Six-Year Demographic Study of the Terrestrial Orchid, *Crepidium acuminatum*: Implications for Conservation

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Studies on population dynamics are helpful for understanding the factors determining population development and predicting the effects of disturbances, such as harvesting of plant species. In an investigation of the demography of a terrestrial medicinal orchid known as *Crepidium acuminatum*, the effects of harvesting on its population dynamics were recorded. Data on recruitment, growth and survival were collected in three populations of *C. acuminatum* over a 6-year period (2012–2017) in central Nepal. A matrix modeling method was used to determine the effect of different harvesting regimes on the population growth and survival of this species. Population growth rates ($\lambda$) of unharvested populations were relatively similar and stable in different years of the study. Harvesting significantly reduced $\lambda$. The results of this study indicate that the sustainable survival of a population that is subject to harvesting can only occur when it is either selective (only flowering individuals or only small amounts of vegetative individuals) or rotational (once every 3–5 or more years). This study demonstrates the necessity of using a sustainable method when harvesting natural populations. Our results are useful for developing efficient management strategies for this species. As each species has a different biology, similar studies are needed for other rare and/or economically important species in the Himalayan region and in other understudied parts of the world.

**Keywords:** elasticity, Himalayan region, Orchidaceae, simulation, sustainable harvesting

**INTRODUCTION**

Collecting and analyzing long-term data on population dynamics is essential for understanding factors determining future fates of populations (Hutchings, 2010; Černá and Münzbergová, 2013; Rokaya et al., 2017; Dostálek et al., 2018; Shefferson et al., 2020). The basic information obtained from studies on population dynamics allows us to assess the present population status, its future fates and predict the effects of different factors, such as harvesting or herbivore damage, on a population's performance (Caswell, 2000a; Coates et al., 2006; Crone et al., 2013; Gamelon et al., 2017; Rokaya et al., 2017; Dostálek et al., 2018) or effects of stochastic effects such as large storms or hurricanes on population viability estimates (Crain et al., 2019). Population dynamic studies can
Crepidium acuminatum (D. Don) Szlach. (Synonym Malaxis acuminata D. Don, Malaxis biloba forma biloba (Lindl.) Tuyama, Microstylis wallichii Lind. of the family Orchidaceae is a long-lived 20–40 cm tall perennial terrestrial herbaceous plant with pseudobulbs at its base. Basal portion of shoot is covered by 1 or 2 sheathing bracts and the bases of the 3 or 4 or more elliptic-lanceolate 7–13 × 2–5 cm sized leaves. Leaf tip is acute to acuminate, sheathing at the base. Inflorescence is terminal and scape ridged. Flowers are dark purple. It is quite rare, but locally abundant and found in moist places in deciduous or subtropical forests. Flowering and fruiting are from May to July (Pearce and Cribb, 2002). In Nepal, it occurs in the central and eastern part (450–3,050 m a.s.l.) and in the rest of the world in China (Guang-dong, SW Guizhou, Taiwan, SE Xizang, S Yunnan), Cambodia, India, Indonesia, Laos, Myanmar, Philippines, Thailand and Vietnam (Raskoti, 2009; Rokaya et al., 2013). In Nepal, it is locally known as Jivak. This plant species is reproduced by means of seeds and also through propagation of pseudobulbs (Pearce and Cribb, 2002).

Pseudobulbs are used in the ayurvedic medicine as one of the ingredients of “ashtawarga” used in tonic preparations (Khare, 2015). The taste of the pseudobulbs is slightly sweet. They are used to treat fever, seminal weakness, burning sensations, excessive thirst, hematemesis or vomiting blood, bronchitis, pulmonary tuberculosis and weakness (Khare, 2007, 2015; Pant and Raskoti, 2013; Vaidya, 2019). Although it is widely used as a medicine, the exact amount traded is unknown. Mostly the whole plant, but some time only underground parts, are harvested, dried and illegally sold in the market for use in local traditional medicine and international trade. However, some traded plants are not well identified up to species level, especially when sterile individuals are harvested (Subedi et al., 2013), and may thus represent other related species.

