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Evaluation of off-label rapamycin use to promote healthspan in 333 adults

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Abstract Rapamycin (sirolimus) is an FDA-approved drug with immune-modulating and growth-inhibitory properties. Preclinical studies have shown that rapamycin extends lifespan and healthspan metrics in yeast, invertebrates, and rodents. Several physicians are now prescribing rapamycin off-label as a preventative therapy to maintain healthspan. Thus far, however, there is limited data available on side effects or efficacy associated with use of rapamycin in this context. To begin to address this gap in knowledge, we collected data from 333 adults with a history of off-label use of rapamycin by survey. Similar data were also collected from 172 adults who had never used rapamycin. Here, we describe the general characteristics of a patient cohort using off-label rapamycin and present initial evidence that rapamycin can be used safely in adults of normal health status.

Keywords Sirolimus · Healthspan · Rapamune · Side effects · Aging · COVID-19 · SARS-CoV-2 · Longevity

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Introduction

Geroscience is a rapidly emerging field that aims to better understand the mechanisms linking biological aging with age-related functional decline and disease [1–3]. Several hallmarks or pillars of aging have been identified that contribute to or reflect biological aging. Interventions targeting these hallmarks of aging are being developed for potential clinical application to increase healthspan and delay the onset of age-related morbidities [4–7]. The long time frames and large cohort sizes required to demonstrate efficacy in clinical trials evaluating age-related endpoints in otherwise disease-free adults are a particular challenge associated with development of novel geroscience therapies [8, 9]. A variety of strategies are being applied to address this challenge, including combining multiple age-related diseases into a single composite endpoint, development of biomarkers or biological aging "clocks" that may one day serve as surrogate endpoints, and targeting individual age-related disorders using traditional clinical trial structures.

Repurposing and off-label use of drugs already approved by the US Food and Drug Administration (FDA) has become a popular approach in translational geroscience [6, 10, 11]. Among the FDA-approved drugs being used in this way are metformin and rapamycin (sirolimus, Rapamune). Both agents have been reported to extend lifespan and healthspan in laboratory animals and are being actively tested in geroscience clinical trials. Based on early data and what appears to be a relatively modest side effect profile, some physicians have begun prescribing these medications off-label for putative healthspan-promoting benefits. While there is no concrete data on the prevalence of off-label use for such medications, metformin is likely more commonly used in this manner compared to rapamycin, both because it is a more widely known drug and is perceived as comparatively safe. Off-label use of rapamycin has been gaining increasing attention, however, due to its efficacy and reproducibility for longevity and healthspan extension in preclinical studies [12, 13].

Rapamycin is an inhibitor of the mechanistic target of rapamycin (mTOR), and has been shown to increase lifespan in laboratory organisms including yeast [14, 15], nematode worms [16], fruit flies [17, 18], and mice [19–29]. In addition to increasing lifespan, rapamycin has numerous healthspan benefits in rodents such as lower cancer incidence [20, 30], improved cognitive function [31, 32], improved kidney function [33], preservation of tendon [34], improved intestinal function and reduced gut dysbiosis [28, 35], preservation of ovarian function [36], and protection against hearing loss [37]. Transient treatment with rapamycin in middle age is sufficient to robustly extend lifespan in mice [28] and reverse a subset of age-related declines including in the heart [38–40], immune system [41], and oral cavity [42, 43]. Initial evidence supports the idea that rapamycin or rapamycin derivatives (rapalogs) may have similar beneficial effects on aging heart in companion dogs [44] and restoration of age-related declines in immune function in older adults [45, 46].

Rapamycin (used clinically under the name sirolimus) is FDA approved for kidney transplant rejection, for use in cardiac stents, and for the treatment of patients with lymphangioleiomyomatosis [47]. Dosing in this context is generally 6–15 mg loading dose followed by daily dosing of 2–5 mg/day. Side effects in these patients include dyslipidemia, mouth sores, thrombocytopenia, leukopenia, and diarrhea [48]. Rapamycin is commonly referred to as an immune suppressant [49]; however, other studies suggest that rapamycin and rapalogs also have the ability to boost at least some aspects of immune function at different doses and delivery schedules [45, 46, 50, 51]. For example, once weekly dosing of the rapalog everolimus (RAD001) at 5 mg/week for 6 weeks was found to boost influenza vaccine response in otherwise healthy older adults without significant side effects [45]. Likewise, few side effects appear to be associated with lower-dose, short-term rapamycin use in healthy adults [52], but safety concerns about off-label use of rapamycin persist within the clinical and regulatory community.

In order to begin to capture real-world data on people using rapamycin off-label for putative

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healthspan-promoting benefits, we established an online data bank where people could self-report their experiences taking rapamycin. The project was open to both current and prior users of rapamycin as well as individuals who had never used rapamycin. Between March and June 2022, a total of 505 people completed the entire survey, of whom 333 had taken rapamycin and 172 had not. Here, we provide an overview of the results obtained from this study.

Methods

Survey design and collection

The study was approved by the University of Washington Institutional Review Board (STUDY00014726). Participants were recruited to the study by direct email from participating physicians, through social media posts, via targeted recruitment within rapamycin user groups such as the Rapamycin Users Facebook page and the Rapamycin News website (https://www.rapamycin.news/), word of mouth, and the study website. Prior to accessing the survey modules, participants were required to provide a working email address and complete an Informed Consent via DocuSign. Participation in the study was available to anyone with an active email account and internet connection from March 1, 2022, through June 30, 2022.

A HIPAA-compliant web-based survey system was constructed consisting of 10 survey modules, each expected to require less than 10 min to complete. The survey modules consisted of questions designed to obtain data about participant demographics, perceived health status, rapamycin usage, medical and dental history, and overall quality of life. The modules and the order in which participants took them are shown in Table 1.

