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Exploring the relationship between polygenic risk for cannabis use, peer cannabis use, and the longitudinal course of cannabis involvement.

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Abstract

Background and aims: Few studies have explored how polygenic propensity to cannabis use unfolds across development, and no studies have yet examined this question in the context of environmental contributions such as peer cannabis use. Outlining the factors that contribute to progression from cannabis initiation to problem use over time may ultimately provide insights into mechanisms for targeted interventions. We sought to examine the relationships between polygenic liability for cannabis use, cannabis use trajectories across ages 12–30, and perceived peer cannabis use at ages 12–17.

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The authors declare no conflicts of interest.

Design: Mixed effect logistic and linear regressions were used to examine associations between polygenic risk scores, cannabis use trajectory membership, and perceived peer cannabis use.

Setting: USA

Participants: From the Collaborative Study on the Genetics of Alcoholism (COGA) study, a cohort of 1,167 individuals aged 12–26 years at their baseline (i.e., first) interview.

Measurements: Key measurements included lifetime cannabis use (yes/no), frequency of past 12-month cannabis use, maximum lifetime frequency of cannabis use, cannabis use disorder (using DSM-5 criteria), and perceived peer cannabis use. Polygenic risk scores (PRS) were created using summary statistics from a large (N = 162,082) genome-wide association study (GWAS) of cannabis use.

Findings: Three trajectories reflecting no/low (n=844), moderate (n=137) and high (n=186) use were identified. PRS were significantly associated with trajectory membership ($p=0.002 - 0.006$, maximum conditional $R^2 = 0.014$, ORs = 1.40 – 1.49). Individuals who reported that most/all of their best friends used cannabis had significantly higher PRS than those who reported that none of their friends were users (OR = 1.35, 95% C.I. = [1.04, 1.75], $p = 0.023$). Perceived peer use itself explained up to 11.3% of the variance in trajectory class membership (OR: 1.50 – 4.65). When peer cannabis use and the cannabis use PRS were entered into the model simultaneously, both the PRS and peer use continued to be significantly associated with class membership ($p < 0.01$).

Conclusions: Genetic propensity to cannabis use derived from heterogeneous samples appears to correlate with longitudinal increases in cannabis use frequency in young adults.

Introduction

The growing controversy regarding cannabis legalization in the United States¹ is based in part on the question of whether increased access is associated with escalations of both use and misuse², with the latter currently affecting about 6% of the population³. Longitudinal studies have classified young cannabis users into those who remain casual users, those who transition to moderate levels of use and remain stable, those who show initial increases followed by declines in use and, importantly, those who demonstrate accelerated use and progression to problem use^{4–11}. Outlining the factors that contribute to the likelihood of progression to problem use might provide insights into targets for intervention.

Cannabis use and misuse are heritable ($h^2=50–70\%$ of the variation). Several genomewide association studies (GWAS) have attempted to identify loci that might contribute to this heritable variation^{12–18}. For cannabis use, the largest published study to date (N = 184,765 individuals of European descent¹⁹; results used here on $n=162,082$; see Supplemental Materials for details) identified four independent genomewide significant loci and found a genomewide single nucleotide polymorphism (SNP) heritability of 10%, suggesting that the aggregated effects of common SNPs captured a sizeable portion of the heritability of cannabis use. Polygenic risk scores (PRS) offer a complementary approach to the study of such aggregated effects²⁰. In brief, a PRS is a person-specific index of genetic propensity to a trait (e.g., cannabis use); PRS are constructed by multiplying the effect size from a discovery GWAS by the number of risk alleles that an individual possesses at that SNP. PRS

approaches are widely used in psychiatric genetics, including substance use and dependence, and can be used to assess whether genetic risk for one disorder or trait is associated with aspects of the same trait, or with a correlated disorder/trait^{21,22}. For instance, one study found that PRS for schizophrenia risk predicted cannabis use in individuals with bipolar disorder²³. However, few studies have explored how genetic propensity to cannabis initiation (i.e., cannabis PRS) influences patterns of cannabis use across development.