Species Studied
Crepidium acuminatum (D. Don) Szlach. (Synonym Malaxis acuminata D. Don, Malaxis biloba forma biloba (Lindl.) Tuyama, Microstylis wallichii Lindl.) of the family Orchidaceae is a long-lived 20–40 cm tall perennial terrestrial herbaceous plant with pseudobulbs at its base. Basal portion of shoot is covered by 1 or 2 sheathing bracts and the bases of the 3 or 4 or more elliptic-lanceolate 7–13 × 2–5 cm sized leaves. Leaf tip is acute to acuminate, sheathing at the base. Inflorescence is terminal and scape ridged. Flowers are dark purple. It is quite rare, but locally abundant and found in moist places in deciduous or subtropical forests. Flowering and fruiting are from May to July (Pearce and Cribb, 2002). In Nepal, it occurs in the central and eastern part (450–3,050 m a.s.l.) and in the rest of the world in China (Guang-dong, SW Guizhou, Taiwan, SE Xizang, S Yunnan), Cambodia, India, Indonesia, Laos, Myanmar, Philippines, Thailand and Vietnam (Raskoti, 2009; Rokaya et al., 2013). In Nepal, it is locally known as Jivak. This plant species is reproduced by means of seeds and also through propagation of pseudobulbs (Pearce and Cribb, 2002).

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Plant Populations and Sampling
A demographic study of three populations of C. acuminatum was carried out on Shivapuri hill in central Nepal. All three populations were at an altitude of about 2,150 m a.s.l. in the forest...
and Population 1 and 3 were close to trails, whereas population 2 was at least 100–150 m from a trail in a deep forest.

In August 2012, we selected approximately 150–200 individuals per population (usually all plants within the population), permanently tagged them and inspected them every year in August until 2017, i.e., for 6 subsequent years. We ensured that individuals at different stages (as defined below) were marked in similar proportions (Münzbergová and Ehrlén, 2005) and also ensured that data are collected each year from 2012 to 2017 as incomplete matrices (holey matrices) may result in biologically non-sensical matrices (Tremblay et al., 2021). Every year, any newly developing plants found at each locality were also tagged. These plants were in areas where previously C. acuminatum was not recorded and very likely had germinated from seed or developed from pseudobulbs. In each census, we recorded the number of leaves, total height and status (vegetative/flowering) of each plant. Mortality of plants was also recorded the following year.

**Seedling Recruitment**

After flowering C. acuminatum produces capsules containing many minute dust-like seeds. Due to the difficulty of monitoring the germination of its dust-like seeds, we marked all the plants in specific areas in this study of its population dynamics. In addition, we marked C. acuminatum plants growing close to the areas studied in 2012 and recorded new seedlings that appeared the following year, 2013. This was repeated again each year until 2017, the end of study period. The seedling recruitment recorded in this study is the number of new seedlings that appeared each year over the total number of flowering plants the previous year. Thus, we assumed that there is germination of all seeds to seedlings (small plants) within 1 year in our species. It is also possible that some of the new individuals arise vegetatively from the pseudobulbs. While this assumption is likely not correct, it is considered as the best solution to the problem that the seeds can stay in dormancy for a variable number of years and the time of development to seedling is unknown (Shefferson, 2002; Gregg, 2011). This approach is used also in other studies on other species of orchids (Ackerman et al., 1996; García-González et al., 2017; Raventós et al., 2018).