Upon completion of the Informed Consent, participants were randomly assigned a unique 6-digit identification code that was required to access the survey modules and were also provided a direct URL to access Module 1 of the survey. Upon completion of each Module, participants were automatically directed to the next Module in numerical order. If participants indicated in Module 2 that they had never taken rapamycin, they were automatically directed to Module 4 and did not complete Module 3. Participants were not required to complete all modules in one sitting and received at least two email reminders if they had completed some, but not all, modules. Only those participants who completed all of the offered modules were included in the analysis. A dedicated study coordinator was available by email to answer any questions that participants had throughout the entire process from Informed Consent through survey completion. The results presented here correspond to data from Modules 1–4 and the COVID-19 module (Module 8). Analysis of results from the other modules will be presented elsewhere.

Statistical analysis

For comparison of potentially negative health conditions experienced in the past 3 months by survey

Table 1 Description of survey modules used in the UW Rapamycin Study

Survey module	Purpose
1. Demographics	Obtain basic information about each participant
2. Rapamycin use history	Obtain information about prior use of rapamycin or other mTOR inhibitors
3. Experiences with rapamycin	Obtain participant perception on quality of life factors since taking rapamycin
4. Recent health history/potential side effects	Obtain information about perceived overall health and specific health experiences over the past 3 months that might be side effects of rapamycin use
5. 10-year reflection	Obtain participant perception of current health status compared to 10 years ago
6. Sleep quality	Obtain information about sleep quality
7. Immune status	Obtain information about immune function
8. COVID-19	Obtain information about SARS-CoV-2 exposure, infection, and severity
9. Medical health	Obtain basic information about medical care and conditions
10. Oral health	Obtain basic information about oral health care and conditions

respondents, only rapamycin users who had been taking rapamycin for at least 90 days ($n = 245$) were included in this analysis along with all non-users who completed the survey modules ($n = 172$). Chi-square tests or Fisher's exact tests (when the expected frequency of a frequency cell is less than 5) were used to compare the differences in these health conditions. False discovery rate (FDR) adjusted p-values were calculated for multiple comparisons.

The frequency and percentage of the severity of SARS-CoV-2 infection were described among non-users (Non-users), rapamycin users who only took rapamycin after recovering from infection (After only), rapamycin users who took rapamycin prior to but not during infection (Prior not during), and rapamycin users who took rapamycin continuously before, during, and after infection (Continuous). The distribution differences of the severity of COVID-19 among these groups were tested using Fisher's exact tests.

Results

A total of 505 participants successfully completed the survey modules and were included in the final data set. Of these, 333 had previously used rapamycin, while 172 had never used rapamycin. A description of the basic demographic and self-reported lifestyle characteristics of study participants is provided in Table 2.

Rapamycin users

Among rapamycin users, 77.7% (202) of men and 82.2% (60) women reported taking rapamycin under the supervision of a physician. Among the reasons given for taking rapamycin, the most common answer was "healthy longevity/anti-aging" reported by 95% (313) of users. Other responses included 62 individuals who reported taking rapamycin as a potential preventative for higher risk of dementia as a result of carrying one or more APOE4 alleles, 27 individuals selected "cardiovascular disease," 12 individuals selected "cancer," and 2 individuals reported taking rapamycin for PTEN Hamartoma Syndrome. None of the study participants reported taking rapamycin for organ transplant rejection.

Participants reported a variety of different dosing strategies (Supplemental Table 1) and lengths of rapamycin use (Table 3). By far, the most common current dosing interval among rapamycin users is once weekly dosing, with 88.1% (229) of male and 91.8% (67) of female respondents reporting using this strategy. The second most common interval for dosing was 14 days, reported by 5.8% (15) of men and 2.7% (5) of women. Only four participants (three men and one woman) reported using rapamycin daily.

Table 2 Demographic and basic lifestyle features of the study participants. For age, height, BMI, and weight, values are shown as average ± standard deviation. For other categories, values shown as percent of respondents (n). *BMI*, body mass index; *TRF*, time-restricted feeding. Δ = Change in

	Rapamycin users		Non-users	
	Men	Women	Men	Women
Gender at birth	260	73	109	63
Age (years)	61.1 ± 11.6	60.7 ± 10.5	51.8 ± 14.6	56.4 ± 12.2
Height (inches)	70.8 ± 2.6	65.0 ± 2.6	70.6 ± 2.8	65.7 ± 2.8
Current weight (lbs)	178.0 ± 29.5	136.9 ± 24.1	180.2 ± 25.8	143.1 ± 21.6
Current BMI	24.9 ± 3.6	22.8 ± 3.9	25.4 ± 3.3	23.4 ± 3.7
ΔWeight (age 25-current, lbs)	8.7 ± 23.2	8.1 ± 17.4	11.6 ± 18.5	12.3 ± 17.4
Exercise regularly	89.6% (233)	84.9% (62)	84.4% (92)	87.3% (55)
Healthy diet	97.3% (252)	93.2% (68)	97.2% (106)	98.4% (62)
Fasting/TRF	70.4% (183)	63.0% (46)	61.5% (67)	71.4% (45)
Tobacco use	1.2% (3)	0% (0)	1.8% (2)	4.8% (3)
> 2 alcoholic drinks/day	12.7% (33)	8.2% (6)	7.3% (8)	4.8% (3)
Cannabis > 1/wk	8.5% (22)	11.0% (8)	10.1% (11)	6.3% (4)
Medical exam in last 12 months	83.8% (218)	90.4% (66)	76.1% (83)	85.7% (54)
Dental exams in last 12 months	78.8% (205)	90.4% (66)	75.2% (82)	87.3% (55)

Table 3 Reported length of rapamycin usage among men and women at the time the survey was completed

Length of rapamycin usage	Women	Men
< 1 month	23.3% (17)	11.9% (31)
1–6 months	26% (19)	30.4% (79)
6 months–1 year	16.4% (12)	19.2% (50)
1–3 years	23.5% (18)	24.7% (61)
> 3 years	15.0% (7)	9.6% (39)

