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Estimating the range shift and harvesting intensity of Junipers in Bhutan (Eastern Himalaya)

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Estimating the Range Shift and Harvesting Intensity of Junipers in Bhutan (Eastern Himalaya)

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A thesis submitted to the Graduate School of the University of Missouri - St. Louis in partial fulfillment of the requirements for the degree of Master of Science in Biology (Ecology, Evolution and Systematics)

December 2017

Advisory Committee Robert J. Marquis, Ph.D. (Chairperson) Peter F. Stevens, Ph.D. Robbie Hart, Ph.D.

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ABSTRACT

The authors of more than 97% of the scientific publications on climate change agree that global temperature has increased in the last six decades and is caused by human beings via emission of greenhouse gases in the atmosphere. This phenomenon has altered natural ecosystem functions, causing many species to shift to a more suitable habitat. Throughout the world, because of recent climate warming, many forms of range shift associated with climate warming have occurred. While animals can move as warming happens, plants, being stationary organisms, cannot move, so the difference in the distribution of different plant life stages is an indication that plants are responding to climate change effects. The genus *Juniperus* is the most diverse genus in the Cupressaceae, the cypress family. Species of *Juniperus* are found in many varied climatic conditions throughout the world. Junipers in Bhutan are high altitude plant species and extensively harvested for incense manufacturing and construction purposes. The current study analyzed the elevational distribution of *J. recurva* and *J. squamata* in Bhutan along six transects, two in each of the eastern, central and western portions of Bhutan. The range of *J. squamata* is found to be shifting at the leading edge (upper elevation). The distribution of seedlings is on overage 83 m higher than that of adults at these high elevations. No differences were found in distributions at lower elevations or in the middle. *J. recurva*, in contrast, does not show comparable evidence of a range shift. The harvesting of *J. recurva* was found to be greater than that for *J. squamata*, and the intensity of harvest is positively linked to the proximity of the nearest road from sampling points. Thus, humans appear to be having an indirect effect on the distribution of *J. squamata*, via changes in climate, and a direct effect on the abundance of *J. recurva*, via harvesting of adults.

KEYWORDS: Global warming, range shift, *J. recurva*, *J. squamata*, harvesting intensity

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CHAPTER ONE: LITERATURE REVIEW

Abstract

The world is experiencing an increase in both land and ocean surface temperature caused by human beings via emission of greenhouse gases to the atmosphere. This phenomenon has caused many species to shift in their distribution to areas more favorable in climate. This range shift can be multi-dimensional in direction: toward the poles or equator (latitudinal), higher or lower elevation (altitudinal), and east or west (longitudinal). Most commonly, range shifts are detected at the leading edge of the range or the trailing edge of the range of the organism, but not east or west.

Juniper plants (*Juniperus*) are one of the most diverse group in the conifers, and are found in various climatic conditions throughout the world. The junipers found in Bhutan are high altitude species and some species of junipers in Bhutan are very local e.g., *J. recurva, J. rushforthinia* and *J. pseudosabina*, while *J. squamata* is widely distributed. Junipers in Bhutan have multiple uses, and thus are under serious threat from unregulated harvesting. Although the plant is protected under Forest and Nature Conservation Rule of Bhutan-2006 (FANCR), the harvesting is rampant and unregulated in the wild. As elsewhere, population growth in human and increasing use of resources is the biggest threat to organism survival. Thus, the fate of junipers in Bhutan looks ambiguous, unless due importance is given in understanding the factors affecting the harvesting intensity.

Key word: climate change, range shift, junipers, Bhutan, harvesting intensity

1. Climate change and range shift

1.1. Introduction

Four leading global scientific institutes (the NOAA National Climatic Data Center, the NASA Goddard Institute for Space Studies, the Japanese Meteorological Agency, and the Met Office Hadley Centre/Climatic Research Unit) have all agreed that the average global temperature in the last six decades has increased and is caused by human activities (Cook et al., 2016 & Buis, 2017) figure 1. In addition, the global organization responsible for the study of climate change, the Intergovernmental Panel on Climate Change (IPPC), has reported that climate change is real and is occurring across the globe (IPCC, 2014b). Multiple lines of evidence support these claims, including detailed observations of variability and changes in temperature of the land surface, the atmosphere, the ocean and the cryosphere. The last decade (2001 to 2010) has been the warmest of all decades (Johnson, 2014) as seen in figure 1. The authors of over 90% of all peer-reviewed science journal articles who have discussed the causes of climate change agree that current climate change is caused by human activities (Cook et al., 2016).

At the current rate of global warming, Urban 2015, predicts that one and six species will go extinct by the end of this century as a result of global climate change. There is much evidence that plant and animal range are shifting towards more suitable habitats, both altitudinally and latitudinally. While animals can move in response to climate warming, plants are stationary organisms. Phenotypic plasticity in plants may be more important than in animals as a response to climate change. Nicotra et al. (2010) reviewed the likelihood that plants might express plasticity in the face of increases in $CO₂$ and temperature.

The study of the effects of climate change on plant distribution and range shifts have focused on analyzing differences in the distribution of different life stages, particularly seedlings versus adults, across time and space. These studies generally assume that seedling distribution represents the current climate, juvenile distribution represents the climate of the most recent past, and adult distribution represents the past climate (Malis et al. 2016). Any differences in the distribution among life stages represent the effects of climate change.

1.2. Global climate change scenario

Current climate change is primarily caused by global warming, which in turn is caused by increased greenhouse gas (GHG) concentrations in the atmosphere. GHGs include carbon dioxide, water vapor, ozone, nitrous oxide and methane that are emitted by human activities like farming, burning fossil fuels, production of industrial wastes, and changing land use, particularly from increased timber harvest and logging (IPCC, 2014b). Nitrous oxide (N₂O), methane (CH₄), and carbon dioxide (CO₂) constitute a major portion of GHG concentration in the atmosphere that is causing the warming of both land and ocean surface temperature (IPCC, 2014a). The consequences are diminishing snow cover and ice volumes, rising sea levels, destruction of coral reefs, and the shift of suitable habitats distributions for plants and animals on the Earth.