**Data Analysis**

**Construction of Life Cycle Stages**

Using generalized linear model (GLM) with a binomial link, we looked for thresholds in plant traits (number of leaves and height of plant individuals) that differentiated between vegetative plants with low and high probability of flowering the following year. In the regression, flowering the following year (yes/no) was used as dependent variable and plant traits as independent variables. GLM analysis was carried out using glm() function in lmerTest package (Kuznetsova et al., 2017) in R 4.0.0 (R Development Core Team, 2020). We used the knowledge obtained in the tests to classify individuals into three stages, based on the number of leaves and flowering status: small vegetative (with fewer than 3 leaves and not flowering), large vegetative (more than 4 leaves and not flowering) and flowering individuals (bearing flowers, but leaves ranged from 1 to 7 in numbers, meaning that flowering plants do not always bear high number of leaves). Small vegetative plants included small plants with zero or very low probability of flowering the following year and made up approximately one-third of the tagged plants. Small vegetative plants were usually large vegetative plants the following year. Most of the large vegetative plants remain in this stage for more than one year and until they gather enough resources for flowering and producing dust-like seeds. However, there were very less flowering plant individuals and it shows that this particular species is resource limited for reproduction (Tremblay et al., 2004). In general, the plant usually dies after flowering, although it is not strictly monocarpic—it can be vegetative the next year and flower the following year. In very rare cases, it can flower in two consecutive years. We thus consider this species to be perennial and non-clonal with polycyclic bulbs (Klimešová et al., 2016).

**Comparison of Vital Rates**

We compared differences in the probability of stasis, growth, retrogression, flowering and mortality in different years and populations. For this comparison, we used logistic regression with stage in the previous year, population type and interaction of year × population as independent variables. Stasis was defined as plants surviving and remaining in the same stage, growth as development to a later stage, i.e., growth from a small vegetative to a larger vegetative or flowering plant, or large vegetative to flowering plants and retrogression as a reduction in growth from a large to a smaller stage, such as a flowering plant to a large vegetative plant. Analyses of vital rates were carried out using GLM using glm() function in lmerTest package (Kuznetsova et al., 2017) in R 4.0.0 (R Development Core Team, 2020).

**Population Dynamics**

Data on recruitment, growth and survival of individuals, classified by size, were gathered over a period of 6 years (2012–2017). Stage-based population projection matrix models were used to estimate demographic parameters (Caswell, 2000a). The model used was:

\[ n_{t+1} = An_t \]

where \( n \) is a column vector containing the number of individuals in each stage at time \( t \) or \( t+1 \), and \( A \) is a square matrix with the matrix elements representing transition probabilities between stages. Three stages were used in this study: small vegetative \((a_1)\) termed as seedling \((S)\), large vegetative \((a_2)\) termed as non-flowering adults \((V)\) and flowering individuals \((a_3)\) termed as flowering adults \((FA)\). \( a_{ij} \) indicates transitions in matrix \( A \) from stage \( j \) to stage \( i \) in 1-year time intervals. From each transition matrix constructed for each population and year (Supplementary Table 1), lambda (\( \lambda \)) was calculated, which is known as the population growth rate (Caswell, 2000a), and elasticity (Supplementary Table 1), which is usually used as a measure of the relative contribution of a matrix element to fitness (de Kroon et al., 2000). Elasticity analyses can be used to identify stages for potential management because the high elasticity of changes in vital rates will result in large changes in \( \lambda \) (Caswell, 2000b).

Both the 95% confidence intervals (CI) of \( \lambda \) and elasticity values of each transition matrix were estimated using the
rates with high elasticity values. In addition, LTRE analyses sometimes not realized, because it is difficult to change vital best management actions identified by elasticity analysis are population growth rate, as opposed to expected effects. Potential effects of single matrix elements on observed variation in the elasticity analysis, the LTRE analysis quantifies the observed growth rates and is analogous to ANOVA. Compared to the of factors responsible for the observed variation in population is a form of retrospective analysis that allows quantification contribution of each transition to the observed variation in

\[ \lambda \]

simulated two types of population growth rate, \( \lambda_s \). (1) There were five \( \lambda_s \) values for each year, which were based on years 2012–2017. (2) Overall stochastic \( \lambda_s \) values were based on a combination of data from all years 2012–2017 and populations. Simulations were performed using a MATLAB script developed in a previous study (Münzbergová, 2005). We ran the same simulations for the bootstrapped matrices as described above. Therefore, we were able to construct 95% confidence intervals for the population growth rates and elasticity (Černá and Münzbergová, 2013).

