

# Conservation and ecological interactions of *Astragalus bozakmanii* Podlech (Fabaceae)

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**ABSTRACT** - (Conservation and ecological interactions of *Astragalus bozakmanii* Podlech (Fabaceae)). *Astragalus bozakmanii* Podlech, an endemic plant species to Turkey, was investigated to provide an understanding the conservation status of this species and to fill the gap in the knowledge of habitat of this species. Field studies were carried out randomly by sampling  $2 \times 2$  m sizes, 10 quadrats from each region. The number of individuals was estimated by calculating the average individual numbers in small quadrats and extrapolating this result for approximate population area. Reproductive success was evaluated using the mean number of seeds per fruit and the number of ovules per flower. *A. bozakmanii* prefers saltless soils whose pH values range from 7.48 and 8.33. The seed set was 45.8% of the ovules. In this study, we observed that the flowers of *A. bozakmanii* were visited by *Oxythyrea cinctella* to feed on their reproductive parts, thus rendering them infertile. Considering all these findings, the global and national conservation status for *Astragalus bozakmanii* was assessed as Endangered (EN) B1ab (i, ii, iii) + 2ab (i, ii, iii) according to the new IUCN Red List criteria. In conclusion, appropriate conservation actions need to be developed so that natural enemies of agricultural pests can re-enter their habitats. **Keywords:** conservation, IUCN, Turkey, endemic, Anatolian steppe

**RESUMO** - (Conservação e interações ecológicas de *Astragalus bozakmanii* Podlech (Fabaceae)). *Astragalus bozakmanii* Podlech, uma espécie de planta endêmica da Turquia, foi investigada para fornecer uma compreensão do estado de conservação dessa espécie e para preencher a lacuna no conhecimento do seu habitat. Os estudos de campo foram realizados de forma aleatória por amostragem de tamanhos de  $2 \times 2$  m, 10 quadrados de cada região. O número de indivíduos foi estimado calculando os números médios individuais em pequenos quadrantes e extrapolando esse resultado para a área aproximada da população. O sucesso reprodutivo foi avaliado usando o número médio de sementes por fruto e o número de óvulos por flor. *Astragalus bozakmanii* prefere solos sem sal, cujos valores de pH variam de 7,48 a 8,33. A formação de sementes foi de 45,8% dos óvulos. Neste estudo, observamos que as flores de *A. bozakmanii* foram visitadas por *Oxythyrea cinctella* para se alimentar de suas partes reprodutivas, tornando-as inférteis. Considerando todas essas descobertas, o status de conservação global e nacional de *Astragalus bozakmanii* foi avaliado como Ameaçado (EN) B1ab (i, ii, iii) + 2ab (i, ii, iii) de acordo com os novos critérios da Lista Vermelha da IUCN. Em conclusão, ações de conservação adequadas precisam ser desenvolvidas para que os inimigos naturais das pragas agrícolas possam reentrar em seus habitats. **Palavras-chave:** endêmica, estepe da Anatólia, conservação, IUCN, Turquia

## Introduction

*Astragalus* L. (Fabaceae), one of the largest genera of flowering plants on Earth, comprises almost 3,000 taxa and more than 250 sections (Podlech, 1986, Maassoumi, 1998, Podlech and Zarre, 2013, Atasagun et al., 2018). Nearly 476 taxa 64 sections have been recorded with 203 endemic taxa and 51% endemism rate in Turkey known as the important endemism and gene diversity centre of the genus. (Chamberlain and Mathews 1970, Aytaç et al. 2012, Karaman Erkul et al. 2014, İlçim & Behçet 2016). *Astragalus bozakmanii* Podlech was first collected in 1970 by Bozakman and K. Fitz on the Kızılcahaman-Çeltikçi road in Ankara province. It was known as an endemic species from Turkey, was published by Podlech (2001) as a new species in the revision studies. *A. bozakmanii* is a perennial species that grows up to approximately 15 cm. Leaves are 15-20 cm long and petioles are 4-6 cm long. Leaflets are

generally in 15 pairs (in first leaves usually in 10 pairs), elliptic and 10-13(-18) x 5-6(-8) mm. Racemes are loosely 15-20-flowered. Calyx is 12-13 mm long, tubular. Petals are glabrous, yellow, the standard slightly reddish-suffused (Podlech 2001). Because of its limited distribution range and few known populations, it was considered as critically endangered; however, there is no information about exact number of individuals (Eker et al. 2015). Further, there are a lot of incomplete information about habitat preference, environmental requirements, ecological thresholds, limiting its distribution, population size, and potential threats on *A. bozakmanii* populations. The objective of our research is to fill this gap. The main goals of our research are (1) to provide data about the habitat and population size of *A. bozakmanii*; (2) to determine the environmental drivers of the species distribution range; (3) to identify its major threats; and (4) to re-evaluate its conservation status at global scale.