In addition to genetic risk, affiliations with cannabis-using peers are believed to be amongst the leading contributors to persistent cannabis use^{8,11,24,25}. However, results from longitudinal samples remain mixed (e.g.,⁷). While peer use is readily viewed as an “environmental” agent of risk, it can also represent heritable aspects of underlying behavior, with at least one study suggesting a heritability of 25–28% for general peer group deviance, a broad measure including peer marijuana use²⁶. That study also found that about 50–78% of the genetic variance in peer group deviance was attributable to genetic factors related to cannabis use^{27–29}. Another study³⁰ reported that the heritability of perceived peer alcohol use ranged from 7% at age 12–14 up to 38% by age 18, and that the relationship between peer alcohol use and one’s own alcohol use was attributable to genetic factors with a correlation of 0.83. Taken together, these observations raise the possibility that polygenic risk for cannabis use may interface with peer cannabis use in several possible ways, ranging from a main effect to a potential interactive effect. To our knowledge, these hypotheses remain untested.

To better understand the role of genetic propensity and peer use in the longitudinal course of cannabis use, we used data on 1,167 individuals of European descent who were part of a large longitudinal study of the genetics of addictions. We first identified trajectories of cannabis use frequency, and then examined whether trajectory class membership was related to (a) cannabis use PRS and/or (b) perceived peer cannabis use when the subject was 12–17 years old. We also examined whether the relationship between polygenic risk, perceived peer use and trajectory membership could be explained by an interaction model where perceived peer use moderated the influence of polygenic risk on trajectory membership. Results from these analyses can provide a framework for how genetic liability and peer use might interface to shape the developmental unfolding of cannabis use.

Methods

Participants

The Collaborative Study on the Genetics of Alcoholism (COGA) study recruited alcohol dependent probands through substance use treatment programs at 7 sites across the United States. Probands and their family members were invited to participate, resulting in an overrepresentation of densely affected multiplex pedigrees. Control families (2 parents and 3 or more offspring over the age of 14) were also selected from a variety of community sources (e.g., driver license registries). The institutional review boards for all 7 data collection sites, and additional data analysis sites, approved the study³¹.

For the current analyses, data from a cohort of 3,618 individuals (“September 2017” data freeze), who were aged 12–26 years at their baseline (i.e., first) interview, and comprised the

longitudinal component of COGA, were used³². Briefly, participants were offspring of COGA families, with 61.6% having one parent with alcohol use disorder. Since 2004, participants have been interviewed every 2 years with the same structured interview; follow-up interviews are ongoing. We included only subjects with GWAS data and of European-American (EA; as verified by genotype) descent to match the ethnicity of the discovery GWAS¹³ (n=1,897); of these individuals, 1,840 had non-missing data for relevant variables. For the longitudinal growth curve analyses, a further reduction in sample size resulted from sub-setting on those who were EA, had GWAS data, and had 3 or more assessments, including the baseline assessment (final analytic n = 1,167). When compared to the larger subset of 1,840 individuals, those with 3 or more assessments did not vary of any demographic or cannabis-related characteristics, suggesting that selection for those with 3 assessments did not significantly bias findings (Table S1).

Assessment: All individuals were interviewed using a version of the Semi-Structured Assessment for the Genetics of Alcoholism (SSAGA^{33,34}) with individuals aged <18 years administered a child version (C-SSAGA^{33,34}).

Lifetime cannabis use was coded using all assessment responses to an item on whether they had ever used cannabis (response: yes or no).

Frequency of past 12 month cannabis use was also recorded as the response at each interview to an item querying how often the participant had used cannabis in the past 12 months; the range was from 0 to 935. Data were winsorized to remove outliers (>3 standard deviations) at each age and were binned into 31 categories in 20-unit intervals. The interval length was chosen to capture variation in the data and allow for model optimization (sensitivity analyses with a 10-unit interval were also conducted; see Supplemental Materials).

Maximum lifetime frequency of cannabis use was the maximum reported frequency of past 12 month cannabis use; this variable was log-transformed before analysis due to being right-skewed.

Cannabis Use Disorder (CUD) was coded using DSM-5 criteria³⁵ (without the requirement for clustering of symptoms).

Perceived peer cannabis use was coded as the response to an item: When you were 12–17, how many of your best friends used marijuana? (0 = none, 1 = a few, 2 = most, 3 = all; categories representing “most” and “all” were combined as the latter was only endorsed by 35 individuals); To minimize recall bias, peer use reported at the last assessment was used for those aged 12–17 (n=818), and at the assessment closest to age 12–17 for subjects aged 18 and older (349, although 91% were 18–21 years of age).