A life-table response experiment (LTRE) (Caswell, 2000a) with a fixed factorial design was conducted to examine the contribution of each transition to the observed variation in \( \lambda \) between populations in different populations growing sites. LTRE is a form of retrospective analysis that allows quantification of factors responsible for the observed variation in population growth rates and is analogous to ANOVA. Compared to the elasticity analysis, the LTRE analysis quantifies the observed effects of single matrix elements on observed variation in the population growth rate, as opposed to expected effects. Potential best management actions identified by elasticity analysis are sometimes not realized, because it is difficult to change vital rates with high elasticity values. In addition, LTRE analyses enable the identification of variable matrix elements with the highest positive or negative contributions to the population growth rate (\( \lambda \)). The significance of LTRE was estimated using the permutation test used by Münzbergová (2007) and based on 10,000 permutations. To visualize the LTRE values and compare the three populations, we also summed the positive and negative contributions for survival separately (including transitions \( a_{11}, a_{12}, a_{22}, a_{23}, a_{33} \), fecundity (\( a_{13} \)) and growth (\( a_{21}, a_{31}, a_{32} \)) and plotted these values (jongeijans and De Kroon, 2005; Münzbergová, 2013; Černá and Münzbergová, 2013; Rokaya et al., 2017).

Harvesting Simulation
To assess the effects of harvesting on \( C. acuminatum \), we produced projections of population size over 50 years under four different levels of harvesting (see below). To simulate environmental stochasticity, one of the five matrices (five transition intervals) available for each type of population was randomly selected at each simulation step. At each step, the resulting population vector (number of plants in each stage) was replaced by values drawn from a Poisson distribution with the appropriate mean, which is the standard procedure for simulating demographic stochasticity (Caswell, 2000a). This projection was repeated 1,000 times for each type of population (Münzbergová, 2005). Harvesting was simulated by removing 0, 25, 50, 75, and 100% of the natural population every 1, 3, 5, and 10 years (Ghimire et al., 2008; Rokaya et al., 2017). This was carried out in different scenarios, including removal of (i) only plants in the large vegetative and flowering stages, (ii) only large vegetative plants, (iii) only flowering plants and (iv) all plants (small and large vegetative and flowering plants). Small vegetative plants are not intentionally harvested for pseudobulbs. However, they are usually destroyed when commercial collectors randomly dig up mature plants and during this process destroy other small plant individuals too, leading to the destruction of all plant individuals and thus destruction of the whole population. Therefore, in simulation tests, we also used a scenario in which all of the plants are removed. For the initial population vector for the harvesting simulations, we used 100 plants divided according to the stable stage distribution (Caswell, 2000a) and averaged for all populations, i.e., 49 small vegetative, 36 large vegetative and 15 flowering plants. The stable stage distribution was very similar for the populations. By using the mean stable stage distribution, we ensured that all populations started under exactly the same conditions in our simulations. The MATLAB script used during the analysis is that used previously in other studies (Münzbergová, 2005, 2006). All simulations were performed using Matlab, version 6.0.0.88-The MathWorks, Inc., Natick, Massachusetts, United States (MATLAB, 2002).

RESULTS
Comparison of Vital Rates
All the vital rates (probability of stasis, growth, retrogression, flowering and mortality) tested were significantly affected by
stage in the previous year and except for growth, which varied significantly between years. Growth, flowering and mortality significantly varied among populations. The differences in flowering and mortality only varied in population 2. Individuals in population 2 flowered more than in populations 1 and 3 and mortality in population 2 was lower than in populations 1 and 3 (Table 1 and Figure 2).

**Population Growth Rates**

There was no significant variation in the population growth rate between populations and years. The population growth rates ($\lambda_S$) within 1 year varied from 0.81 to 1.05 for population 1, 0.91–1.22 for population 2 and 0.73–1.13 for population 3 over 5 transition intervals (Figure 3). Overall stochastic population growth rates ($\lambda_S$) based on a combination of data from 2012 to 2017 showed that each $\lambda$ had equal probabilities and population 1 had the lowest growth rate ($\lambda_S = 1.05$), followed by population 3 ($\lambda_S = 1.12$) and population 2 ($\lambda_S = 1.27$; see Figure 3).