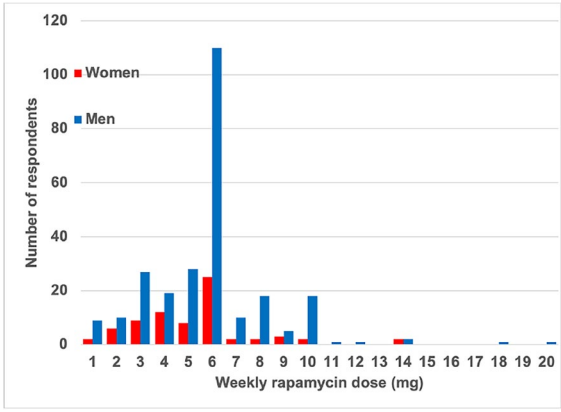

Fig. 1 Weekly dose of rapamycin usage reported by survey respondents. Dosing was determined based on self-reported usage of rapamycin. When more than one dose was reported by a single participant, the most recent dose reported was used. When dosing intervals other than weekly were reported, the dose was normalized to the equivalent weekly dose and rounded to the nearest milligram

Other dosing intervals reported by users were 5, 6, 8, 9, 10, 15, 16, and 17 days.

Similar to the case for dosing intervals, respondents reported a wide range of rapamycin doses taken. Among the individuals taking rapamycin weekly, 6 mg was the most common dose, with 101 out of 229 men and 25 out of 67 women reporting taking this dose. The minimum weekly dose for both men and women was 1 mg and the maximum weekly dose was 20 mg and 14 mg, respectively. Due to the wide range in dosing intervals and concentrations, we normalized dosing to estimate the equivalent weekly amount of rapamycin taken across all respondents rounded to the nearest milligram (Fig. 1). The median length of time on rapamycin among all survey respondents was 218 days (228.5 for men, 183 for women), with a range of 1–3890 days.

Rapamycin user experiences

Survey respondents who indicated that they had taken rapamycin were asked in Module 3 to reflect on their perception of experiences taking rapamycin. For each of the questions asked in Module 3, participants were prompted to "Please consider your overall experience with rapamycin and any changes that you associate with rapamycin. Please select the option that best describes your experiences:". The options given for each question were "Strongly Agree, Agree, Neither Agree nor Disagree, Disagree, Strongly Disagree." For questions that were gender specific, "Not Applicable" was also an option. The results of this module are shown in Table 4 with "Strongly Agree/Agree and Strongly Disagree/Disagree" shown as combined categories. The ratio of combined (Strongly)/Agree to (Strongly)/Disagree is also shown and questions are ranked according to this ratio.

Because many of the survey respondents had only recently begun taking rapamycin, a similar analysis was performed for those respondents who had reported rapamycin use for at least two months. These results are provided in Supplemental Table 2.

Recent health history

We sought to assess whether off-label use of rapamycin is associated with any significant side effects by asking both rapamycin users and non-users to recall if they had experienced any of 44 common health conditions in the past 3 months. For the purposes of this analysis, only those rapamycin users ($n = 245$) who had been taking rapamycin for longer than 3 months (the experience recall period) were included (Table 5). Similar analyses for only men ($n = 196$) and only women ($n = 49$) are provided in Supplemental Tables 4 and 5, respectively.

A total of 7 conditions were significantly different between non-users and rapamycin users. The only condition that was significantly more common in rapamycin users was presence of mouth ulceration, a known common side effect of rapamycin usage [53, 54]. Several conditions were significantly less frequent in rapamycin users compared to non-users: abdominal cramps, depression, abdominal pain, muscle tightness, anxiety, and eye pain. A trend toward a higher frequency of infections (respiratory tract, skin,

Table 4 Reported perceptions of user experiences with rapamycin. Ratio equals the percent of users who agreed divided by the percent who disagreed

	Agree	Disagree	Neither	Ratio
Rapamycin has anti-aging properties	65.5% (218)	0% (0)	34.5% (115)	-
My health has improved since taking rapamycin	44.7% (149)	5.7% (19)	49.5% (165)	7.8
I'm happier	34.8% (116)	4.8% (16)	60.4% (201)	7.3
My brain works better	35.4% (118)	5.1% (17)	59.5% (198)	6.9
I feel younger	37.5% (125)	5.7% (19)	56.8% (189)	6.6
I have more energy	38.7% (129)	6.3% (21)	55% (183)	6.1
Family/friends have commented that I look good	38.7% (129)	7.2% (24)	54.1% (180)	5.4
I feel more confident	28.8% (96)	6.3% (21)	64.9% (216)	4.6
I'm calmer and less likely to get upset or angry	25.2% (84)	7.5% (25)	67.3% (224)	3.4
My anxiety is lower	24.3% (81)	7.5% (25)	68.2% (227)	3.2
I have fewer aches and pains	35.4% (118)	12% (40)	52.6% (175)	3
It's easier to lose weight	35.1% (117)	13.2% (44)	51.7% (172)	2.7
My relationships have improved	12.6% (42)	6% (20)	81.4% (271)	2.1
I have reduced frequency of hot flashes (women)	24.4% (10)	12.2% (5)	63.4% (26)	2
I have reduced severity of hot flashes (women)	25% (10)	12.5% (5)	62.5% (25)	2
I have more erections on waking (men)	27.3% (72)	16.3% (43)	56.4% (149)	1.7
My arthritis has gotten better	18.3% (61)	10.5% (35)	71.2% (237)	1.7
I have more interest in sex	23.7% (79)	16.2% (54)	60.1% (200)	1.5
My quality of sleep improved	23.7% (79)	15.6% (52)	60.7% (202)	1.5
I have improved urinary flow (men)	20.2% (54)	16.9% (45)	62.9% (168)	1.2
I have more stronger vaginal bleeding during menstruation (women)	11.5% (3)	15.4% (4)	73.1% (19)	0.8
My vision got better	11.4% (38)	19.2% (64)	69.4% (231)	0.6
I have decreased tinnitus (ringing of the ear)	8.7% (29)	18.6% (62)	72.7% (242)	0.5
My hearing got better	6% (20)	13.5% (45)	80.5% (268)	0.4
I have increased menstrual cramping (women)	9.5% (2)	28.6% (6)	61.9% (13)	0.3

fungal, and urinary tract) among rapamycin users was noted, but did not reach statistical significance.