Genotypic data:

Members of COGA’s prospective cohort were genotyped as a part of multiple initiatives on different Illumina and Affymetrix arrays. The reported pedigree structure was assessed using

a pruned set of 1,519,440 SNPs. In total, 6,881,872 SNPs passed quality control and data cleaning thresholds and were available for analysis (details in Supplemental Materials).

Polygenic risk for cannabis use: Effect sizes and effect alleles were derived from genome-wide summary statistics from a large GWAS meta-analysis of 162,082 individuals, all of European ancestry (characteristics of discovery GWAS¹⁹ in Supplemental Materials). PRS were created for each COGA individual of genetically verified European descent with SNPs meeting increasingly lenient p-value thresholds from the discovery GWAS (from $p_T < 0.0001$ to $p_T < 0.50$). Details are provided in Supplementary Materials but briefly, for each COGA individual, effect sizes from the discovery GWAS by Pasma et al., were multiplied by the number of effect alleles for each SNP, and then averaged across all SNPs within a certain p-value threshold (e.g., $p_T < 0.10$) (e.g., tuning parameter³⁶) in the discovery GWAS to create one score per individual for that p_T . This p_T threshold is not reflective of significance of the PRS in a traditional statistical sense (i.e., $p < 0.05$). Instead, it is predicated on the assumption of a high degree of polygenicity, which has been found to be true for most complex traits³⁷; therefore, SNPs that do not reach stringent genome-wide significance cutoffs (typically $p < 5e-8$) in the discovery GWAS are still predicted to make small but incremental and additive contributions to risk liability for the outcome^{20,38,39}.

Covariates: Sex, age at first (i.e., baseline) and last interview were included as covariates. Two additional covariates were also included. First, three principal components, reflecting continuous variation in genetic ancestry were derived from all the GWAS data (details in Supplemental Materials) and included to account for subtle ancestral differences⁴⁰. Second, the type of genotyping array used for each individual was included as a covariate in order to control for potential differences in genomic content, quality control or imputation (Table S1 for descriptive data).

Statistical Analyses

Estimation of Trajectories: Only subjects with GWAS data and cannabis frequency of use data available at 3 or more assessment waves ($N=1,167$) were included in the growth mixture models. Latent class growth analysis (LCGA) with a zero-inflated Poisson model in MPlus v8⁴¹ was used to assign these individuals to classes that were derived using cannabis past year frequency of use categories from each of the up to 7 interviews (baseline through 12 - year follow-up). Age at assessment was used as the analytic unit (i.e., the x-axis). Analysis details are available in Supplemental Materials.

PRS analyses: The PRS were standardized (using the 'scale' function in R) before analysis. Mixed effect logistic regression models were used to test the association between PRS (at varying p_T) and trajectory class membership (using pairwise comparisons between classes), the associations between peer use and trajectory class membership, between the PRS and peer use and also to test whether an interaction between PRS and peer cannabis use predicted trajectory membership, while accounting for all second order interaction terms (see⁴²). All analyses included the family identifier and recruitment site as random effects (family nested within site).

All of the above analyses were conducted in R⁴³. To assess model fit and the relative amount of variance explained by the PRS, we used the ‘MuMIn’ package in R to calculate both marginal and conditional R² for each mixed model⁴³. We use the conditional R² to select the most predictive p_T (see Supplemental Materials), but report both statistics for the most predictive PRS threshold. The proportion of variance attributable to the PRS (conditional R²) was estimated as the difference between the conditional R² for a model with covariates alone and the model that included covariates and the PRS (i.e. conditional R²(full model) - conditional R²(model without PRS)). The use of R² (typically Nagelkerke’s pseudo-R² for binary traits²⁰) as an index of the most predictive PRS relates to its role as an index of predictor efficacy^{38,39,44}, such that the addition of the PRS to a model improves the fit of the model, thus indicating unique variance attributable to the PRS, over and above covariates. As peer use was restricted to recall at age 12–17, we did not test whether trajectory membership influenced future peer use. The Bonferroni-corrected significance threshold for the PRS analyses was set at 0.0019 (corrected for 27 tests: 3 class comparisons x 9 PRS thresholds), while the significance threshold was set at $\alpha < 0.05$ for the remaining analyses. In addition, to overcome concerns that uncertainty in class membership might have influenced results, we reran analyses for the most predictive PRS threshold using the BCH approach in MPlus⁴⁵. In this approach, the LCGA model is fitted to data and weights are assigned to likelihood of membership in each class while simultaneously examining between-class differences in PRS and accounting for the effect of covariates on class membership (see Supplemental Materials).

