD = 1.8\%$).

CarePartners were 37.5% adult children or grandchildren (including step, in-law), 26.3% friends (including neighbors and coworkers), 21.8% siblings (including in-law), 5.5% partners or ex-partners, and 10.2% parents, grandparents or other relatives. Most participants reported high emotional closeness with their CarePartner (83.5% reported ≥ 8 on the 10-point scale). Contact between participants and their CarePartner was frequent; 75.8% reported talking at least twice per week and 55.2% reported seeing the CarePartner at least once per week. As can be seen in Table 2, CarePartner relationship characteristics had significant low to moderate correlations with one another (Spearman's rho: 0.14–0.36), suggesting each variable assessed different aspects of the CarePartner relationship.

Table 3 shows results of adjusted direct effect models. In-home supporter presence was associated with less diabetes distress ($\beta = -0.12$, $p = 0.028$), but not with self-management or HbA1c. Greater emotional closeness with the CarePartner was independently associated with greater odds of medication adherence (AOR = 1.19, $p = 0.029$), more fruit/vegetable consumption ($\beta = 0.14$, $p = 0.018$),

and with lower diabetes distress ($\beta = -0.14, p = 0.012$). Neither frequency of talking with nor seeing the CarePartner was associated with patients’ self-management or diabetes distress in direct effect models (see Table 2).

In adjusted models testing the deficit model, we found that in-home supporter presence moderated the association between frequency of contact with the CarePartner (interaction terms significant for both talking with, $\beta = -0.45, p = 0.005$, and seeing $\beta = -0.26, p = 0.021$) and HbA1c. For patients with an in-home supporter, more frequently talking with ($\beta = -0.24, p = 0.018$) or seeing the CarePartner ($\beta = -0.20, p = 0.040$) was associated with lower HbA1c; whereas for patients without an in-home supporter, more frequently talking with ($\beta = 0.12, p = 0.020$) or seeing the CarePartner ($\beta = 0.15, p = 0.042$) was associated with higher HbA1c. Because similar results emerged for talking and seeing, only the adjusted predictions for the frequency of talking with the CarePartner are shown in Fig. 1. Presence of an in-home supporter did not moderate any other associations.

Discussion

Among adults with T2DM and elevated HbA1c levels receiving care at community primary care clinics, approximately a quarter lived alone and most (62%) did not have an adult in their home who helped them manage their T2DM. Most patients identified adult children or grandchildren, friends, and siblings as a CarePartner and most reported contact with their CarePartner at least twice per week. Greater emotional closeness with the out-of-home CarePartner was associated with better diabetes medication adherence, more fruit and vegetable intake and less dia-

betes distress. These associations were not moderated by the presence of an in-home supporter, which suggests that out-of-home support may benefit T2DM management for the vast majority of patients. We also found that having an in-home supporter was associated with less diabetes distress but not with better self-management or glycemic control. Collectively, these findings are consistent with the direct effect model of social support on diabetes distress and, to a lesser degree, on regular self-management.

More frequent contact with the CarePartner was associated with better glycemic control among patients with an in-home supporter but with worse glycemic control among patients with no in-home supporter. For patients with an in-home supporter, support from within and outside the household may work synergistically to help patients achieve and maintain glycemic control, consistent with the direct effect model. However, the findings for patients without an in-home supporter were in contrast to our deficit model hypothesis, given that CarePartner contact was associated with worse rather than better outcomes for those who had no in-home supporter. Understood in the context of the associations between emotional closeness and better self-management and less diabetes distress, this finding may indicate that concerned out-of-home friends and family members more frequently contact poorly controlled patients who receive little or no support within their home. We examined frequency of talking with and seeing the CarePartner separately to understand if in-person interactions played a distinct role, but these variables showed similar associations (or lack thereof) with the outcomes of interest despite the moderate correlation between the two. This suggests long-distance support may be just as relevant. However, relatively few of our participants reported