COVID-19

Among all study participants, 29.5% ($n=149$) reported experiencing a SARS-CoV-2 infection. Reported infection rates were similar between groups, with 31.3% ($n=54$) of non-users and 28.5% ($n=95$) of rapamycin users reporting a prior infection. Survey respondents were asked to characterize their COVID-19 case as either mild (less than 1 week of symptoms), moderate (more than 1 week of symptoms), or severe (requiring a trip to the hospital). Additionally, respondents were asked whether they suffered prolonged symptoms consistent with long-COVID.

Because many rapamycin users did not begin taking rapamycin until after their SARS-CoV-2 infection, we examined three different groups of rapamycin users and compared them to non-users. We defined 37 individuals as continuous rapamycin users based on their reported use of rapamycin continuously before, during, and after their SARS-CoV-2 infection. Seventeen individuals took rapamycin before their SARS-CoV-2 infection but stopped during the course of their infection. The remaining 41 rapamycin users did not start taking rapamycin until after their SARS-CoV-2 infection. Aggregate results are presented in Fig. 2 with percentages and number of participants in each category provided in Supplemental Table 6 (all participants), Supplemental Table 7 (men only), and Supplemental Table 8 (women only).

The 37 respondents who took rapamycin continuously before, during, and after SARS-CoV-2 infection had the lowest rate of moderate or severe infections among all groups. Only 5 individuals (13.5%) reported a moderate infection while the rest (88.5%)

Table 5 Reported frequency of potentially negative health conditions experienced in the past 3 months by survey respondents. Only rapamycin users who had been taking rapamycin for at least 90 days ($n = 245$) were included in this analysis. False discovery rate (FDR) adjusted p-values shown. Shading indicates $p < 0.05$ with green being more frequent in non-users and yellow being more frequent in rapamycin users

	All participants (n=417)	Non-users (n=172)	Rapamycin users (n=245)	FDR adj p value
Abdominal cramps	7.0%(29)	14.0%(24)	2.0%(5)	0.0000
Depression	8.6%(36)	15.1%(26)	4.1%(10)	0.0022
Abdominal pain	13.7%(57)	20.9%(36)	8.6%(21)	0.0044
Muscle tightness	36.2%(151)	45.3%(78)	29.8%(73)	0.0081
Mouth ulceration (canker sore)	10.6%(44)	4.7%(8)	14.7%(36)	0.0081
Anxiety	11.8%(49)	18.0%(31)	7.3%(18)	0.0081
Eye pain	7.0%(29)	11.6%(20)	3.7%(9)	0.0107
Nausea	5.0%(21)	8.1%(14)	2.9%(7)	0.0836
Muscle strain	26.6%(111)	32.6%(56)	22.4%(55)	0.1051
Lower back pain	37.4%(156)	43.6%(75)	33.1%(81)	0.1254
Excessive fatigue	11.8%(49)	15.7%(27)	9.0%(22)	0.1440
Respiratory tract infection	12.0%(50)	8.1%(14)	14.7%(36)	0.1516
Laceration	1.4%(6)	0.0%(0)	2.4%(6)	0.1516
Headache	39.3%(164)	44.8%(77)	35.5%(87)	0.1785
Skin infection	4.6%(19)	2.3%(4)	6.1%(15)	0.1971
Dizziness	6.7%(28)	9.3%(16)	4.9%(12)	0.2115
Cough/tonsilitis	7.7%(32)	9.9%(17)	6.1%(15)	0.4025
Musculoskeletal pain	30.7%(128)	34.3%(59)	28.2%(69)	0.4293
Abnormal vision	5.8%(24)	7.6%(13)	4.5%(11)	0.4293
Tearing	3.8%(16)	5.2%(9)	2.9%(7)	0.4704
Ear pain	5.5%(23)	7.0%(12)	4.5%(11)	0.5470
Open sores	0.7%(3)	0.0%(0)	1.2%(3)	0.5470
Urinary tract infection	2.2%(9)	1.2%(2)	2.9%(7)	0.5997
Bronchitis	3.4%(14)	2.3%(4)	4.1%(10)	0.5997
Loss of hearing	3.1%(13)	4.1%(7)	2.4%(6)	0.6134
Mouth/tooth pain	14.2%(61)	12.8%(22)	15.9%(39)	0.6322
Other fungal infection	3.1%(13)	2.3%(4)	3.7%(9)	0.7099
Skin rash	8.6%(36)	7.6%(13)	9.4%(23)	0.8054
Weight gain > than 5 lbs	5.0%(21)	5.8%(10)	4.5%(11)	0.8234
Leg cramps	29.3%(122)	27.9%(48)	30.2%(74)	0.8299
Abnormal running nose and/or sneezing	15.1%(63)	14.0%(24)	15.9%(39)	0.8299
Sore throat	11.3%(47)	12.2%(21)	10.6%(26)	0.8299
Diarrhea	22.1%(92)	23.3%(40)	21.2%(52)	0.8299
Oral herpes	3.4%(14)	2.9%(5)	3.7%(9)	0.8655
Joint pain/stiffness	55.2%(230)	55.8%(96)	54.7%(134)	0.9762
Tinnitus	22.1%(92)	22.7%(39)	21.6%(53)	0.9762
Constipation	16.3%(68)	16.9%(29)	15.9%(39)	0.9762
Other bacterial infection	2.6%(11)	2.3%(4)	2.9%(7)	0.9999
Pain in extremities	13.9%(58)	14.0%(24)	13.9%(34)	0.9999
Difficulty breathing	1.0%(4)	1.2%(2)	0.8%(2)	0.9999
Abnormal bruising	1.7%(7)	1.7%(3)	1.6%(4)	0.9999
Fall	1.7%(7)	1.7%(3)	1.6%(4)	0.9999
Fever	3.4%(14)	3.5%(6)	3.3%(8)	0.9999
Weight loss > than 5 lbs	10.8%(45)	11.0%(19)	10.6%(26)	0.9999

reported a mild case. There were no reports of hospitalization or long-COVID among this group.