**Elasticity**

The transition with the highest elasticity value for all populations was stasis of large vegetative plants ($a_{22}$) (Supplementary Table 1). Other important transitions were associated with growth ($a_{21}$ and $a_{32}$) and fecundity ($a_{13}$). There were no differences in elasticity values among populations (Supplementary Figure 1).

**Life Table Response Experiment**

According to the LTRE analyses, population growth of *C. acuminatum* in population 1 was negatively affected by fecundity, survival and growth (mainly the transition from small vegetative to large vegetative plants). Population 2 was positively affected by fecundity and survival-related transitions (mainly the high production of large vegetative plants from small vegetative and flowering from large vegetative stages), but negatively affected by growth (the transition from large vegetative plants to flowering plants). Population 3 was positively affected by fecundity, survival and growth (mainly the transition from small vegetative to large vegetative stages) (Table 2, Figure 4, and Supplementary Figure 2).

**Effect of Harvesting on Population Size**

Harvesting 25% or more of all plant individuals every year led to a significant decline in population size and increase in the probability of extinction within 30 years in all three populations. Population 3 was more vulnerable than population 1 and 2 (Figures 5, 6 and Supplementary Table 2).

The negative effect of harvesting was partly reduced when only large vegetative and flowering plants were harvested separately. The lowest extinction probabilities were achieved when only flowering plants were harvested (Figures 5, 6).

Harvesting is sustainable (i.e., the extinction probabilities for all populations in 30 years is less) when harvesting is carried out at the rate of 25% of all plant stages in a rotation of 3–5 years (Figures 5, 6 and Supplementary Table 2).

**DISCUSSION**

A long-term study of the population dynamics of *C. acuminatum* showed that the three populations studied were relatively stable with their growth rates (15) near to 1. However, all populations were seriously affected by harvesting as even in low harvesting percentages (i.e., 25% of all available plants) every year led to a significant decline in population size and increased in the probability of extinction in all populations. Our study showed that harvesting can be sustainable, if the period between harvests is long (at least 3–5 years) or when it is restricted to certain plant stages, such as small numbers of vegetative individuals or only flowering individuals.

*C. acuminatum* is capable of reproducing through bulbs and removing large individuals with fruits may not have very strong effect on seedling recruitment. It was also observed that most of the plant individuals in studied populations were in non-flowering stages meaning that flowering or seed production plays less role in population growth. Experiment related to harvesting of pseudobulb has shown death of no plants as more than 55% produced new pseudobulbs yearly in *Laelia autumnalis* orchid in Mexico (Emeterio-Lara et al., 2021).

Most of the studies on the effect of harvesting on the population dynamics of different economically important plants in the world are based only on data collected during 2–3 years (Ghimire et al., 2008; Schmidt et al., 2011; Ticktin et al., 2019). However, the previous study on medicinal plants (*Rhoeas acuminatum* and *R. australe*) in Nepal (Rokaya et al., 2017) was based on 6-year study period as the present study. Results of both studies were similar showing a significant decline in population size and increase in the probability of extinction within 30 years.
FIGURE 2 | Comparison of the vital rates of three populations. The vital rates are the probability of stasis (survival and remaining in the same stage), growth (growth to a larger stage), retrogression (shrinkage from a large to a small stage), flowering and mortality the following year. Boxes show means, standard errors and 1.96 * standard errors (defining 95% confidence interval of the mean) of the percentage of individuals undergoing a given transition. Different letters indicate significant differences in vital rates between populations (Tukey's post hoc test, \( P < 0.05 \)).

FIGURE 3 | Population growth rates and their 95% confidence intervals for three populations. The last value for each population (indicated by asterisks) shows stochastic population growth rate for all transition intervals combined (2012–2017). Dashed line (\( \lambda = 1 \)) indicates a stable population.
TABLE 2 | Results of LTRE analyses comparing the contribution of single matrix elements to the recorded variation in overall population growth rate between populations.

