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The geographic distribution, variations and floristic associations of wild Cruciferae: *Cardamine hirsuta* and *Arabidopsis thaliana;* with historical reference on herbarium voucher specimens in Kenya

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Abstract

Monitoring of plant species presence in past recorded sites helps to establish whether there is a shift of the species with respect to elevation gradient; and also assess other potential natural and anthropogenic factors that may be attributed to any possibility of shift or change in the species spatial, temporal and geographic distribution. This study attempted to trace the spatial and geographic distribution of the two related members of the family Cruciferae: Cardamine and Arabidopsis. The variations observed in leaf morphology of the Cruciferae representatives in this study have been associated with local growth and patterning, whereas their siliquae show striking similarity in structure and dispersal mechanisms. Floristic associations of the genus Cardamine and Arabidopsis comprise Salvia nilotica, Geranium arabicum, Artemisia afra, Veronica javanica, Trifolium sp., Hebenstretia angolensis, Galium sp., Oxalis corniculata and Kniphofia thomsonii. From the survey of the favourable growth sites for Cardamine and Arabidopsis, Mt. Elgon and Mt. Kenya (Chogoria Route) provided good conditions for germination, establishment and spatial distribution. Observed anthropogenic impacts to the montane Cruciferae habitats include large wild herbivore grazing, livestock grazing, road construction, and forest fires.

Introduction

The species *Cardamine hirsuta* is in the genus Cardamine and the family Cruciferae or Brassicaceae which is comprised of several other genera which include Subularia, Farsetia, Rorippa, Oreophyton, Raphanus, Diceratella, Arabidopsis, Arabis, Barbarea, Sisymbrium, Erucastrum, Brassica, Crambe,

Capsella, Thlaspi, Camelina, Lepidium and Coronopus (Agnew, **2013**). The main focus of this study was to explore the geographic distribution, morphological and anatomical variations of mature and reproductive wild *Cardamine hirsuta* sampled in its natural habitat (Figure 1) and compare the information collated with that of the herbarium voucher specimens collected over the years. Some publications have documented clearly the similarities and differences among the two closely related genera particularly the Bitter Cress (*Cardamine hirsuta*) and its wild relative, Thale (*Arabidopsis thaliana*).

Figure 1: Mature *Cardamine hirsuta* showing prominent white flowers and greenish apical siliques growing on a recovering burnt grassland from wild fire in the Mt. Elgon National Park and Reserve in Western Kenya (Photograph by Chebii Kibet).

The differences of gene function and regulatory mechanisms in Cruciferae have been widely researched on the leaf development (Hay & Tsiantis, **2006**). Many differences in leaf form have been established both between and within species, *A. thaliana* have simple leaves (**Figure 2**) as compared to the compound and somewhat complex leaves of *C. hirsuta*. These differences in leaf morphology and development have been attributed to the local growth and patterning whereas *A. thaliana* is normally considered as a reference or model plant species (Hay *et al*., **2014**; Cartolano *et al*., **2015**) whereas the genus Lepidium is considered a reference model for fruit development among the Cruciferae. These model species are critical in developmental genetics and evolutionary relationships (Mummenhoff *et al*., **2008**). Although *Cardamine hirsuta* siliques are somewhat similar to those of *Arabidopsis thaliana* they can disperse seeds to about five (5) metres whereas *Arabidopsis thaliana* disperse seeds with the aid of gravity (Vaughn K.C. *et al*. (**2011**). Many studies on genetic aspects particularly on leaf development and reproductive structures have been carried out on *Cardamine hirsuta* and *Arabidopsis thaliana* plants.

Figure 2: *Arabidopsis thaliana* growing on a roadside and partly rocky substratum and soil-leaf-litter vegetation in the Mt. Kenya (Chogoria route) expressing conspicuous white flowers and showing a rosette leaf pattern.wers (Photograph by Chebii Kibet).