Among the 17 individuals who took rapamycin prior to but not during their SARS-CoV-2 infection, 10 (58.8%) reported mild symptoms, 6 (35.3%) reported moderate symptoms, and 1 reported a (5.9%) severe case (trip to hospital). Only one of these individuals did not resume rapamycin use after SARS-CoV-2 infection. There were no reports of long-COVID among this group.

A total of 41 rapamycin users had not yet started taking rapamycin prior to or during their SARS-CoV-2 infection. Thus, this group can be considered similar to the non-users for assessing

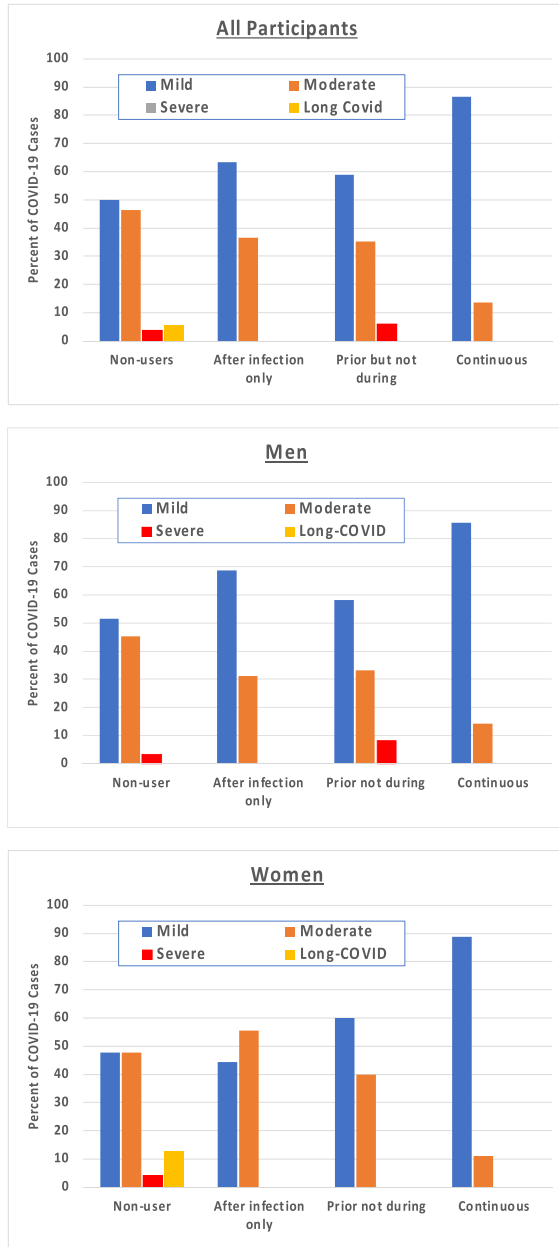

Fig. 2 Severity of COVID-19 outcomes among survey participants reporting an infection. Survey participants self-reported SARS-CoV-2 infection as mild (less than 1 week of symptoms, no hospitalization), moderate (more than 1 week of symptoms, no hospitalization), or severe (trip to hospital). Participants also noted whether they suffered from prolonged symptoms consistent with long-COVID. **A** All participants. A total of 54 individuals who have never take rapamycin are included in the "Non-users" group. Rapamycin users are split into three groups to reflect their use of rapamycin relative to the timing of SARS-CoV-2 infection. Rapamycin users who did not begin rapamycin use until after their SARS-CoV-2 infection ($n=41$) are included in the "After infection only" group. Rapamycin users who stopped taking rapamycin during their SARS-CoV-2 infection ($n=17$) are included in the "Prior but not during" group. Rapamycin users who took rapamycin continuously throughout their SARS-CoV-2 infection ($n=37$) are shown in the "Continuous" group. **B** Men only. **C** Women only. Percentages and number of participants in each group are provided in Supplemental Tables 6–8

potential impacts of rapamycin on COVID-19 severity, although rapamycin use could impact risk of long-COVID in this group. Among this group, 15 (36.6%) individuals reported a moderate infection and 26 (63.4%) reported mild symptoms. There were no reports of severe infection or long-COVID among this group.

There were a total of 54 reported SARS-CoV-2 infections among non-users. Of these, 27 (50.0%) were reported as mild, 25 (46.3%) were reported as moderate, and 2 (3.7%) were reported as severe. A total of 3 (5.6%) non-users report that they are still experiencing symptoms consistent with long-COVID. Among the individuals reporting long-COVID symptoms, one experienced a mild infection, one experienced a moderate infection, and one experienced a severe infection requiring hospitalization.

Among all survey respondents, continuous rapamycin users were significantly less likely to have experienced a moderate or severe infection or long-COVID symptoms, compared to non-users ($p<0.005$). Continuous rapamycin users were also significantly less likely to have experienced moderate or severe infection compared to rapamycin users who took rapamycin prior to but not during their infection ($p=0.039$) or those who took rapamycin only after recovering from infection ($p=0.037$). Comparisons among other groups were not statistically significant.

Discussion

Survey-based data of 333 off-label rapamycin users indicates a high perceived quality of life and good health status. No major differences in basic demographics or lifestyle features were noted between the rapamycin users and non-users. Among survey respondents, rapamycin users were on average slightly older than non-users for both men (9 years)

and women (6 years). Rapamycin users reported consuming more alcohol than non-users, and a greater percentage of rapamycin users were male (78% male) compared to non-users (63% male).