The IPCC 2014 fifth assessment report projected that there will be an increase in global temperatures between 1.4°C and 5.8°C by the end of 2100 at the current rate of global warming. Plotting the earth's temperature for both ocean and land surface (Figure 1) shows a warming trend of 0.07°C per decade between 1850 and 2016, compared to average means for the period 1901 - 2000. The increase is greater in Asia at $+0.13^{\circ}$ C per decade (National Centers For Environmental Information 2017).

Figure 1: "The global average annual temperatures since 1880-2016 compared to the longterm average (1901-2000)". Source: National Centers for Environmental Information, 2017.

1.3. Climate change and biodiversity

The Biodiversity and Climate Change (2007) has published a report to commemorate the international day for biological diversity, which explicitly mentioned the impacts of climate change on biodiversity as a major concern facing the Earth now. The Millenium Ecosystem Assessment (2007) also stated that nearly 60% of ecosystems have been changed in the last 50 years so rapidly due to developmental activities taking place around the world, thereby increasing the atmospheric concentration of greenhouse gases. Of course, global biodiversity has been affected by climate change and development since the beginning of life, but the current rate of climate change boosted by human-induced activities is happening so fast that organisms are being challenged to adapt to the changed conditions where they grow now or shift their ranges to the places where environmental conditions are like those they have previously experienced.

1.4. Climate change and range shift

Grace et al. (2002) listed increasing carbon dioxide concentrations, increasing temperature, and deposition of nitrogen as the main atmospheric factors of climate change to which plants respond. Lenoir et al. (2008) and Solr (2013) stated that a small fluctuations in such variables alter an organism's range more than any changes in other set of factors combined. Other factors (secondary factors) which are less studied, and therefore less well-understood, like seed dispersal, soil type, aspect, fire, and ontogeny, are likely important and probably contribute to climate change effects and range shifts of organisms.

Climate change directly affects an individual plant's growth, alters populations and affects ecosystem structure and function (IPCC, 2014a). The joint action of many climate change effects will eventually cause a shift in the geographic and ecological ranges of plants. The shift could be toward higher or lower elevations (altitudinal), towards the pole or equator (latitudinal) or east or west (longitudinal). Woodall et al. (2008), Bell et al. (2014), and Monleon & Lintz (2015) have found that some plant species in the United States are currently moving latitudinally toward the poles at rates nearing 100 km/century. These studies were confirmed through analysis of mean changes in distribution of plant life stages along latitudinal gradients. A meta-analysis of latitudinal and elevational changes in plant distribution in Europe and North America has also found that both plants and animals are moving to higher elevations at a median rate of 11 meters/decade and to higher latitudes at a median rate of 16.9 kilometers/decade as a consequence of global climate change (Chen et al. 2011).

A plant range shift can be defined as a distributional change in one or more life stages (seedling, juvenile and adult) across latitudes, longitudes or elevations over time, aided by biotic and abiotic factors, and eventually leading to a change in the distribution of the entire species. The effect of climate change is most likely to be manifested first at the margins of species' ranges because they are very sensitive to changing climatic conditions (Cavender-Bares & Bazzaz 2000). As climate warms, it will drive seedling recruitment to a previously colder location when compared to the distribution of adult trees which represent past climates. This method of comparing the distribution of plant life stages across one or more censuses has been used by many researchers to document responses of plants to climate change (Woodall et al. 2008, Lenoir et al. 2009, Telwala et al. 2013, Bell et al. 2014, Monleon & Lintz 2015).

A literature review as of April 2014 by Lenoir & Svenning (2015) found 212 studies related to climate and range shift. They reported a range shift in at least one of the seven parameters that they studied, covering the period from 1885 onward (Table 1). Of the 212 studies, 123 were conducted on terrestrial plants and animals. Seventy-two percent ($N =$ 89) of these terrestrial studies focused on animal range shift, while 28% (N = 34) were on plants. In marine ecosystems, 91% (N = 81) of the studies focused on animals, and 9% (N=8) on plants (Lenoir & Svenning 2015).

As shown in Table 1, most studies of terrestrial plants reported an elevational range shift ($N = 24$ out of 34 studies), and next most frequent ($N = 19$) was a range shift at the leading edge, and just one study showeda longitudinal range shift. Overall, studies conducted so far have been very limited geographically: 90% (N = 140 total) of all studies, aquatic and territorial, have focused on Europe and North America (Lenoir & Svenning 2015).

Table 1. Global climate-related range shift studies segregated by ecosystem, kingdom, and range shift parameters. (LON=Longitudinal, LAT=latitudinal, E/D=Elevational/Depth, LE=Leading edge, TE=Trailing edge, O=Optimum (central), A=Abundance (uniform increased throughout the range). Source: Lenoir & Svenning, (2015)

	Terrestrial		Marine		
	Plants	Animals	Plants	Animal	
LON		8	$\mathbf{2}$	q	
LAT	3	44	5	43	
E/D	24	26		6	
LE	19	57	$\mathbf{2}$	35	
TE	11	41		25	
O	12	27		16	
A	15	44		64	
TOTAL	34	89	81	8	

1.5. Recent studies of climate change-induced range shift in trees

A study of elevational range shift by Telwala et al. (2013) for 124 endemic alpine (4000-5500 m) plant species (that include *Juniuperus recurva* and *J. indica*, the focal species of the current study) in Sikkim, India, showed that 87% of plant species have moved to a higher elevation. Historical (1849-1850) and current (2007-2010) data on climate and plant ranges, specifically those of adults, were used in a correlative analysis between the two time periods. Species migration in their study was attributed to recent climate warming. The average winter temperature in the last two centuries in the study area had increased by $0.76 \ (\pm 0.25)^{\circ}\text{C}$ in the study area. The range of *J. indica* range changed by -178 m at the lower and +28 m at the higher edges, and that of *J. recurva* moved by +5 m at lower and $+77$ m at the higher edges.