| From stages          | To stages          | Population 1 |          |          | Population 2 |          |          | Population 3 |          |
|---------------------|--------------------|--------------|----------|----------|--------------|----------|----------|--------------|----------|
| Small vegetative    | Large vegetative   | 0.017        | -0.013   |          | 0.630        | -0.002   |          | 0.009        | 0.015    |
| Large vegetative    | Large vegetative   | 0.068        | -0.018   |          | 0.025        | 0.020    |          | 0.814        | -0.003   |
| Large vegetative    | Flowering          | 0.554        | 0.009    |          | 0.025        | -0.033   |          | 0.170        | 0.024    |
| Flowering           | Small vegetative   | 0.310        | -0.005   |          | 0.310        | 0.002    |          | 0.310        | 0.003    |
| Flowering           | Large vegetative   | 0.573        | 0.003    |          | 0.043        | -0.009   |          | 0.237        | 0.006    |
| Flowering           | Flowering          | 0.062        | -0.014   |          | 0.887        | -0.001   |          | 0.069        | 0.015    |
| Overall             |                    | 0.016        | -0.038   |          | 0.104        | -0.023   |          | <0.001       | 0.061    |

The presented values indicate the magnitude of the contribution, with significant values (P < 0.05) shown in bold. For each factor, the contributions to the population growth rate at all levels of a given factor sum to zero. As a result, a positive contribution indicates that the transition increases overall population growth at a given level of the factor, while a negative contribution indicates that the transition decreases the overall population growth at a given level of the factor. The negative contribution values therefore indicate that a specific transition represents a weak point in the life cycle at the given level of the factor.

in all populations due to harvesting of the plant. The similarity in results in both studies is because studied species were perennial in nature and underground parts of both species were used as medicine (Khare, 2007, 2015; Pant and Raskoti, 2013; Rokaya et al., 2017; Dostálek et al., 2018; Vaidya, 2019). We focused in having a long-term study because such study can result in novel insights into the temporal variability in the overall dynamics of plant species (Oostermeijer et al., 1996; Fréville et al., 2004; Marrero-Gómez et al., 2007; Jongejans et al., 2010; Crone et al., 2011; Bucharová et al., 2012; Reineke et al., 2019).

Our study showed variability in stasis, growth, retrogression, flowering and mortality over time that depended on the stages present in previous years. There was also variability in growth, flowering and mortality among populations. These findings are similar to previous findings for other perennial plants (Münzbergová, 2005, 2013; Heiniken-Šmídová and Münzbergová, 2012; Černá and Münzbergová, 2013; Rokaya et al., 2017; Dostálek et al., 2018) and terrestrial orchids (Shefferson et al., 2020). The variability in stasis, growth, retrogression, flowering and mortality over time is mostly driven by fluctuations in climate from 1 year to the next (Gamelon et al., 2017; Shefferson et al., 2020), which can markedly affect flowering, fruiting, seed production and also seedling recruitment (Fréville et al., 2004). In addition to this, finding of variability in growth, flowering and mortality among populations is due to the presence of variation in the local environmental factors that ultimately have influence in the growth of plant individuals in different habitats (Rokaya et al., 2017).

Population growth rates (λ) for C. acuminatum in all the years were close to 1 meaning that the populations were almost stable and do not fluctuate. This is because the studied species is perennial in nature and similar results were obtained for many other perennial plant species in the world (García et al., 2008; Ramula et al., 2008; Shefferson et al., 2020). It is pointed out that stable population dynamics is a common feature of perennial species with tuberous bulbs that usually
store resources in underground parts and are as a consequence better able to withstand changing environmental conditions (García et al., 2008).

In this study, we report a high elasticity value for stasis, which is similar to that reported for epiphytic orchids in Southeast México (Mondragón, 2009) and other epiphytic angiosperms (Mondragón et al., 2015). High elasticity value for stasis, among the different vital rates (such as stasis, growth, retrogression and reproduction), means that stasis plays a vital role and contributes most to population growth rates. The retrospective (LTRE) analysis revealed both positive and negative contributions of fecundity, survival and growth to the population growth of *C. acuminatum*. From our results, we found that population 1 can be best managed by increasing fecundity, survival and growth and population 2 by increasing growth as variation in these vital rates contributed negatively to population growth rates. However, there is no need to manage population 3 as all the vital rates contributed positively (Heinken-Šmídová and Münzbergová, 2012).