Role of externalizing behaviors: To examine whether cannabis use PRS represented a general propensity to externalizing behaviors, we examined their association with (a) the thrill/adventure-seeking and the disinhibition subscales from Zuckerman’s sensation-scale (from baseline assessments; for adults⁴⁶; Russo’s modified sensation-seeking scale for children⁴⁷; standardized) and (b) with a lifetime diagnosis of conduct disorder from the SSAGA.

Negative control analyses: As a negative control, we also tested whether the PRS significantly predicted height at baseline, a trait not expected to be genetically associated with cannabis use.

Results

Trajectories of recent cannabis use

As shown in Figure S1, three classes were identified as the three-class model had a lower Bayesian Information Criterion (BIC) than the two-class solution, the Lo-Mendel-Rubin Adjusted Likelihood Ratio Test (LMR-ALRT) p-value for the 4 class solution ($p = 0.1002$) was not significant, and the entropy for the 3 class solution (0.917) was high (fit statistics in Table S2; parameter estimates for the best-fitting model in Table S3). Classification probabilities were high (0.975, 0.928, 0.977). Sensitivity analyses with 10-unit intervals of cannabis use frequency were similar (Table S2; **Supplemental Materials**). Broadly speaking (Table 1), the classes represented (a) users who consistently used cannabis infrequently during the entire period of follow-up, and included never users of cannabis, that

we termed the “no-low” use class ($N = 844$); (b) another class that included individuals with very high frequency of initial use that continued to escalate during the follow-up period and remained elevated at the final assessment, that we termed the “high” use class ($n=186$); and (c) a class that included escalating use that involved similar high use at baseline but a less steep increase in use during the follow-up, that we termed the “moderate” use class ($N = 137$). Also, as shown in Table 1, those in the high and the moderate use trajectories were significantly more likely to be male, have used cannabis at an earlier age, and meet criteria for a lifetime history of cannabis use disorder (CUD) as well as conduct disorder.

Associations between cannabis use PRS and overall cannabis use in the sample

In the analytic sample ($n=1,167$), we found no evidence that the cannabis PRS was associated with a binary measure of lifetime cannabis use ($p = 0.111$), nor with frequency of use at baseline (in ever-users, $p = 0.390$), frequency of use at last assessment (in ever-users, $p = 0.513$), or maximum frequency of cannabis use (in ever-users, $p = 0.090$). However, the cannabis use PRS was associated with lifetime history of DSM5 CUD ($p = 0.028$) but was no longer significant in the subset of ever-users ($p = 0.090$, Table S4). The pattern of association with cannabis use was similar when individuals with <3 assessments ($n=1,840$) were studied, although in this larger sample the PRS was associated with maximum frequency of cannabis use ($p = 0.013$), and with DSM5 cannabis use disorder in both the full sample ($p = 0.005$) including in ever-users ($n = 1,144$, $p = 0.014$).

Cannabis use PRS predicting cannabis use trajectories

The cannabis use PRS was significantly associated with cannabis use trajectory class membership (Table 2). At the most significantly-associated PRS threshold of $p_T < 0.1$, the cannabis use PRS explained approximately 1.4% of the conditional variance in high vs. no-low class membership (2.30% of the marginal variance); for every unit increase in PRS, membership in the high vs. no-low class increased by an odds of 1.40 (95% CI = [1.13, 1.74]) (Figure S2; full results for all thresholds in Table 2, all covariates in Table 3). Cannabis PRS also explained 3.6% of the conditional variance in high vs. moderate class membership, although this comparison did not survive Bonferroni correction (OR = 1.49, 95% CI = [1.12, 1.97], $p = 0.006$). There was no evidence that cannabis use PRS was associated with height at baseline ($p = 0.730$). Results from the BCH approach identified identically significant differences in mean PRS across the high class when compared with the moderate and the no-low class (Table S5).