Lenoir et al. (2009) studied the 17 most common tree taxa found in the French mountain ranges ("the northern Pyrenees, the Vosges, the western Alps, the western Jura, the Corsican and the Massif central ranges") using a vegetation data bank for the period 1986-2006. The study area includes species growing in northern temperate to southern Mediterranean forest types and at elevations from lowland to sub-alpine $(50 - 2250 \text{ mas}$). Using the frequency distribution of each life stage along the elevational gradient, the differences between the life stages of plant were calculated at the $1st$, $5th$ and $9th$ deciles. They found that for 13 of the 17 species, the mean occurrence of seedlings at the 1st decile was 29 meters higher compared with that of adults along the same elevation gradient. At the $5th$ decile (optimum) the occurrence of seedlings was 69 m higher than that of adults, and at the $9th$ decile, it was not significant. They attributed this phenomenon to climate change as a primary driver coupled with secondary drivers like land abandonment, hunting of vertebrate grazers, nitrogen deposition and ontogenetic shifts (changes in the habitat that an organism needs as it develops and grows).

Bell et al. (2014) used data from forest inventories to quantify whether the climate envelope (climatic conditions suitable for growth) of six conifer tree species at two life stages (seedling and adult) showed a range shift due to recent global warming and if it differed between montane (warm climate) and subalpine (cold climate) forest types in the western United States. The species selected for the study were the most common and widely distributed tree species in western United States. They found that the distribution of life stages differed significantly between montane and subalpine species, and temperature and precipitation as climate envelope affects this variation between the species and the location. In a similar study, Monleon and Lintz (2015) examined the mean temperature ranges for seedlings and adults of 46 tree species in the three U.S. states of Washington, Oregon and California. They found that the mean temperature in areas inhabited by seedlings was 0.12° C colder than that of adults, suggesting that seedlings colonize colder areas than adults are currently living in. Thus, they concluded that seedlings have moved to significantly higher and colder areas relative to the adult life stage.

Gehrig-Fasel et al. (2007) found a significant increase in forest coverage at the tree line in the Swiss Alps, using a data set "GEOSTAT". They compared the forest coverage using GIS data between the periods (1979-1985) and (1992-1997). They found that forest cover has increased with elevation at 3% at the lowest elevation to 86% at the tree line and beyond. The increase in forest cover at tree-line was attributed to the effect of land abandonment, and the increases of forest coverage beyond tree line was attributed to recent climate warming in Swiss Alps of 2.5°C above the mean temperature since 1980.

2. Junipers-the study plant species

2.1. Introduction

Juniperus (juniper) is the largest and most diverse genus in the Cupressaceae (Adams, 2014). The genus is concentrated in the Northern Hemisphere, ranging from the deserts of Mexico, north and east to deciduous and boreal forests, and from the Mediterranean to east-central Asia (Adams, 2014). *Juniperus* species are found from sea level to above the tree line, and range in growth form from a few centimeter-high shrubs to 20-25 m tall trees. Junipers, like most conifers, are evergreens. The species can be either monoecious or dioecious. The leaves are awl-shaped or scale-like. *Juniperus* has a fleshy female cone, the cone itself called a "berry". The berries vary in color from red-brown or orange to blue-black depending on the species, and have 1 to 13 hard-shelled seeds between 4-27 mm long (Adams, 2014). These berries are consumed by birds and mammals, which help in long-distance dispersal.

2.2. Phylogeny and Evolution

The genus *Juniperus* has 75 species in 3 sections: *Caryocedrus*, *Juniperus* and *Sabina* (Adams, 2014). These sections are characterized by the morphology of seeds and cones, leaves, and their distribution. Section *Caryocedrus* has blue, woody three-seeded cones. This section has one species, *J. drupacea,* growing in the Mediterranean region (Adams, 2014)*.* Section *Juniperus* includes 14 species growing in the Eastern Hemisphere. These species have blue or red cones with three seeds (Adams, 2014). Finally, section *Sabina* has 60 species equally distributed in both the Eastern and Western Hemispheres. They have black-blue, red-copper, rose and brown cones that have 1-13 seeds (Adams, 2014).

Leaf structure differs among the sections. The sections *Caryocedrus* and *Juniperus* have acicular leaves (awl or needle-like) as shown in Figures 2A and 2B, while species of the section *Sabina* have decurrent and scale-like leaves (Figure 2C). Section *Sabina* is further divided into three clades based on morphological characteristics and supported by molecular analyses (Mao et al., 2010). These include: Clade 1. Serrate leaf margin, mostly in Northern America (Adams & Schwarzbach, 2013), Clade 2. Entire leaf margin with turbinate seed cones, single seed, mostly in Eastern Hemisphere (Adams & Schwarzbach,

2013), and Clade 3. Entire leaf margin with multiple seeds, found in both Western and Eastern Hemispheres (Adams & Schwarzbach, 2013).

Figure 2: Types of leaves in Juniperus spp. Adapted from (https://gobotany.newenglandwild.org)

A phylogeny of the *Juniperus* was generated using nuclear ribosomal DNA and chloroplast DNA (Adams & Morris, 2008) and is shown in Figure 3. The colored boxes stand for the sections and the clades. The blue rectangular boxes are the *Caryocedrus* and *Juniperus* sections, and the green for clade 1, red for clade 2 and purple for clade 3 under section *Sabina*.

Figure 3: Bayesian phylogeny tree for Juniperus. Adapted from Adams and Schwarzbach. (2013).

2.3. Ecology

Junipers are one of the most diverse genera of conifers and are found at sea level, e.g., *Juniperus lutchuensis,* and at and above the tree line, e.g., *J. monticola*. Most species prefer to grow on limestone, e.g., *J. thurifera.* However, there are species that grow in sand dunes, e.g., *J. macrocarpa,* sphagnum moss bogs, e.g., *J. communis,* glaciated granites, e.g., *J. communis*, or at the desert edge, e.g., *J. californica* (Adams, 2014)*.*

Juniperus species are pioneer and opportunistic colonizers. A key element in the evolution of *Juniperus* is its fleshy seed cones, which are eaten by birds and mammals that may help in long distance dispersal (Farjon, 2005).

2.4. Uses of Junipers

The major commercial products from *Juniperus* are cedar wood oil and gin. The oils are commonly applied to strengthen gums, improve hair growth, reduce skin irritation, relieve spasms, cure toothaches and fungal infections, and as anti-inflammatory agents (Karchesy, 1998). The berries of junipers, especially of *J. communis,* are used to brew gin. Junipers, especially in Asian Buddhist countries, are used in the manufacture of incense sticks. In Bhutan, juniper is used to make furniture and construct Dzong (fortresses) due to its durability and resistance to fungal and insect attack.