Cardamine hirsuta taxonomic description

C. hirsuta L. is described as a hairy annual herb with pinnate phyllotaxy usually in a basal rosette containing 3-7 pairs of somewhat circular leaflets. They have a white corolla and their staminate count is usually four (4), stamen count is often used to distinguish *Cardamine hirsuta* from its wild relatives. The spreading fruit, a 'pod' or also referred to as siliqua measures up to 30 mm long; their habitat mostly is comprised of an open, disturbed areas within an altitudinal range of 500-4600 m. Species samples have been previously recorded in the alpine and subalpine zones of Mt. Kenya, Aberdares N. Park and Mt. Elgon. On the other hand, *Arabidopsis thaliana (*L.*)* Heynh is described as a tiny (less than 10 cm high) hairy annual rosette-leaved herb. The habitat for *A. thaliana* include disturbed areas in the alpine grassland and can also be found in frost heaved ground with an elevation range of 1750-4250 m. Unlike *C. hirsuta*, *A. thaliana* can be found in diverse floral zones of Mt. Kenya, Mt. Elgon, Rift Valley, Mau Complex and the Aberdares N. Park among others ecogeographic locations (Agnew, **2013**).

Methodology

Herbarium voucher references

Preliminary stages of the study involved study of herbarium material for historical collections, sites and analysis of information recorded on the herbarium data labels to aid in the tracking of the species distribution and obtain raw data expressed by initial collectors as expressed through their field notes.

Vegetation sampling

Vegetation sampling was conducted in the months of January, February, March, April, June, September and December in the year 2017 following an altitudinal gradient (Odland & Birks, **1999**) where site coordinates and altitude was determined using a GARMIN GPS. A number of belt transects measuring 5m × 20m was laid, with subplots measuring 5m × 10m, (5 × 5) m^2 and (1 × 1) m^2. Important morphological parameters collected in the field included: herb height, silique length, leaf length and diameter alongside growth patterns. The sampling procedure shall follow the protocols laid by Mueller-Dombois & Ellenberg (**1974**) with regard to the herbaceous layer sample collection. Most of the plant species were identified in the field by taxonomic experts, and voucher

specimens were collected for herbarium verification and determination using relevant taxonomic literature. A species checklist and a comprehensive species list in each transect was also compiled.

Sample collection points for mapping *Cardamine* and *Arabidopsis* distribution

We used GPS records derived from herbarium field notes, although most herbarium vouchers of old species folders lacked GPS coordinates. Sampling was done with transects radiating from positively determined GPS locations, where semi-random transects were virtually created and sampling done along the wet areas, particularly in the banks of rivers and streams and along human track paths to the montane peak. Where species of interest was encountered the new location (GPS coordinates and elevation) was marked using a Garmin GPS (*etrex* legend HCx). These new GPS locations were used in generating and updating the distribution maps prepared using Global mapper version 15 generated from the few GPS coordinates captured in the past collections was (Englund *et al*., **2001**).

Results

Sampling points for fresh Cruciferae sample collection for the year 2017

From the fresh sample collection done periodically, a record of the specific locality, elevation points, Cruciferae collected and their described species-habitat description were tabulated as shown on Table 1 below.

Table 1: Tabulation points of fresh sample collection for *Cardamine hirsuta* and *Arabidopsis thaliana*, with a historic representation of their specific GPS localities, elevation their habitat description

Locality	Route/Specific site	Species-Habitat description	Cruciferae collected
Mt. Kenya National Park	Sirimon Track	*Hagenia-Juniperus* Zone: Roadside with *Cineraria deltoides, Clutia abyssinica, Berberis, Leonotic cymifolia, Bothriocline, Salvia, Alchemilla, Galium, Agrostis*	*Cardamine obliqua*
		Moorland/Heath zone: Comprising *Stoebe, Hebenstretia, Carex monostachys, Anthemis, Alchemilla argyrophylla, Artemisia and Hypericum revolutum*	
	Old Moses Camp	Moorland: On wet ground with *Euryops brownei, Stoebe, Trifolium, Rumex steudelii, Poa annua, Agrostis* and *Crassula schimperi*	
Aberdare National Park	Chania Falls	River Gorge with *Rubus, Crepis, Kyllinga odonata, Plantago palmata, Poa annua, Hypericum, Senecio, Asplenium aethiopicum* and *Geranium*	*Arabis alpina*
	Queen's Cave Waterfalls	River Gorge on rocky damp ground with *Asplenium aethiopicum, Schefflera volkensii, Solanum terminale, Conyza sp., Plantago*	*Cardamine obliqua*