Peer cannabis use predicting cannabis use trajectories

Of the 1,162 individuals with peer use data available, 57.5, 28.3 and 14.2% reported that none, few and most-all of their close peers used cannabis. Perceived peer cannabis use explained up to 11.3% of the variance in trajectory class membership (ORs = 1.50 – 4.65). When peer cannabis use and the cannabis use PRS were entered into the model simultaneously, the association between the cannabis use PRS and membership in high vs. moderate class was only slightly attenuated (OR: 1.46 [95% C.I. = [1.09, 1.94]; $p = 0.010$), as was the association with the high vs. no-low class comparison (OR: 1.34 [95% C.I. = [1.07, 1.68]; $p = 0.012$). Peer cannabis use was independently and significantly associated

with all three comparisons in these models that also included the PRS as a predictor (both high vs. no-low and moderate vs. no-low $p < 0.001$, high vs. moderate $p = 0.017$).

Cannabis use PRS predicting peer cannabis use

Those who reported that most-all of their best friends used cannabis had significantly higher PRS than those who reported that “none” of their best friends used cannabis (most significant OR = 1.38, 95% C.I. = [1.07, 1.78], $p = 0.012$). Other comparisons (e.g., none vs. few: $p = 0.799$; few vs. most-all, $p = 0.096$) did not significantly differ from each other on cannabis PRS (details in Supplemental Material).

Role of externalizing behaviors:

The disinhibition scale score was significantly associated with both trajectory membership and all peer cannabis use comparisons, while thrill-seeking was only significantly associated with belonging to the high trajectory class vs. no-low class and with the peer cannabis use comparison between none vs. a few (details in Table S6). Cannabis use PRS did not significantly predict either scale, but was significantly associated with conduct disorder diagnosis, as were peer use and all three of the trajectory class comparisons (Table S6). The association between peer use and trajectory membership (high vs. no-low class: $p < 0.001$; high vs. moderate: $p = 0.037$; moderate vs. no-low: $p < 0.001$) was only somewhat attenuated when including conduct disorder as a covariate. Inclusion of conduct disorder also modestly attenuated the association between PRS and class membership (e.g., high vs. no-low class: OR_{conduct} = 1.37, 95% CI = [1.11, 1.69], $p = 0.003$; vs OR_{no-conduct}: 1.40, 95% CI = [1.13, 1.74], $p = 0.002$; high vs. moderate OR_{conduct} = 1.45, 95% CI = [1.10, 1.90], $p = 0.008$ vs. OR_{no-conduct} = 1.49, 95% CI = [1.12, 1.97], $p = 0.006$).

PRS x peer use predicting cannabis use trajectories

PRS-by-peer use interaction was not significant (Table S7), suggesting independent effects of PRS and peer use on trajectory membership.

Discussion

There are *three key implications* from our study. First, we found a statistically significant association between cannabis PRS and trajectory membership, and the effect size (conditional R^2 up to 3.6%) was consistent with other PRS analyses²¹. Thus, genetic propensity to cannabis initiation derived from a large, heterogeneous discovery sample appears to differentiate between classes derived from frequency of cannabis use in an ascertained, longitudinal cohort. Interestingly, lifetime cannabis use was not significantly related to PRS. However, maximum frequency of use and DSM5 CUD were associated with PRS in the larger sample of 1,840. It is possible that even though the discovery GWAS was aimed at assessing genetic propensity to lifetime use, that polygenic liability is better captured along a developmental spectrum in these data. While, to some extent, the classes differed in severity of use (e.g., CUD), associations with class membership (e.g., high vs. no-low) far exceeded cross-sectional associations with CUD, suggesting that class membership in this young and ascertained sample may be a superior index of genetic propensity than cross-sectional indices alone.

Second, the “environmental” risk factor in our study, perceived peer cannabis use explained up to 11.3% of the variance in trajectory membership. This suggests that, although genetics certainly plays a role in the progression of cannabis use, established environmental influences such as peer use are better predictors of cannabis use than PRS at the moment, and this is likely to be true for other complex behavioral traits as well. Uniquely, genetic propensity to cannabis use was also associated with greater perceived peer engagement in cannabis use. Consistent with prior heritability studies, this finding of genetic contributions to perceived peer use might reflect gene-environment correlation^{48,49} or causal processes, such as Mendelian randomization⁵⁰. However, both PRS and peer use remained significantly associated with class membership when simultaneously modeled suggesting some independent effects.