2.5. Junipers in Bhutan

Six species of *Juniperus* are recorded from Bhutan all in section sabina: *J. recurva, J. squamata, J. pseudosabina, J. communis, J. rushforthinia* and *J. indica*. They are found from 2900 masl to 4800 masl (Grierson& Long, 1983). Of the six species, *J. communis* records in Bhutan seem to be unlikely, given the known geographic distribution of this juniper in the western Himalayas(Adams,2014).

Juniperus recurva is a monoecious or rarely dioecious shrub or tree and can grow up to 20 m tall. It has decurrent drooping needle-like leaves usually 5-7 mm long appressed to the stem in whorls of three. Seed cones are bluish-black with a solitary hard seed. This species grows in open spaces as a pioneer and can be found mixed with other conifers (Adams, 2014). Its elevational range is 2900 to 4200 masl (Grierson & Long, 1983).

According to Adams (2014), its distribution range includes Afghanistan, the Himalayas (Nepal and Bhutan), China and Tibet.

Juniperus squamata is a dioecious and occasionally monoecious shrub or tree, growing up to 12 m tall. It has both needle-like and scale-like decurrent spreading leaves 6-7 mm long in whorls of three. The seed cone is black or bluish black with a solitary seed (Adams, 2014). It grows mixed with other alpine conifer species and has an elevational range of 3200 to 4700 masl (Grierson& Long, 1983).

Juniperus indica var. caesptosa is a dioecious or monoecious shrub, rarely a small tree, from 0.5 to 1 m tall height. Leaves are scale-like, and closely appressed to the stem, 3-8 mm long and borne in whorls of three. It grows near tree-line and in rocky habitats from 3600 to 4800 masl (Adams, 2014).

3. Harvesting intensity of Juniper in Bhutan

Juniperus plays an important social, cultural and religious role in Bhutan. The entire tree, including the leaves, is used for the manufacturing of incense sticks. The leaves

and branches are burnt on hilltops as a smoke offering to the local deities to bless sentient beings (Buddhist concept of all living beings on the earth that are not enlightened) so that they achieve happiness, good luck and accumulate merit. Juniper wood is considered good

properties, thus preventing it

because of its aromatic *Figure 4: Junipers harvested for construction purposes*

from insect and fungal attacks, and it is used for making altars, expensive furniture and Dzongs (Buddhist temples and administrative buildings). Juniper wood is also required as fuel wood to burn the corpse during Bhutanese funerals.

As juniper, especially *Juniperus recurva* and *J. squamata,* have many uses in Bhutan, they are seriously under threat from unregulated over-harvesting across the country. This overharvesting is occurring despite the fact that junipers are classified as class A plant species under the Forest and Nature Conservation Rules of Bhutan (2006), which in theory restricts their harvest. The IUCN red list of threatened species has listed *J. recurva* as a species of least concern in its home range (IUCN, 2017), but in Bhutan, overharvesting of the species is of major concern. (Figure 4). The Department of Forest and Parks Services is the sole authority responsible for managing forestry resources in Bhutan, but at present, the department has no record of how many junipers are harvested annually in the country nor does the Department have specific guidelines for the sustainable management of juniper forest. The most important information needed to ensure that the current stock of junipers in Bhutan are managed sustainably would be to know the immediate threat areas and the factors affecting the harvesting intensity.

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CHAPTER TWO: ESTIMATING CURRENT DISTRIBUTION AND HARVESTING INTENSITY OF *JUNIPERUS* **IN BHUTAN**

Abstract

There is much evidence that global warming is affecting the habitats of organisms throughout the world. While animals can migrate easily when warming happens, plants being stationary organisms cannot move. Differences in plant life stage distributions with altitude, latitude and longitude give an indication that plant ranges are responding to the effects of climate change. In this study, three species of *Juniperus, J. indica, J. recurva* and *J. squamata*, were sampled along six elevational transects, two transect each in the eastern, central, and western portions of Bhutan. The elevational ranges for all three species along these transects is smaller than previously reported in the literature. The range of *J. squamata* was has shifted by an average 83 m towards higher elevations, as the distribution of seedling life stage was significantly higher than that of adult life stage at the leading edge (90 percentile). There was no evidence for a range shift in *J. recurva,* and *J. indica* was too rare to determine whether its range is changing or not. Regarding harvesting intensity of junipers, both the percentage of *J. recurva* harvested and the percentage of basal area harvested in a plot increased as the distance of the nearest road to the sampling point decreased. Together, these results suggest that humans are having indirect effects on junipers in Bhutan via climate change and direct effects through harvesting.

KEYWORDS: Global warming, range shift, *Juniperus indica, Juniperus recurva*, *Juniperus squamata*, harvesting intensity

1. Introduction

The Intergovernmental Panel for Climate Change (IPCC, 2014b) predicts that at the current rate of warming, global temperature will increase between 1.4° C to 5.8° C by the end of this century. The warming is expected to be felt more in the Asian continent, the change being $+ 0.13^{\circ}$ C/decade (National Centers For Environmental Information, 2017), including the study area in Bhutan. The effects of climate change are impartial when it comes to country and environment. Bhutan, although a carbon negative country, will not be able to escape these impacts even though it is carbon negative (Dorji, 2016). Climate data for Bhutan, although not extensive, show both an increasing trend of warming and increasing precipitation over the last century, and continuing into the $21st$ century (Figure 5).