Rapamycin users generally reported perceived improvements in quality of life since beginning off-label use of rapamycin. Ratios of greater than 3:1 in agreement were observed for self-reported improvements in health, happiness, brain function, feelings of youthfulness, confidence, calmness, anxiety, and generalized aches and pains. Interestingly, greater than fivefold more rapamycin users agreed with the comment that "family/friends have commented that I look good" than disagreed, suggesting that these perceived self-benefits may also be apparent to others.

Rapamycin use by organ transplant patients is associated with a long list of potential side effects, including immune suppression, mouth sores, hypertriglyceridemia, anxiety, abdominal pain, nausea, dizziness, and many others. Organ transplant patients typically also take additional strong immunosuppressants such as tacrolimus, cyclosporine, or mycophenolate mofetil along with rapamycin or rapalogs, potentially exacerbating side effects or inducing side effects which would not be present in the absence of these drugs Interestingly, among survey respondents, only mouth sores was significantly more prevalent in rapamycin users compared to non-users. Unexpectedly, several supposed side effects associated with rapamycin use were significantly less frequent in rapamycin users, including abdominal cramps, other abdominal pain, anxiety, and eye pain. While it is difficult to know the extent to which self-reporting may have biased these results, it seems plausible that any bias could act to both increase and decrease the reported frequency of rapamycin-associated side effects. For example, rapamycin users are likely to know that mouth sores and infection are common side effects associated with rapamycin use in organ transplant patients, and therefore may be more likely to note and recall such events than non-users. Indeed, several rapamycin users reported that their prescribing physician specifically warned them to be watchful for signs of infection and to begin taking antibiotics if they suspected an infection, suggesting a heightened awareness for certain potential side effects among this group. The lack of apparent side effects associated with off-label rapamycin use here is also consistent with prior reports that once weekly administration of 5 mg of the rapamycin derivative everolimus has side effects comparable to placebo among healthy older adults [45].

Of particular interest was the analysis of COVID-19 severity among rapamycin users and non-users. Rapamycin has been proposed as a potential therapy for preventing and treating severe COVID-19 cases via its possible effects on anti-viral gene expression and attenuation of the "cytokine storm" [51, 55, 56]. No obvious difference in risk of infection was apparent between groups. Interestingly, continuous rapamycin use throughout the SARS-CoV-2 infection period was associated with a significant reduction in moderate or severe COVID-19 cases, and rapamycin users did not report any cases of long-COVID, despite being an average of 7.5 years older than non-users.

More research is needed to determine whether this represents a clinically meaningful benefit from rapamycin use for reducing severity and long-term consequences of infection with SARS-CoV-2 and other viruses. Long-COVID in particular is being recognized as having a substantial detrimental effect on quality of life for many people [57]. We speculate that the anti-inflammatory effects of rapamycin may be helpful in reducing symptoms and restoring immune homeostasis in people suffering from long-COVID. Additionally, mTOR inhibitors including everolimus (RAD001) and dactolisib (RTB101) have repeatedly shown beneficial effects on vaccine response and subsequent viral infections in otherwise healthy people in placebo-controlled clinical trials [45, 46, 51]. Given the long history of clinical use, low cost, and low risk profile associated with off-label use of rapamycin observed here, we propose that controlled clinical trials to assess benefits of rapamycin for long-COVID should be prioritized.

This study has several limitations that make the data less reliable than what would be obtained from a double-blind, randomized clinical trial. The self-reported nature of the data and the possibility of unintended bias in the participant pool reduce confidence that these results would be recapitulated in a larger, more heterogenous population. In particular, we cannot rule out the possibility that the population of rapamycin users is self-selected against people who started taking rapamycin and stopped because of negative experiences; however, we attempted to recruit as broadly as possible to include such individuals both through social media and through direct recruitment

of prior patients who had been prescribed rapamycin in the past.

It is also possible that individuals taking rapamycin off-label are more likely to practice healthy lifestyle habits or take other substances that could confound this analysis. We attempted to evaluate this and found no major differences between groups. Indeed, both rapamycin users and non-users in this study appear to be atypical in that they report higher rates of exercise and healthy dietary habits, lower body mass index, and lower rates of alcohol consumption and tobacco use, relative to the general population. It is possible that potential benefits and side effects from off-label rapamycin use would be different in a less healthy population. For example, there are several reports that rapamycin or genetic inhibition of mTOR can reduce weight gain and improve metabolic parameters in mice subjected to diet-induced obesity [58–61], but paradoxically, has negative effects in a murine genetic model of obesity caused by mutation of the leptin receptor [62].

An additional limitation of this study is that was large variation in dose and duration of rapamycin use. Approximately 20% of rapamycin users also reported taking rapamycin without the supervision of a physician. Unsupervised use of prescription medications appears to be common within the biohacking community, based on comments found in multiple online forums, and is unlikely to be unique to rapamycin. We chose not to exclude these individuals from the analyses because none of the data collected here is derived from medical records or underwent prior physician review. While it is possible that obtaining rapamycin from sources that do not require a valid prescription could influence bioavailability or purity of the product, a cursory examination of the data suggests there are no obvious differences based on prescription versus non-prescription use.

Despite these limitations, we find no evidence for significant increases in health risks, other than mouth sores, from off-label rapamycin use. A trend toward increased risk of bacterial and fungal infection was associated with rapamycin use, but did not reach statistical significance and appear to be small in magnitude. Overall, the risk of side effects from off-label use of rapamycin may compare favorably with other drugs commonly used off-label, such as statins [63] and metformin [64]. Interestingly, several positive effects were associated with off-label rapamycin use, including significant reductions in eye pain, stomach pain, anxiety, and depression relative to non-users. We also found a significant reduction in severity of COVID-19 and risk of long-COVID among respondents who took rapamycin continuously during the time of their SARS-CoV-2 infection. Rapamycin users expressed overwhelmingly positive impressions of their experiences with rapamycin. Taken together, these observations support further study of rapamycin as a potential intervention to improve quality, and perhaps quantity, of life in otherwise healthy older adults.