Third, we found no evidence that peer cannabis use is a moderator of polygenic contributions to cannabis use trajectories. Previous studies have found some evidence for interaction effects between peer substance use and genetic liabilities for substance use⁴⁸ but few have used genomewide PRS to do so.

Although results from the discovery GWAS for cannabis use were genetically correlated with risk-taking ($\text{SNP-}r_g = 0.425$, $p = 3.4 \times 10^{-42}$)¹⁹, we found no evidence that our measures of risk-taking were consistently related to the cannabis use PRS. Even though PRS were correlated with conduct disorder, associations between the PRS and trajectory membership persisted even after controlling for conduct disorder. Thus, general deviance does not appear to fully account for these associations.

Our study had several limitations, including a modest target sample size (target $N = 1,167$, discovery sample size $N = 162,082$; given the current sample size and a significance level of $\alpha = 0.05$, our study had 80% power⁵¹ to detect an effect size of $R^2 = 0.0068$). Further replication studies in larger, independent samples are warranted. Also, the current analyses were restricted to individuals of European ancestry, so we cannot confidently extrapolate our conclusions to other populations. Third, COGA is ascertained for genetic liability to addiction, which may have influenced findings. Our “high” group (16%) is somewhat larger than those noted in two prior general-population longitudinal studies^{6,8} but similar to one study that oversampled for tobacco⁴ smoking and lower than a study with overrepresentation of individuals from high-crime neighborhoods⁷. Thus, similar classes have been noted, although there is much variability in their class size. Fourth, while self-report of perceived peer use is commonly studied, and does not significantly differ from actual peer use⁵², it is possible that it is less objective than reports by peer nominees⁵³. Furthermore, as we did not have reports of concurrent peer cannabis use at older ages (and the sample has a diverse age range at final assessment), we cannot speculate whether trajectory membership was associated with subsequent affiliations with cannabis-using peers. Fourth, we binned frequency of use data into 20-unit intervals and this may have obscured the identification of smaller classes. For instance, our method combined those using 1–2 times in the past year with those who may have used cannabis 15–20 times. However, sensitivity analyses with 10-unit intervals provided similar results. It is also possible that reported frequency at the upper end of use was imprecise (e.g., using 550 vs. 600 times).

It is hoped that with larger discovery efforts of both cannabis use⁵⁴ and of cannabis use disorders, the predictive quality of PRS, not merely in terms of *what* they predict, but also *when* and *how* they do so, will be better elucidated. However, this study highlights that even as discovery GWAS sample sizes grow and PRS begin to attain a greater level of precision^{21,39}, it will be of paramount importance to consider not only how genetic liability shapes health and behavior, but also the environmental context within which such behavior unfolds (e.g.,⁵⁵).

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Table 1.

Characteristics of European-American individuals in classes representing high, moderate and no-low cannabis use frequency.