Figure 5: Annual deviation from mean annual temperature and precipitation 1901-2015 in Bhutan. (World Bank, 2017)

There is mounting evidence that plant species are shifting to higher elevations and higher latitudes, inhabiting locations previously unoccupied (Chen et al., 2011, Lenoir & Svenning, 2015). Often these shifts are associated with local climate warming (Stenseth et al., 2002, Van Bogaert et al., 2011, Rabasa et al., 2013, & Benavides et al., 2013), however shifts to higher elevations in particular might be due to changes in land use (Gehrig-Fasel et al., 2007), decreased air pollution (Koo et al., 2014), or changes in climate associated with post-Pleistocene glaciation (Davis & Shaw, 2001). Lenoir et al. (2008)

stated that distribution changes for stationary species like plants that have a long lifespan, trees in particular, are more difficult to detect compared to mobile organisms like animals. One common method used to study distributional changes in trees is to document differences in range between adults and established juveniles, either at a single point in time or across multiple censuses. The assumption of this approach is that adult distribution reflects past local climate, while the established seedling distribution, if different from that of adults, reflects a change in space in the distribution of microclimates that allow establishment. If these differences are caused by climate warming, the predictions are that established seedling distributions should be further poleward latitudinally, then that of adults at both the northern and southern boundaries of the species, and higher altitudinally at the lower and higher edges along an elevational boundary. Repeated censuses, depending on the time span between censuses and the lifespan of the tree species, should show a shift in the distribution of adult trees as well.

Compared to the number of studies conducted in Europe and North America about climate effects on range shifts of terrestrial plants, only a few have been conducted in Asia (Telwala, et al., 2013, Lenoir & Svenning, 2015 & Schickhoff et al., 2015). There are no records of such studies in Bhutan despite that fact that the geography of the country lends itself to such studies. There is a steep elevational gradient from south to north, going from 100 m to more than 7000 m in some locations (DoFPS, 2016). Many of the mountain regions are covered by conifer forests, reaching to almost 5400 m in elevation in some areas (DoFPS, 2016).

Species of the genus *Juniperus* dominate a number of these conifer forests. Both *J. recurva* and *J. indica* have moved to higher elevations in Sikkim, India (Telwala, et al., 2013). This change in distribution of *Juniperus* can be considered as an indirect consequence of human-induced climate change. In contrast, humans can also have a more direct effect on the abundance of plant species in general and *Juniperus* in particular by harvesting plants. The uses of juniper plants in Bhutan are varied. The plant is used in manufacture of incense, construction of monumental buildings, and in funerals as fuel wood. Harvesting is normally restricted, however, as this species is categorized as a class A timber in Bhutan Forest and Nature Conservation Rule-2016. Despite this, unregulated harvesting of juniper plants appears to be rampant and increasing in the forests of Bhutan.

Now is the right time to develop base line data on the current status of junipers in Bhutan, and learn the factors causing more harvesting of junipers, so that some form of regulation can be implemented to ensure sustainable harvesting in the future. Understanding the current distribution of junipers in Bhutan is very important given that we are at a cross roads both of global warming on one hand and conservation of species on other hand.

The main objectives of this study were to answer the following two questions. First, has the distribution of junipers in Bhutan been affected by recent climate change, and if so, in which direction? To answer this question, I sampled the distribution of various life stages (seedling, juvenile, and adult) of *Juniperus recurva, J. squamata,* and *J. indica* in the areas spanning the geographic range of Bhutan. I hypothesized that because plant distribution is influenced by climate, changes in climate may trigger a change in plant distribution. To test this hypothesis, I estimated the current distribution of seedlings, juveniles and adults of each juniper species along an elevational gradient in three study areas. I predicted that seedlings and juveniles would be found at higher elevation than adults, since current adult distribution would reflect the past climate and current seedling and juvenile distributions would reflect the present and recent past climate. The differences among each life stage of species distributions would reflect the shift that has occurred over time.

Second, what is the intensity of juniper harvest, and is it related to the proximity of roads to the harvested sites? To answer these questions, I estimated the basal area of trees harvested and total number of trees harvested, and correlated each with estimated proximity to the nearest road. The prediction is that the intensity of harvesting (percentage of harvest and basal area) increases as the proximity to the road increases.

2. Methodology

2.1. Study area

The study was conducted in Bhutan (27.41688° N, 90.43476° E), a small landlocked mountainous country in Asia (Figure 6). The study sites were three, representing three regions of the country, western, central and eastern, in order to sample the full range of climatic conditions where *Juniperus* species are known to occur in Bhutan. The southern region of Bhutan, where there is no record of juniper species occurring, was not sampled.

The criteria used for the selection of sites in all the three regions were: 1. All sites were on south-facing slopes as far as possible, 2. All sites were inside the mixed conifer forest type, and 3. All study sites have similar disturbance levels. These criteria were used to help ensure uniformity in environmental conditions. As the study focused on high altitude plant species, the timing of the data collection coincided with winter months in Bhutan, thus accessibility to one of the juniper growing areas in western Bhutan is restricted due to heavy snow fall. The study sites are Lamchey in the west (Figure 7), Merek in the east (Figure 8) and Yotongla in the center Bhutan (Figure 9).

Figure 6: Map of the study area (source: http://www.asia-atlas.com/bhutan.htm)

Figure 7: Study area in west Figure 8: Study area in east

2.2. Data Collection

The study was carried out between February-April 2017. At each study site, two transects were laid using Google Earth and field truthing. The total number of plots inventoried was 60 (10 plots × 2 transects × 3 study sites). The search for the *Juniperus* began at 2900 masl, the lower limits of the genus as per the flora of Bhutan (Grierson, and Long, 1983). When no junipers (and this was true for all transects) were found at 2900 masl, the next higher elevation where there were junipers served as the first plot for a given transect line. The transect line continued all the way to elevations where no junipers of any life stage were to be found, usually, that was at the tree line. On each transect line, 10 sample plots were laid out (excluding the lowest elevation where junipers are not found). The sample plots were circular with 12.62 m radius (500.34 m²) as shown in Figure 9. The distance between each sample plot was 120 meters of elevational distance, calculated based upon the range of junipers of Bhutan by Grierson and Long (1983), listed as 2900 to 4200 masl for *J. recurva* and 3200 to 4700 masl for *J. squamata*. It was found that the range of junipers, at least in this study areas was 3005 to 3620 masl for *J. recurva* and 2995 to 4091 masl for *J. squamata*. There are no historical data of juniper distribution for the study areas.