Table 2 Pairwise Spearman’s rho correlations among predictors and outcomes

	1	2	3	4	5	6	7	8	9	10
1. In-home supporter present	1.00									
2. Emotional closeness with CarePartner	-.059	1.00								
3. Frequency of talking with CarePartner	-.045	.321***	1.00							
4. Frequency of seeing CarePartner	.008	.137*	.362***	1.00						
5. Perfect medication adherence	-.029	.134*	.000	.054	1.00					
6. Following healthy eating plan	.077	-.003	.023	.043	.133*	1.00				
7. Fruit/vegetable consumption	.041	.159**	.051	.014	.159**	.236***	1.00			
8. High-fat food consumption	-.012	-.088	-.032	-.018	-.124*	-.184**	.020	1.00		
9. Physical activity	.017	-.078	-.016	-.039	.146*	.115*	.092	.066	1.00	
10. Diabetes distress	-.144*	-.078	.003	.003	-.219***	-.196***	-.089	.046	-.137*	1.00
11. Hemoglobin A1c, %	-.033	-.086	.073	.073	-.077	.011	.001	.009	-.122*	.184**

* $p < .05$, ** $p < .01$, *** $p < .001$

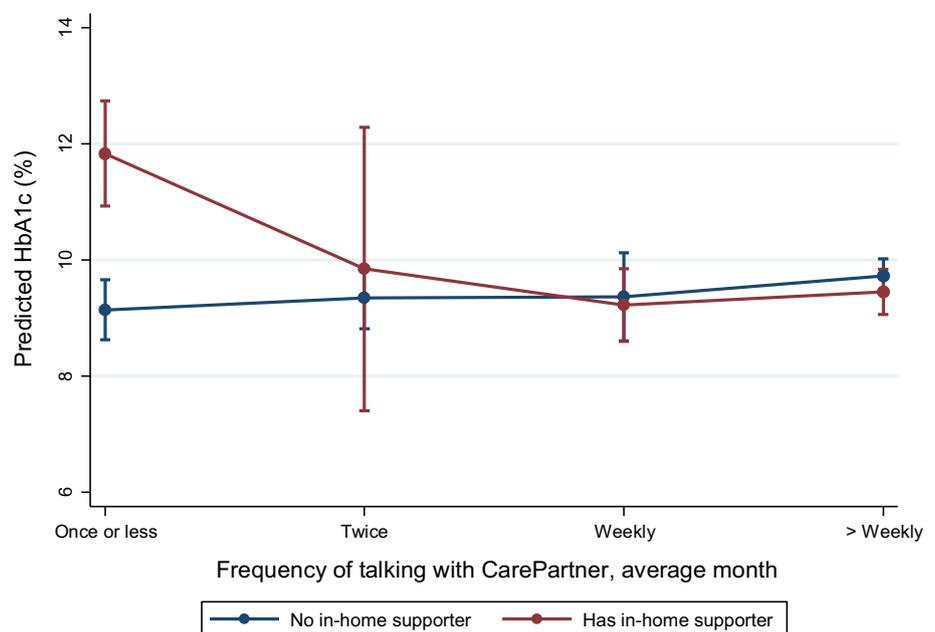
Table 3 Adjusted associations between in-home supporter presence and CarePartner relationship characteristics on patients' self-management, diabetes distress, and hemoglobin A1c

	Perfect medication adherence		Following healthy eating plan		Fruit/vegetable consumption		High-fat food consumption		Physical activity		Diabetes distress		HbA1c, %	
	AOR	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>
In-home supporter present	0.86	.580	.12	.057	.07	.233	-.06	.270	.00	.966	-.12	.028	.00	.986
Emotional closeness with CarePartner	1.19	.029	-.06	.376	.14	.018	-.04	.474	-.07	.327	-.14	.012	-.11	.111
Frequency of talking with CarePartner	1.05	.822	.04	.519	.05	.395	.00	.969	-.01	.838	-.04	.470	-.03	.634
Frequency of seeing CarePartner	1.20	.223	.06	.330	.03	.557	-.02	.755	-.04	.514	.01	.915	.03	.613

Each predictor examined in a separate model, adjusted for age, gender, minority race/ethnicity, annual income < \$15,000, diabetes duration, and insulin use

AOR adjusted odds ratio, β standardized regression coefficients

Fig. 1 Adjusted association between frequency of contact with the CarePartner and HbA1c depended on presence of an in-home support person. Graph depicts adjusted predictions of interaction with 95% confidence intervals for effect of CarePartner contact frequency on HbA1c. More frequently talking with the CarePartner associated with lower HbA1c among patients with an in-home supporter, but with higher (worse) HbA1c among patients with no in-home supporter. A similar interaction term and pattern of results for frequency of seeing the CarePartner is not shown



infrequent CarePartner contact, and therefore this finding requires confirmation in other samples.