| | High-use class (N=186) | | Moderate-use class (N=137) | | No-Low-use class (N=844) | | High vs. No-Low OR (95% CI) | High vs. Moderate OR (95% CI) | Moderate vs. No-Low OR (95% CI) |
|--|------------------------|--------|----------------------------|--------|--------------------------|-------|--------------------------------|----------------------------------|------------------------------------|
| | Mean | SD | Mean | SD | Mean | SD | | | |
| Baseline age | 15.74 | 3.00 | 16.14 | 3.29 | 15.43 | 3.12 | 1.01 (0.96, 1.07) | 0.96 (0.89, 1.03) | 1.07 (1.00, 1.13)* |
| Age at last assessment | 23.71 | 4.07 | 25.04 | 4.05 | 23.91 | 4.38 | 0.97 (0.93, 1.01) | 0.92 (0.87, 0.98)** | 1.06 (1.01, 1.11)* |
| Age at first cannabis use | 15.08 | 2.08 | 15.98 | 1.99 | 17.87 | 2.76 | 0.61 (0.56, 0.68)** | 0.80 (0.71, 0.90)** | 0.71 (0.708, 0.715)** |
| Frequency of use at baseline | 87.66 | 153.63 | 32.12 | 89.62 | 0.72 | 4.15 | 1.07 (1.04, 1.10)** | 1.004 (1.002, 1.007)** | 1.11 (1.07, 1.15)** |
| Frequency of use at last assessment | 278.28 | 193.02 | 120.57 | 139.19 | 5.21 | 25.58 | 1.08 (1.06, 1.10)** | 1.007 (1.005, 1.009)** | 1.033 (1.026, 1.041)** |
| Male gender | % | N | % | N | % | N | OR (95% CI) | OR (95% CI) | OR (95% CI) |
| | 67.20 | 125 | 64.96 | 89 | 41.94 | 354 | 3.00 (2.99, 3.00)** | 1.10 (0.68, 1.77) | 2.90 (1.90, 4.43)** |
| Lifetime cannabis use | 100.00 | 186 | 100.00 | 137 | 51.66 | 436 | -- | -- | -- |
| Cannabis use disorder | 84.40 | 157 | 69.34 | 95 | 7.10 | 60 | 127.14 (41.19, 392.41)** | 2.45 (1.39, 4.31)** | 39.73 (19.40, 81.37)** |
| Conduct disorder diagnosis | 34.41 | 64 | 21.17 | 29 | 7.23 | 61 | 7.38 (4.61, 11.80)** | 1.96 (1.17, 3.28)* | 4.09 (2.28, 7.31)** |

* p<0.05

** p<0.01

Table 2. Results from mixed effect logistic regression models predicting cannabis use trajectory class membership by polygenic risk scores for cannabis initiation.

All models controlled for age at baseline, age at last assessment, sex, the first three ancestry principal components, and array type, and site and family id were included as nested random effects. The N SNPs column is the minimum number of SNPs included in each PRS threshold (some individuals had fewer SNPs included in the score due to missing genotypes). The PRS p-value threshold of $p_T < 0.1$ is bolded, as this PRS was the most significantly threshold associated with belonging to the high use class compared to both the no-low use class and the moderate use class, and this PRS threshold explained the most marginal variance (see Fig.S2). Thus, this PRS was used in all analyses reported in the main manuscript.

| p-value threshold (p_T) | N SNPs | Moderate class vs. no-low class | | | High class vs. no-low class | | | High class vs. moderate class | | |
|-----------------------------|---------------|---------------------------------|--------------|--------------|-----------------------------|--------------|---------------|-------------------------------|--------------|--------------|
| | | Beta | SE | p | Beta | SE | p | Beta | SE | p |
| p5 | 298,678 | 0.054 | 0.128 | 0.676 | 0.242 | 0.106 | 0.022 | 0.185 | 0.132 | 0.162 |
| p4 | 258,733 | 0.042 | 0.121 | 0.727 | 0.284 | 0.107 | 0.008 | 0.222 | 0.137 | 0.106 |
| p3 | 211,705 | 0.034 | 0.128 | 0.792 | 0.306 | 0.111 | 0.006 | 0.256 | 0.136 | 0.060 |
| p2 | 157,496 | 0.022 | 0.124 | 0.857 | 0.271 | 0.106 | 0.011 | 0.239 | 0.138 | 0.083 |
| p1^{&} | 92,504 | -0.055 | 0.121 | 0.651 | 0.339 | 0.109 | 0.0018 | 0.396 | 0.145 | 0.006 |
| p05 | 52,656 | -0.116 | 0.116 | 0.315 | 0.263 | 0.104 | 0.011 | 0.390 | 0.140 | 0.005 |
| p01 | 14,102 | -0.122 | 0.115 | 0.289 | 0.011 | 0.097 | 0.910 | 0.109 | 0.132 | 0.408 |
| p001 | 2,166 | 0.066 | 0.111 | 0.553 | -0.003 | 0.097 | 0.978 | -0.081 | 0.123 | 0.508 |
| p0001 | 372 | 0.131 | 0.111 | 0.240 | 0.076 | 0.099 | 0.445 | -0.099 | 0.132 | 0.451 |

[&]Corresponding results for PRS $p_T < 0.1$ using the BCH approach are in Supplemental Table S5.

Table 3.
Associations between cannabis initiation polygenic scores and cannabis use trajectories.