Figure 9: Lay out of sample plots and the transect in the center study area

At each sampling plot, plants with a diameter at breast height (DBH) greater than 10 cm were counted as adults, DBH greater than or equal to 5 cm but less than or equal to 10 cm as juveniles, and DBH less than 5 cm but height greater than or equal to 10 cm as seedlings. Plant diameter was measured at 1.37 m above the ground except for stumps for which diameter was measured at the highest stump height. The categorization of plants into different life stages as a seedling, juvenile and adult is based on the Bhutan national forest inventory tree classification guide. The seedling stage class likely included plants at least 3-5 years old. In addition to measuring DBH of all encountered *Juniperus* greater than 10 cm in height and counting and measuring stumps, the distance to the nearest road from the sampling point was measured in meters using the Google Earth measurement tool "ruler", which gives the actual ground distance.

Each sampled plant was identified to species, and voucher specimens of the species were collected and deposited at the National Biodiversity Center, Bhutan and the Missouri Botanical Garden, St. Louis. Grierson and Long (1983) and Adams (2014) were used for species identification.

3. Analysis

IBM SPSS statistics 24 and SAS 9.2 were used for the data analysis. To understand the current distribution of junipers in the study area, and to estimate the range shift of junipers, the data for life stages of junipers were analyzed as cumulative frequencies and percentages (relative abundance) from the raw count data. This computation was done to calculate the percentile of each life stage's current distribution with elevation. The cumulative percentages and the frequencies are used to calculate the elevation of each life stage's 10^{th} , 50^{th} and 90^{th} percentile for the current distribution of the species.

> FreqNoH−Percentilevalue qNoH−Percentilevalue = ElvH−ElvX
FreqNoH−FreqNoL = ElvH−ElvL ElvH−ElvL

where the FreqNoH and FreqNoL are the highest and the lowest frequency number corresponding to the elevation X. ElvX is the elevation that is required to calculate. Percentile value is the percentage of the total frequency number. ElvH and ElvL are the highest and lowest elevation of the corresponding frequency number. Using the formula above, the elevation of a 10^{th} , 50^{th} and 90^{th} percentile of each life stage was calculated, indicating whether there is a difference between life stages at the lower, middle and upper edge.

Wilcoxon signed rank test was used to test the hypothesis whether there is a difference between the life stages of junipers with elevation. Spearman's rho was used to test the relationship between the percentage of total harvested junipers and the percentage of total basal area harvested with the distance of sampling point to the nearest road.

4. Results

4.1. General patterns

We found three species of *Juniperus*, *J. recurva, J. squamata,* and *J. indica,* but only two species *(J. recurva* and *J. squamata)* are considered here as *J. indica* was found only in one sample plot in the study area (Figure 10). Seedlings of *J. recurva* and *J. squamata* were easily distinguished (Figure 11). The basic identification key used in the field for seedlings are; 1. Presence of parent tree species 2. Leaf and branches (color and shape)-leaves light green and branches drooping/ weeping, and leaves appressed (closed)-

J. recurva. Leaves dark green, branches upright/erect, leaves open when compared to *J. recurva- J. squamata.*

Figure 10: Juniper species in the study area (Top-J. squamata, middle- J. recurva and bottom-J. indica)

Figure 11: Seedlings of junipers (right- J. squamata and left- J. recurva)

Figure 12: Standard error line graph showing the current distribution of Junipers with elevation. A. J. recurva. B. J. squamata. *Orange lines show seedlings, blue lines the juveniles and the green lines the adults. J. recurva was not found in the West region.*

Figure 13: Median elevation at which a given proportion of individuals was encountered (triangle: 10th percentile; square: 50th percentile; circle: 90th percentile, based on the lower limit of the distribution for each life stage). Comparisons were made across life stages but within the specified distribution cutoff. Medians not sharing the same letter are significantly different at P < 0.05 (Wilcoxon signed rank test).

4.2. Range shift of *J. recurva*

There was broad elevational variation in the mean number of individuals per plot among seedlings (\bar{x} ± SE = 8.7 ± 1.2), juveniles (\bar{x} ± SE = 2.5 ± 0.6) and adults (\bar{x} ± SE = 4.0 ± 1.23) of *J. recurva* (Figure 12). The analysis using the Wilcoxon signed rank test at the $10th$, $50th$ and $90th$ percentiles for each life stage showed no significance differences between the life stages across elevations (Figure 13 and Table 2). Therefore, there is no evidence suggesting an ongoing range shift in *J. recurva* in the study areas. Based on the sampling, *J. recurva* is restricted to the central and eastern portions of the country (it was not found in any of the plots of the western transects) and has a lower elevation limit than that of *J. squamata* (Figure 12).

Life stage	10th Percentile	50th Percentile	90th Percentile
Seedling vs. Juvenile		$Z = -0.45$, $P = 1.00$ $Z = -1.00$, $P = 0.38$ $Z = -1.10$, $P = 0.38$	
Juvenile vs. Adult		$Z = -1.00$, $P = 1.00$ $Z = -1.60$, $P = 0.25$ $Z = -0.73$, $P = 0.63$	
Adult vs. Seedling		$Z = -1.34$, $P = 0.50$ $Z = -1.83$, $P = 0.13$ $Z = -1.83$, $P = 0.13$	

Table 2: Wilcoxon signed rank test results for J. recurva

4.3. Range shift of *J. squamata*

Juniperus squamata also varied greatly in the number of seedlings per plot ($\overline{x} \pm \overline{S}E$ $= 17.0 \pm 2.2$, juveniles ($\bar{x} \pm SE = 4.9 \pm 1.4$) and adults ($\bar{x} \pm SE = 5.7 \pm 1.1$) with elevation (Figure 12). Based on the results generated by Wilcoxon signed rank test, there were no significance differences between life stages at $10th$ percentile and $50th$ percentile elevation and even at the 90th percentile elevation for juvenile vs. adult and juvenile vs. seedling life stages (Table 3). But there was a significance difference for seedlings vs. the adult life stage at 90th percentile elevation. Seedlings of *J. squamata* are on average 33 m higher in elevation in the western transects, 35 m higher in center transects and 182 m higher in eastern transects than adults at the leading edge (mean elevation of seedling – mean elevation of adult at 90th percentile), which shows that the range of *J. squamata* is moving towards higher elevations (Figure 13).