This study revealed that the effects of harvesting on population dynamics of *C. acuminatum* depend on different plant stages that are harvested for economic use. Populations are negatively affected by frequent harvesting large vegetative individuals with high elasticity values, as was reported in previous studies (Ghimire et al., 2008; Rokaya et al., 2017) or by frequent harvesting all the plants would also completely eradicate the species (Rock et al., 2004; Ticktin, 2004; Lázaro-Zermeño et al., 2011; Ticktin et al., 2019). However, selective harvesting of numbers of vegetative individuals or only flowering individuals in our study species or a long-term rotational harvesting (preferably once in 3–5 years) ensures the survival of populations in their natural habitat and this practice helps to protect *C. acuminatum* from complete eradication in their habitat as in *Rheum austral* and *R. acuminatum* in Nepal (Rokaya et al., 2017). Similar to our findings, previous studies have reported that populations of perennial plants are not seriously endangered by harvesting their flowers or flowering individuals (Ticktin, 2004; Emanuel et al., 2005; Schmidt and Ticktin, 2012; Castro et al., 2015). Our study indicated that frequent harvesting of all plants is not sustainable as shown by Ticktin et al. (2019). As most of the plant individuals are in vegetative stage and this species can also reproduce through bulb, selective harvesting of only flowering

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**FIGURE 5** | Probabilities of three populations of *C. acuminatum* becoming extinct in 30 years when 25% of the individuals in all stages, only the large vegetative and flowering stages (Large veg. and flow.), only the large vegetative stage (Large vegetative) or only the flowering (Flowering) stage are harvested at different frequencies. 1, 3, 5, 10 on X-axis indicates the interval of harvesting in years.
individuals or a small number of vegetative individuals will not deplete the population and such practices will ensure the survival of the studied terrestrial orchid, *C. acuminatum*.

**Implications of Our Study in Species Conservation**

Our sampling was carried out for a long time period (for 6 years), and was quite extensive. Different demographic processes and future fates of our study species are well illustrated for less studied orchid species in Nepal. Despite the lack of field harvesting experiments, we have used simulations that are commonly used in demographic studies to show the effect of harvesting on demographic structure and population performance (Ticktin et al., 2019) and also to evaluate demographics and extinction risk of rare and endemic plants such as *Lepanthes caritensis* (Orchidaceae) (Crain et al., 2019). These simulations allow us to formulate management plans to protect plant species from getting extinct (Ghimire et al., 2008; Rokaya et al., 2017; Crain et al., 2019; Ticktin et al., 2019).

Overharvesting and random harvesting of many medicinal plant species throughout the Himalayan region has negatively affected the performance and also their existence. It is thus important to protect these plant species. As our study is in line with previous studies from Nepal (Ghimire et al., 2008; Rokaya et al., 2017; Dostálek et al., 2018) and results can serve as baseline information for other similar species that grow in the similar climatic conditions and habitat, mainly in Nepal.

**CONCLUSION**

This study revealed that although the natural populations of *C. acuminatum* studied are almost stable, they are negatively affected by harvesting. If harvesting is random or too frequent, this species may be seriously threatened or even become extinct. Thus, sustainable harvesting strategies are necessary as: (i) selective harvesting or rotation harvesting is needed to protect natural populations of *C. acuminatum* and (ii) transplantation/cultivation or tissue culture techniques should be used to protect natural populations that are over harvested or destroyed. The present research results indicate that it is possible to maintain natural populations of orchids, as reported for 15 Mexican orchids (Ticktin et al., 2019) and long-term research should be undertaken on other vulnerable species of plants, especially orchids that are highly endangered (Fay, 2018).
DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

AUTHOR CONTRIBUTIONS

BT, PK, and MR conceived the idea. BT and MR collected field data and wrote the manuscript. BT, MR, and ZM analyzed the data. PK and ZM contributed significantly to the ideas presented and edited the manuscript. All authors contributed critically to the final manuscript and approved its publication.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fevo.2021.676993/full#supplementary-material

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