		palmata and *Dichrocephala chrysanthemifolia*	

Locality	**Specific Locality**	**Habitat**	**Plant community**	
Mt. Kenya	Mt. Kenya, Teleki Valley	Moorland, growing on shady places on rocks	*Lobelia, Alchemilla argyrophylla, Helichrysum forskahlii,* Grass tussock *(Eleusine sp.),* and *Sedum sp.*	*Arabis alpina* (Species on high altitude drew huge similarity with *Arabidopsis thaliana*
			Lobelia, Alchemilla argyrophylla, Helichrysum forskahlii, Grass tussock *(Eleusine sp.), Sedum sp., Ranunculus oreophytus, and Dendrosenecio sp.,*	
	Sirimon track intake	Stream edge	*Sambucus africana, Podocarpus latifolius, Olea europaea ssp. africana, Arundinaria alpina, Alchemilla argyrophylla, Agrocharis incognita* and *Impatiens tinctoria*	*Cardamine trichocarpa* (Emergent seedlings drew huge similarity with C. hirsuta)
	Chogoria route		*Podocarpus latifolius, Olea europaea ssp africana, Pteris sp., Arundinaria alpina, Rumex sp., Cyperus ajax,* and *Impatiens tinctoria*	
	Old Moses Camp	Marshy vegetation	*Cyperus sp., Poa annua and Epilobium sp.*	*C. trichocarpa & C. obliqua*
Aberdares National Park	Along Mutubio Gate Steep hill	Stream edge	*Nuxia congesta, Acacia melanoxylon, Dombeya torrida, Rubus pinnatus, Asplenium sp., Trifolium semipilosum, Droguetia iners, and Clutia abyssinica*	*C. trichocarpa*
	On wet roadside	Hagenia-Bamboo zone	*Trifolium semipilosum, Rumex steudelii, Sambucus africana, Bothriocline fusca, Plectranthus sp., Galium thunbergianum, Euphorbia sp., Geranium sp., Clinopodium uhligii, and Polystichum sp.*	*C. obliqua*
Mt. Elgon	Track to the peak at End Road	.	*Trifolium spp, Dichondra repens, Galium spp*	*Cardamine hirsuta*

Saiwa Swamp National Park	Wetland tracks	Wetland streams	*Cyperus spp., Ensete ventricosum, Bersama abyssinica, Stephania abyssinica, Galinsoga parviflora, Laxogramme lanceolate, Oxalis spp., Centella asiatica, Panicum sp., Veronica anagallis-aquatica*	*Cardamine hybrid (C. flexuosa), C. hirsuta*

Floristic association of the Cardamine hirsuta plant community

Cardamine hirsuta is known to grow on wet soils, wet road sides/pathsides and on clean flowing streams. From three sampled locations of the Mt. Elgon habitat, the species that were recorded in the field to provide a conducive habita for *C. hirsuta* include: *salvia nilotica, Geranium arabicum, Artemisia afra, Veronica javanica, Trifolium sp., Pseudognaphalium luteoalbum, Heliotropium sp., Hebenstretia angolensis, Cyperus rigidifolius, Haplocarpha sp., Micromeria imbricata, Anthemis tigrensis, Oxalis corniculata, Galium sp., Conyza sp., Helichrysum sp., Kniphofia thomsonii* and quite notable species was *Pycreus nigricans*.