To our knowledge, this study is the first to examine how characteristics of patients' relationships with out-of-home friends or family members, specifically, are associated with T2DM self-management and outcomes. However, our findings are consistent with research indicating that social and family support are associated with improved diabetes self-management (DiMatteo, 2004a; Glasgow & Toobert, 1988; Mayberry & Osborn, 2012; Rosland et al., 2012; Schiøtz et al., 2012) and lower diabetes distress (Baek et al., 2014; Schiøtz et al., 2012). More research on the specific types of diabetes-specific support provided by out-of-home friends and family is needed. The direct effect and deficit models focus on the structural aspects of patients' support relationships (i.e., who is available, frequency of

contact) and perceived support (e.g., emotional closeness) (Cohen & Wills, 1985; Thoits, 2011), which are reflected in our measures. However, this area of inquiry would be significantly advanced by future collection of information on functional support and received support (e.g., emotional, instrumental, informational, appraisal) from in-home versus out-of-home supporters. Prior research suggests some forms of instrumental support important for self-care may be more difficult for out-of-home supporters to provide (e.g., buying healthy foods, prepping healthy meals, exercising with the patient), but others such as sending medication reminders, facilitating medication refills, and sharing healthful recipes are relatively easy to perform from a distance (Mayberry & Osborn, 2012, 2014). Furthermore, there is some evidence that compared to in-home supporters, out-of-home supporters may be

better positioned to provide emotional support or assist patients with diabetes problem-solving with less risk of conflict (Lee et al., 2017). Future research should investigate the types of support provided by out-of-home supporters and how it might vary in the context of high and low HbA1c and in the presence or absence of an in-home supporter to expand and deepen our understanding of out-of-home support for adults with T2DM. Similarly, we did not examine the occurrence of harmful involvement (e.g., nagging, arguing, or undermining behaviors) (Mayberry & Osborn, 2012; Rosland et al., 2010, 2012) from CarePartners. Doing so may help to elucidate why more frequent contact was associated with worse HbA1c for patients without an in-home supporter.

Limitations of our approach include the use of measures collected at a single time point about a single primary CarePartner. Multiple family members and friends may be involved in patients' diabetes self-management. Patients' ratings of their emotional closeness to their CarePartner may be characteristic of their social relationships in general and not specific to the patients' primary CarePartners identified in this study. Assessment of patients' out-of-home support from multiple sources may result in more consistent associations between received support from out-of-home friends and family and patients' self-management. Additionally, we did not specifically assess whether patients' contact with their CarePartner included discussion of or help with diabetes or self-management; frequency of *diabetes-specific* interactions with the CarePartner may be more strongly associated with diabetes-related outcomes than general interactions with the CarePartner. Finally, because self-management behaviors required of adults with T2DM are many and diverse, we examined associations with multiple outcomes which can increase the likelihood of finding significant associations by chance (type 1 errors). We made analytic decisions to reduce type 1 error, including using conservative standard errors and adjusting for a priori covariates with established associations on the outcomes of interest, but the issue of multiple tests remains.

Despite these limitations, the present findings alongside others (Lee et al., 2017; Piette et al., 2010) help to clarify the potential benefits of engaging out-of-home family and friends in the self-management of adults with T2DM. A close relationship with an out-of-home supporter may help enhance self-management and reduce diabetes distress regardless of the presence of an in-home supporter. However, out-of-home supporters feel less informed about care recipients' chronic conditions and recommended regimen than in-home supporters (Lee et al., 2017; Rosland et al., 2013). This suggests that family-centered care for adults with chronic conditions may focus too narrowly on the spouse/partner despite the engagement and willingness to

help of other family members (Piette et al., 2010). Efforts to share information with designated adult children, siblings, and friends of adults with T2DM via technology [e.g., proxy on patient portals (Mayberry et al., 2011; Wolff et al., 2016) or technology-delivered communications (Burner et al., 2018; Mayberry, Berg, et al., 2016; Piette et al., 2014)] may enhance the quality of support patients' receive from out-of-home supporters to manage T2D successfully.

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Compliance with ethical standards

Conflict of interest Lindsay S. Mayberry, John D. Piette, Aaron A. Lee and James E. Aikens declare that they have no conflict of interest.

Human and animal rights and Informed consent All procedures were in accordance with the ethical standards of the institutional research committees and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all individual participants included in the study.

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