The most significant PRS is reported, which was defined with a p -value threshold of $p_T < 0.1$ (see Table 2 for results for all thresholds). Results in bold are significant predictors in the model after multiple testing corrections ($\alpha < 0.0019$).

| | Moderate vs. No-Low | | | High vs. No-Low | | | High vs. Moderate | | |
|------------------------|---------------------|--------------|-----------------|-----------------|--------------|-----------------|-------------------|--------------|-----------------|
| | Beta | SE | p | Beta | SE | p | Beta | SE | p |
| Cannabis Use PRS | -0.055 | 0.121 | 0.651 | 0.339 | 0.109 | 0.0018 | 0.396 | 0.145 | 0.006 |
| Sex | 1.117 | 0.222 | 5.03e-07 | 1.037 | 0.194 | 9.79e-08 | 0.015 | 0.258 | 0.955 |
| Age at Baseline | -0.046 | 0.065 | 0.482 | 0.140 | 0.057 | 0.014 | 0.207 | 0.078 | 0.008 |
| Age at Last Assessment | 0.101 | 0.050 | 0.042 | -0.116 | 0.043 | 0.007 | -0.225 | 0.064 | 4.12e-04 |
| Principal component 1 | -276.537 | 124.649 | 0.027 | -224.026 | 129.429 | 0.083 | -51.340 | 219.127 | 0.815 |
| Principal component 2 | -81.749 | 82.385 | 0.321 | 101.019 | 101.375 | 0.319 | 69.935 | 168.781 | 0.679 |
| Principal component 3 | -29.864 | 49.378 | 0.545 | 7.594 | 48.627 | 0.876 | 38.246 | 70.134 | 0.586 |
| Array design 1 | -0.043 | 0.260 | 0.869 | -0.505 | 0.228 | 0.027 | -0.312 | 0.279 | 0.265 |
| Array design 2 | -0.268 | 0.633 | 0.672 | -0.437 | 0.518 | 0.399 | -0.089 | 0.713 | 0.900 |

Note: Array 1 and Array 2 are two dummy-coded variables included in the model to control for the genotyping arrays. Principal components reflect genetic ancestry.

Table 4. Associations between cannabis initiation polygenic score and perceived peer cannabis use.

The PRS that was most strongly associated with cannabis use trajectories is reported ($p_T < 0.1$; see Table 2). Results in bold are significant predictors in the model ($\alpha < 0.05$).

| | None vs. A few | | | None vs. Most/All | | | A few vs. Most/All | | |
|------------------------|----------------|--------------|--------------|-------------------|----------------|------------------|--------------------|--------------|--------------|
| | Beta | SE | p | Beta | SE | p | Beta | SE | p |
| Cannabis Use PRS | 0.021 | 0.083 | 0.799 | 0.324 | 0.129 | 0.012 | 0.184 | 0.111 | 0.096 |
| Sex | 0.419 | 0.145 | 0.004 | 0.528 | 0.222 | 0.018 | 0.073 | 0.201 | 0.717 |
| Age at Baseline | 0.099 | 0.045 | 0.026 | 0.315 | 0.072 | <0.001 | 0.134 | 0.061 | 0.028 |
| Age at Last Assessment | -0.021 | 0.033 | 0.529 | -0.124 | 0.052 | 0.018 | -0.055 | 0.044 | 0.212 |
| Principal component 1 | -380.544 | 255.575 | 0.137 | -407.265 | 95.435 | <0.001 | 116.519 | 309.088 | 0.706 |
| Principal component 2 | -53.450 | 135.154 | 0.692 | -261.639 | 125.320 | 0.037 | -309.740 | 188.027 | 0.099 |
| Principal component 3 | 5.692 | 52.102 | 0.913 | 93.302 | 63.840 | 0.144 | 82.689 | 67.168 | 0.218 |
| Array design 1 | 0.365 | 0.181 | 0.043 | -0.416 | 0.269 | 0.122 | -0.631 | 0.226 | 0.005 |
| Array design 2 | 0.402 | 0.382 | 0.292 | -0.846 | 0.672 | 0.208 | -0.715 | 0.582 | 0.219 |

Note: Array 1 and Array 2 are two dummy-coded variables included in the model to control for the genotyping array types. Principal components reflect genetic ancestry.