Table 3: Wilcoxon signed rank test results for J. squamata

Life stage	10th Percentile	50th Percentile	90th Percentile
Seedling vs. Juvenile $Z = -0.86$, $P = 0.63$		$Z = -0.73$, $P = 0.56$ $Z = -1.15$, $P = 0.31$	
Juvenile vs. Adult	$Z = -0.73$, $P = 0.56$ $Z = -1.57$, $P = 0.57$ $Z = -0.73$, $P = 0.56$		
Adult vs. Seedling	$ Z = -0.11, P = 1.00$ $ Z = -1.79, P = 0.94$ $ Z = -2.20, P = 0.03$		

4.4. Harvesting intensity of Junipers

Among the three study areas, the center has the highest numbers of juniper trees harvested at 43.2%. The comparable figures for the eastern and western regions were 40.5% and 16.2%, respectively. Based on my sampled plots, *Juniperus recurva* is harvested more than *J. squamata* (69% vs. 31% of all harvested trees, respectively). Thus, a perceived preference by the Bhutanese for *J. recurva* for incense manufacturing, timbers for fortress construction and as firewood in funerals is apparent in the higher harvesting rate seen in this study.

The harvesting intensity of junipers as shown in Figures 14 and 15 supported the hypothesis for *J. recurva* that the proximity of roads increases the amount of harvesting.

The two-tail test of significance shows that there is a negative correlation between distance to road and percentage of total trees harvested in a plot $(r_s(40) = -0.50, p \le 0.05)$ (Figure 14) and percentage of total basal area harvested in a plot $(r_s(40) = -0.52, p \le 0.05)$ for *J*. *recurva* (Figure 15).

Regarding *J. squamata,* there was a positive, but not significant correlation (*r^s* (60) $= 0.03$, $p > 0.05$, and $r_s (60) = 0.02$, $p > 0.05$) (Fig. 16 and 17) for proximity of road affecting the percentage of total trees harvested and percentage of total basal area harvested respectively. This could be because *J. squamata* is less preferred than *J. recurva* for manufacturing of incense and constructional purposes because of its smaller size and therefore lower timber volume.

Figure 14: Scatter plot for J. recurva showing the relationship between the percentage of total harvested trees (y-axis) with the distance of road to sampling point (x-axis).

Figure 15: Scatter plot for J. recurva showing the relationship between the percentage of total basal area harvested (y-axis) with distance of road to sampling points (x-axis).

Figure 16: Scatter plot for J. squamata showing the relationship between the percentage of total harvested trees (y-axis) with the distance of road to sampling point (x-axis).

Figure 17: Scatter plot for J. squamata showing the relationship between the percentage of total basal area harvested (y-axis) with distance of road to sampling points (x-axis).

5. Discussion

The elevational range of junipers in Bhutan sampled in this study is smaller than that reported in the flora of Bhutan, i.e., 2900-4700 m (Grierson &Long, 1983). The elevational range of *Juniperus recurva* is 3005 to 3685 masl, while that of *J. squamata is* 2995 to 4091 masl in the study areas. The elevational range of these two species in other non-sampled parts of the country may be greater, as the current study focused only in three areas of Bhutan. Since this study suggests that the range and abundance of *J. recurva* is much more restricted in Bhutan than that of *J. squamata*, this is of serious concern and suggests that the former species may be under greater threat of overharvesting, given the apparent preference of the Bhutanese for *J. recurva*.

As hypotheses, global climate change, especially temperature warming and increased precipitation, in conjunction with changes in other factors, like declining grazing pressure, may have triggered the range shift of *Juniperus squamata* at the leading edge (higher elevation). In contrast, *J. recurva* appears not to be responding to such changes. However, in Sikkim, Telwala et al. (2013) found that *J. recurva* has been moving upwards at the upper edge of its range. The reasons for different responses are unclear, but in the east study sites in Bhutan, both seedlings and juveniles had a secondary peaks at 3620 m, even though there are very few adults at that altitude.

As predicted, harvesting intensity of junipers is directly related to road proximity to the sampling point. From the analysis, this holds true for *Juniperus recurva,* but not for *J. squamata*. This in fact is very much true, as the preferences of *J. recurva* over *J. squamata* for the incense manufacturing and construction purposes is comparatively high. Therefore, road building is a serious threat to the sustainable use of *J. recurva* in Bhutan. *Juniperus squamata* is harvested more at higher elevations, and usually farther away from the road because it is used for smoke offerings at high elevations, where *J. recurva* is absent, and as a fuelwood for yak herders.

6. Conclusion and recommendation

The study suggests three reasons for changes in distribution of junipers with elevation in Bhutan 1. Human-induced climate changes is a primary driver of these changes, in association with secondary biotic factors, especially the decline of grazing by yaks. Although not statistically significant, the distributions of seedlings, juveniles and adults were consistently different throughout the ranges of both species, implying that climatic conditions are affecting the distribution of both plant species particularly at mid and high elevations (Figure 12). 2. In addition, there is some anecdotal evidence from observations by people residing in the area, that there have been phenological changes in most plant species growing at higher elevations, with advanced flowering of fruit plants and successes in the cultivation of vegetables like chilies, cucumber, maize, rice, etc. at higher elevations. Cultivation of these species at high elevations was not possible 40 - 50 years ago. 3. Yak grazing is proposed here as an additional factor affecting the distribution of junipers. Yak were found grazing in high altitude grasslands of the east and there was a recently

abandoned yak hut near the tree line in the center study area. Although our methodologies were not designed to collect social data, the yak herders told us that yak grazing has drastically reduced compared to the past, as people have reduced dramatically their nomadic lifestyle. This may have helped the plant communities, including juniper seedlings, to thrive and colonize the higher elevations where grazing pressure has been reduced or is now absent.

With development, many roads are being constructed in the country. It is clear from this study that road construction is likely to increase the harvesting intensity of *J. recurva.* This phenomenon is only likely to increase as the human population continues to increase accessibility to the remote forests, putting the species under threat of over-harvesting.