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Declarations

Conflict of interest MK and GH are co-founders of Optispan Geroscience, which has no material interest in rapamycin or rapamycin derivative production or sales at this time. ASG and BSR are practicing physicians who may prescribe rapamycin off-label for certain patients when appropriate. AI, AN, and SZ are employees and/or shareholders of AgelessRX, a longevity-focused telemedicine platform that sponsors several clinical trials, including interventional and observational efforts for the repurposed use of rapamycin.

References

1. Sierra F, Kohanski R. Geroscience and the trans-NIH Geroscience Interest Group, GSIG. Geroscience. 2017;39:1–5.
2. Lee MB, Kaeberlein M. Translational geroscience: from invertebrate models to companion animal and human interventions. Transl Med Aging. 2018;2:15–29.
3. Sierra F. The emergence of geroscience as an interdisciplinary approach to the enhancement of health span and life span. Cold Spring Harb Perspect Med. 2016;6: a025163.
4. Lopez-Otin C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. Cell. 2013;153:1194–217.
5. Kennedy BK, et al. Geroscience: linking aging to chronic disease. Cell. 2014;159:709–13.
6. Kulkarni AS, et al. Geroscience-guided repurposing of FDA-approved drugs to target aging: a proposed process and prioritization. Aging Cell. 2022;21: e13596.
7. Kaeberlein M, Rabinovitch PS, Martin GM. Healthy aging: the ultimate preventative medicine. Science. 2015;350:1191–3.
8. Justice JN, et al. Development of clinical trials to extend healthy lifespan. Cardiovasc Endocrinol Metab. 2018;7:80–3.
9. Wissler Gerdes EO, Misra A, Netto JME, Tchkonia T, Kirkland JL. Strategies for late phase preclinical and early clinical trials of senolytics. Mech Ageing Dev. 2021;200:111591.

10. Romashkan S, Chang H, Hadley EC. National Institute on Aging Workshop: repurposing drugs or dietary supplements for their senolytic or senomorphic effects: considerations for clinical trials. J Gerontol A Biol Sci Med Sci. 2021;76:1144–52.
11. Vaiserman A, Koliada A, Lushchak O, Castillo MJ. Repurposing drugs to fight aging: the difficult path from bench to bedside. Med Res Rev. 2021;41:1676–700.
12. Johnson SC, Rabinovitch PS, Kaeberlein M. mTOR is a key modulator of ageing and age-related disease. Nature. 2013;493:338–45.
13. Kennedy BK, Lamming DW. The mechanistic target of rapamycin: the grand conducTOR of metabolism and aging. Cell Metab. 2016;23:990–1003.
14. Powers RW 3rd, Kaeberlein M, Caldwell SD, Kennedy BK, Fields S. Extension of chronological life span in yeast by decreased TOR pathway signaling. Genes Dev. 2006;20:174–84.
15. Medvedik O, Lamming DW, Kim KD, Sinclair DA. MSN2 and MSN4 link calorie restriction and TOR to sirtuin-mediated lifespan extension in Saccharomyces cerevisiae. PLoS Biol. 2007;5: e261.
16. Robida-Stubbs S, et al. TOR signaling and rapamycin influence longevity by regulating SKN-1/Nrf and DAF-16/FoxO. Cell Metab. 2012;15:713–24.
17. Bjedov I, et al. Mechanisms of life span extension by rapamycin in the fruit fly Drosophila melanogaster. Cell Metab. 2010;11:35–46.
18. Lu YX, et al. A TORC1-histone axis regulates chromatin organisation and non-canonical induction of autophagy to ameliorate ageing. eLife. 2021;10:e62233.
19. Harrison DE, et al. Rapamycin fed late in life extends lifespan in genetically heterogeneous mice. Nature. 2009;460:392–5.
20. Anisimov VN, et al. Rapamycin increases lifespan and inhibits spontaneous tumorigenesis in inbred female mice. Cell Cycle. 2011;10:4230–6.
21. Komarova EA, et al. Rapamycin extends lifespan and delays tumorigenesis in heterozygous p53+/- mice. Aging (Albany NY). 2012;4:709–14.
22. Wilkinson JE, et al. Rapamycin slows aging in mice. Aging Cell. 2012;11:675–82.
23. Livi CB, et al. Rapamycin extends life span of Rb1+/- mice by inhibiting neuroendocrine tumors. Aging (Albany NY). 2013;5:100–10.
24. Neff F, et al. Rapamycin extends murine lifespan but has limited effects on aging. J Clin Investig. 2013;123:3272–91.
25. Fok WC, et al. Mice fed rapamycin have an increase in lifespan associated with major changes in the liver transcriptome. PLoS ONE. 2014;9: e83988.
26. Miller RA, et al. Rapamycin-mediated lifespan increase in mice is dose and sex dependent and metabolically distinct from dietary restriction. Aging Cell. 2014;13:468–77.
27. Popovich IG, et al. Lifespan extension and cancer prevention in HER-2/neu transgenic mice treated with low intermittent doses of rapamycin. Cancer Biol Ther. 2014;15:586–92.
28. Bitto A, et al. Transient rapamycin treatment can increase lifespan and healthspan in middle-aged mice. eLife. 2016;5:e16351.
29. Miller RA, et al. Rapamycin, but not resveratrol or simvastatin, extends life span of genetically heterogeneous mice. J Gerontol A Biol Sci Med Sci. 2011;66:191–201.
30. Zhang Y, et al. Rapamycin extends life and health in C57BL/6 mice. J Gerontol A Biol Sci Med Sci. 2014;69:119–30.
31. Halloran J, et al. Chronic inhibition of mammalian target of rapamycin by rapamycin modulates cognitive and non-cognitive components of behavior throughout lifespan in mice. Neuroscience. 2012;223:102–13.
32. Majumder S, et al. Lifelong rapamycin administration ameliorates age-dependent cognitive deficits by reducing IL-1beta and enhancing NMDA signaling. Aging Cell. 2012;11:326–35.
33. Shavlakadze T, et al. Short-term low-dose mTORC1 inhibition in aged rats counter-regulates age-related gene changes and blocks age-related kidney pathology. J Gerontol A Biol Sci Med Sci. 2018;73:845–52.
34. Zaseck LW, Miller RA, Brooks SV. Rapamycin attenuates age-associated changes in tibialis anterior tendon viscoelastic properties. J Gerontol A Biol Sci Med Sci. 2016;71:858–65.
35. Yilmaz OH, et al. mTORC1 in the Paneth cell niche couples intestinal stem-cell function to calorie intake. Nature. 2012;486:490–5.
36. Garcia DN, et al. Effect of caloric restriction and rapamycin on ovarian aging in mice. Geroscience. 2019;41:395–408.
37. Altschuler RA, et al. Rapamycin added to diet in late midlife delays age-related hearing loss in UMHET4 mice. Front Cell Neurosci. 2021;15: 658972.
38. Chiao YA, et al. Rapamycin transiently induces mitochondrial remodeling to reprogram energy metabolism in old hearts. Aging (Albany NY). 2016;8:314–27.
39. Dai DF, et al. Altered proteome turnover and remodeling by short-term caloric restriction or rapamycin rejuvenate the aging heart. Aging Cell. 2014;13:529–39.
40. Flynn JM, et al. Late-life rapamycin treatment reverses age-related heart dysfunction. Aging Cell. 2013;12:851–62.
41. Chen C, Liu Y, Zheng P. mTOR regulation and therapeutic rejuvenation of aging hematopoietic stem cells. Sci Signal. 2009;2:ra75.
42. An JY, et al. Rapamycin rejuvenates oral health in aging mice. eLife. 2020;9e54318.
43. An JY, et al. Rapamycin treatment attenuates age-associated periodontitis in mice. Geroscience. 2017;39:457–63.
44. Urfer SR, et al. A randomized controlled trial to establish effects of short-term rapamycin treatment in 24 middle-aged companion dogs. Geroscience. 2017;39:117–27.
45. Mannick JB, et al. mTOR inhibition improves immune function in the elderly. Sci Transl Med. 2014;6:268ra179.
46. Mannick JB, et al. TORC1 inhibition enhances immune function and reduces infections in the elderly. Sci Transl Med. 2018;10eaaq1564.
47. Kaeberlein M. mTOR inhibition: from aging to autism and beyond. Scientifica (Cairo). 2013;2013:849186.
48. Kuypers DR. Benefit-risk assessment of sirolimus in renal transplantation. Drug Saf. 2005;28:153–81.
49. Sehgal SN. Sirolimus: its discovery, biological properties, and mechanism of action. Transpl Proc. 2003;35:7S-14S.