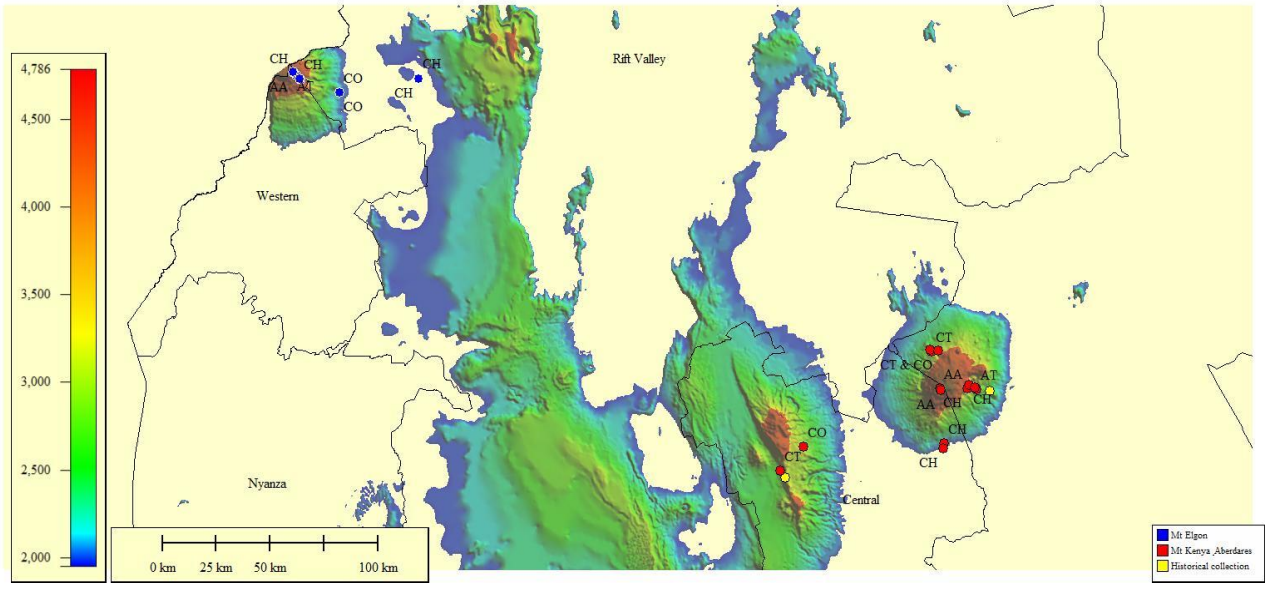

Figure 3: *A map showing the spread and distribution of members of Cruciferae showing the individual species collected: CH= Cardamine hirsuta, CT= Cardamine trichocarpa, CO= Cardamine obliqua, and AA= Arabidopsis thaliana. The colour coding:* ***Red*** *spots represents a present and spatial distribution of Cruciferae in the Mount Kenya regions of Kenya, whereas the* ***blue*** *spots represents a spatial distribution of Cruciferae in the Mt. Elgon region, and the* ***yellow*** *spots represent past collection spots as recorded in the herbaria collections of the National Museums of Kenya and the University of Nairobi herbarium.*

DISCUSSION

From the herbarium voucher specimens, no recent collections have been made and this could be attributed to few collection expeditions targeting the study of Cruciferae in Kenya coupled with their complex montane habitats. Previous recordings largely lacked GPS coordinates and one can only trace species localities using well written field notes and description of the habitats as shown on herbaria specimens. There is need for herbaria to arrange for fresh collections using past herbaria

voucher records and help in the assessment of potential shifts in Cruciferae habitats and evaluate the associated natural and anthropogenic causes of habitat changes. GPS recordings and analysis of plant communities for these related Cruciferae genera are crucial in order to improve understanding of the associated floristic composition in the wild and the community contribution in their growth and establishment. This would also help in the evaluation of upward shift of species due to meteorological elements variation following an elevation gradient. Unlike the Cherangani Hills, Mt. Elgon still has considerable patches of bamboo zone as shown on Figure 4 below.

Figure 4: The growing bamboo of the Mt. Elgon National park in the *Podocarpus* dominated zone (Photograph by Chebii Kibet)

CONCLUSION

The habitats of *Cardamine hirsuta* has been declining and there is increased frequency in the identification of the hybrid *Cardamine flexuosa.* Most of the past collection sites indicated lack of living vouchers and this could be attributed to livestock grazing inside the protected national parks and reserves, particularly in the grasslands of Mt. Kenya and Mt. Elgon. Other anthropogenic disturbances that may affect the spatial and temporal distribution of the montane *Cardamine* include grassland fires, road construction inside the parks, large herbivore grazing patterns and behaviour (elephant, buffalo, antelopes), and the ever changing weather patterns. Road construction negatively impacts the roadside vegetation by introducing alien species to the forest ecosystem, particularly colonizing the roadside plant communities.

Figure 5: Controlled grazing at the grassland zone, a compromise conservation approach that involve the community in forest conservation (Photograph by Chebii Kibet)

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