As the range of *Juniperus recurva* is comparatively smaller than that of *J. squamata*, and people prefer *J. recurva* over *J. squamata*, the species is relatively under higher threat from over-harvesting. Therefore, while climate change mitigation for the conservation of species must happen at both the local and global level, at the national level, it is time to develop proper guidelines for a sustainable harvesting of junipers in the country.

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Study area	Transect No	Altitude	Species	Counts	Lifestage	
West	1	3020	J. squamata	$\overline{1}$	Juvenile	
West	1	3020	J. squamata	3	Adult	
West	1	3020	J. squamata	5	Seedling	
West	1	3140	J. squamata	5	Juvenile	
West	1	3140	J. squamata	6	Adult	
West	1	3140	J. squamata	11	Seedling	
West	1	3260	J. squamata	$\overline{2}$	Juvenile	
West	1	3260	J. squamata	10	Adult	
West	1	3260	J. squamata	9	Seedling	
West	1	3380	J. squamata	3	Juvenile	
West	1	3380	J. squamata	13	Adult	
West	1	3380	J. squamata	16	Seedling	
West	1	3500	J. squamata	$\overline{\mathbf{4}}$	Juvenile	
West	1	3500	J. squamata	11	Adult	
West	1	3500	J. squamata	17	Seedling	
West	1	3620	J. squamata	4	Juvenile	
West	1	3620	J. squamata	$\overline{7}$	Adult	
West	1	3620	J. squamata	37	Seedling	
West	1	3740	J. squamata	$\mathbf{1}$	Juvenile	
West	1	3740	J. squamata	$\overline{7}$	Adult	
West	1	3740	J. squamata	18	Seedling	
West	1	3860	J. squamata	$\overline{7}$	Juvenile	
West	1	3860	J. squamata	$\overline{\mathbf{r}}$	Adult	
West	1	3860	J. squamata	9	Seedling	
West	$\mathbf{1}$	3980	J. squamata	$\overline{2}$	Seedling	
West	$\overline{2}$	2995	J. squamata	$\overline{1}$	Juvenile	
West	$\overline{2}$	2995	J. squamata	$\overline{1}$	Adult	
West	$\mathbf{2}$	2995	J. squamata	$\overline{2}$	Seedling	
West	$\overline{2}$	3020	J. squamata	$\overline{2}$	Juvenile	
West	$\mathbf 2$	3020	J. squamata	7	Adult	
West	$\overline{2}$	3020	J. squamata	10	Seedling	
West	2	3140	J. squamata	1	Juvenile	
West	$\mathbf{2}$	3140	J. squamata	8	Adult	
West	$\overline{2}$	3140	J. squamata	9	Seedling	
West	$\overline{2}$	3260	J. squamata	$\overline{1}$	Juvenile	
West	$\mathbf{2}$	3260	J. squamata	9	Adult	
West	$\overline{2}$	3260	J. squamata	37	Seedling	
West	$\overline{2}$	3380	J. squamata	3	Juvenile	

Table 4: Life stage of junipers data from the study area

Table 5: Wilcoxon signed rank test

a. Species = *Juniperus squamata*

b. Wilcoxon Signed Ranks Test

c. Based on negative ranks.

d. Based on positive ranks.

Test Statisticsa,b

a. Species *= Juniperus recurva*

b. Wilcoxon Signed Ranks Test

c. Based on positive ranks.

Table 6: Spearman rho correlation test for harvesting intensity

**. Correlation is significant at the 0.01 level (2-tailed).

a. *Juniperus recurva*

Correlations^a

**. Correlation is significant at the 0.01 level (2-tailed).

a. *Juniperus squamata*

								Road
								to
	Study		Stump	Stump		% Basal	%	sample
Altitude	area	Species	Juvenile	Adult	Total	Area	Harvest	point
2900	West	J.recurva	0	0	0	0	0	0
2995	West	J.recurva	0	0	0	0	0	0
3020	West	J.recurva	0	0	0	0	0	0
3140	West	J.recurva	0	0	0	0	0	0
3260	West	J.recurva	0	0	0	0	0	0
3380	West	J.recurva	0	0	0	0	0	0
3500	West	J.recurva	0	0	0	0	0	$\pmb{0}$
3620	West	J.recurva	0	0	0	0	0	0
3740	West	J.recurva	0	0	0	0	0	0
3860	West	J.recurva	0	0	0	0	0	0
3980	West	J.recurva	0	0	0	0	0	$\pmb{0}$
4100	West	J.recurva	0	0	0	0	0	0
2900	West	J.recurva	0	0	0	0	0	0
2995	West	J.recurva	0	0	0	0	0	0
3020	West	J.recurva	0	0	0	0	0	$\pmb{0}$
3140	West	J.recurva	0	0	0	0	0	0
3260	West	J.recurva	0	0	0	0	0	0
3380	West	J.recurva	0	0	0	0	0	0
3500	West	J.recurva	0	0	0	0	0	$\pmb{0}$
3620	West	J.recurva	0	0	0	0	0	0
3740	West	J.recurva	0	0	0	0	0	0
3860	West	J.recurva	0	0	0	0	0	0
3980	West	J.recurva	0	0	0	0	0	$\pmb{0}$
4100	West	J.recurva	0	0	0	0	0	0
2900	Center	J.recurva	0	0	0	0	0	0
3029	Center	J.recurva	0	0	0	0	0	350
3140	Center	J.recurva	0	0	0	0	0	30
3260	Center	J.recurva	0	0	0	0	0	149
3380	Center	J.recurva	0	$\mathbf{1}$	$\mathbf{1}$	7	14	350
3500	Center	J.recurva	0	0	0	0	0	486
3620	Center	J.recurva	0	0	0	0	0	833
3740	Center	J.recurva	0	0	0	0	0	1139
3860	Center	J.recurva	0	0	0	0	0	1449
3980	Center	J.recurva	0	0	0	0	0	1865
4091	Center	J.recurva	0	0	0	0	0	2499
4100	Center	J.recurva	0	0	0	0	0	0
2900	Center	J.recurva	0	0	0	0	0	0
3029	Center	J.recurva	0	$\mathbf 1$	$\mathbf{1}$	44	10	470

Table 8: Percentage of harvested and basal area of junipers and road distance to sample points