50. Cox LS, et al. Tackling immunosenescence to improve COVID-19 outcomes and vaccine response in older adults. Lancet Healthy Longev. 2020;1:e55-e57.
51. Mannick JB, et al. Targeting the biology of ageing with mTOR inhibitors to improve immune function in older adults: phase 2b and phase 3 randomised trials. Lancet Healthy Longev. 2021;2:e250–62.
52. Kraig E, et al. A randomized control trial to establish the feasibility and safety of rapamycin treatment in an older human cohort: Immunological, physical performance, and cognitive effects. Exp Gerontol. 2018;105:53–69.
53. de Oliveira MA, et al. Clinical presentation and management of mTOR inhibitor-associated stomatitis. Oral Oncol. 2011;47:998–1003.
54. Saigal B, Guerra L. Prevention of stomatitis: using dexamethasone-based mouthwash to inhibit everolimus-related stomatitis. Clin J Oncol Nurs. 2018;22:211–7.
55. Bischof E, Siow RC, Zhavoronkov A, Kaeberlein M. The potential of rapalogs to enhance resilience against SARS-CoV-2 infection and reduce the severity of COVID-19. Lancet Healthy Longev. 2021;2:e105–11.
56. Blagosklonny MV. From causes of aging to death from COVID-19. Aging (Albany NY). 2020;12:10004–21.
57. Mehandru S, Merad M. Pathological sequelae of long-haul COVID. Nat Immunol. 2022;23:194–202.
58. Bitto A, Tatom N, Krivak T, Grotz P, Kaeberlein M. Evidence that C/EBP-beta LAP increases fat metabolism and protects against diet-induced obesity in response to mTOR inhibition. Front Aging. 2021;2: 738512.
59. Um SH, et al. Absence of S6K1 protects against age- and diet-induced obesity while enhancing insulin sensitivity. Nature. 2004;431:200–5.
60. Chang GR, et al. Rapamycin protects against high fat diet-induced obesity in C57BL/6J mice. J Pharmacol Sci. 2009;109:496–503.
61. Makki K, et al. Beneficial metabolic effects of rapamycin are associated with enhanced regulatory cells in diet-induced obese mice. PLoS ONE. 2014;9: e92684.
62. Sataranatarajan K, et al. Rapamycin increases mortality in db/db mice, a mouse model of type 2 diabetes. J Gerontol A Biol Sci Med Sci. 2016;71:850–7.
63. Jacobson TA, et al. The STatin Adverse Treatment Experience Survey: experience of patients reporting side effects of statin therapy. J Clin Lipidol. 2019;13:415–24.
64. de Vries ST, Denig P, Ekhart C, Mol PGM, van Puijenbroek EP. Sex differences in adverse drug reactions of metformin: a longitudinal survey study. Drug Saf. 2020;43:489–95.