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## Materials and methods

**Data collection** - The herbarium specimens of *Astragalus bozakmanii* were provided a single data source of its locality. [(Holotypus: Turkey, Prov. Ankara, Zwischen Kizilcahamam und Çeltikçi road, 6.6.1970, I. Bozakman and K. Fitz 906 (W).)] Field studies was carried out to verify the location of species stated in the literature and to discover new locations for determination of the distribution area between June and August 2018. Coordinates, population size (number of the individuals), elevation, habitat type (according to IUCN habitats classification scheme ver 3.1; IUCN 2012a), and the threats (according to IUCN threats classification scheme ver 3.2; IUCN 2012b) were recorded for all sites. The coordinates of the locality were taken by using a GPS (Global Positioning System) device in the field. Habitat types and major threats of each site were obtained from fieldworks. They were carried out randomly by sampling 2 x 2 m sizes and 10 quadrats from each region. The number of individuals was estimated by calculating the average individual numbers in small quadrats and then extrapolating this result for approximate population area. The closest climatic description of the area was provided by the Çamlıdere OGM meteorological station (Çamlıdere Forest Operation Directorate, Station No: 18075, <https://www.mgm.gov.tr>), which is a slightly higher altitude (1234 m), but also about ca 10 km to the northeast (40° 29.466' E, 32° 28.536' N).

**Data analysis** - The GeoCAT programme (Bachman & al., 2011; <http://geocat.kew.org/>) was utilized for calculating the extent of occurrence (EOO) and the area of occupancy (AOO) and the cell width was chosen as 2x2 km based on our observations. Area of Occupancy (AOO, km<sup>2</sup>), Extent of Occurrence (EOO, km<sup>2</sup>), population numbers and field observations were used to re-evaluate the conservation status of this species according to the IUCN (2012c) Categories and Criteria (ver 3.1). Soil samples were obtained from two randomly chosen places in the distribution area of the species. Samples were taken from 0-20 cm soil depth and the laboratory analyses were performed based on Walkley and Black (1934), Jackson (1958), Richards (1954), and Allison and Moode (1965).

The average seeds per fruit and ovules per flower were counted by evaluating the seed set. To determine fruit set, 100 flowers (randomly selected 20 individuals from each site) were randomly taken from *A. bozakmanii* population and labelled. All mature fruits were collected 45 days after anthesis. Following this, the mean seeds per fruit (n = total 100 fruits, randomly selected from all sites) was calculated. Seed set (%) was measured by the ratio of the number of seeds per fruit to the number of ovules per flower.

Pest determination observations were performed in 10 randomly assigned individuals for three days between the hours of 09:00-17:00, when the weather conditions were appropriate. Pests were collected from *A. bozakmanii* by hand and visual observation methods were used in the collection of samples. They were moved in a small dark

glass bottle with 70% alcohol. In the laboratory, every sample were identified using a stereomicroscope.

## Results and Discussion

**Distribution, habitat, and environmental drivers** - *Astragalus bozakmanii* was first collected in 1970 by I. Bozakman and K. Fitz on the Kızılcahamam-Çeltikçi road. It has been identified only from its holotype locality since 1970 (Podlech, 2001) which was also confirmed by us. In this study, the species was established in four new sites (at Yahşihan, Yoncatepe, Bayındır, Elmalı villages) around Çamlıdere dam in Ankara in addition to the known locality (figure 2, table 1). The area of the new localities ranges between 0.33 and 3.331 km<sup>2</sup> and the elevation varies between 965 and 1185 m. *A. bozakmanii* grows on the calcareous main mass (figure 1) and is mostly distributed in the steppe, degraded oak areas and abandoned fields.

The soils that the species grows on are characterized by alkaline soils, rich in organic matter, nitrogen, phosphorus, and potassium. Due to the fact that the Çamlıdere dam basin an important water resource in the region, is under protection, the species such as *Quercus pubescens* Willd., *Crataegus monogyna* Jacq., *Prunus spinosa* L. *Scutellaria orientalis* L., *Astragalus vulnerariae* DC., *Ajuga salicifolia* (L.) Schreb., *Acanthus hirsutus* Boiss., *Sedum acre* L. subsp. *acre*, *Centaurea urvillei* DC., *Aethionema armenum* Boiss., and *Hedysarum varium* Willd., which represent the natural vegetation of this region, had the opportunity to redevelop in the abandoned agricultural lands around the dam.

According to climatic data, the average annual temperature is 9.45 °C. *Monthly average maximum temperature reaches 20.5 °C in July. The average minimum temperature reaches -1.9 °C in January. Extreme temperatures records are -14.26 °C in January and 33.32 °C in July.* The highest and lowest precipitation is 65.2 and 6.9 mm in June and July, respectively. The highest and lowest amount of humidity is 86.2 and 52.2 mm in January and September, respectively. The first flower buds of *Astragalus bozakmanii* were observed after the second week of May and the flowering period began in May and continued until mid-July. Fruits that began to mature by the end of June could be observed until the end of August (table 2).

*Astragalus bozakmanii* prefers saltless soils with the pH between 7.85 and 8.06 (slightly alkaline) and clayey with 1.57-23.65% lime. The soils in which *A. bozakmanii* grows include poor organic matter content (1.03-1.39 %), nitrogen (0.05-0.07 %), and phosphorus (1.71- 2.62 %). In additionally, the soil was also rich in CaCO<sub>3</sub> and potassium (table 3).

**Seed set** - In *A. bozakmanii*, seed production in individuals exposed to pollinator visits under natural conditions, the average number of seeds per fruit was calculated as 4.58 ± 2.21, and the number of ovules per flower was 10. The seed set was 45.8 % of the ovules. The mean number of seeds per legume was reported as 11.8 in populations of *Astragalus gines-lopezii* Talavera



Figure 1. Characters of *Astragalus bozakmanii* Podlech. a. General appearances of flower. b. Legume. c. Habit. d. Habitat.

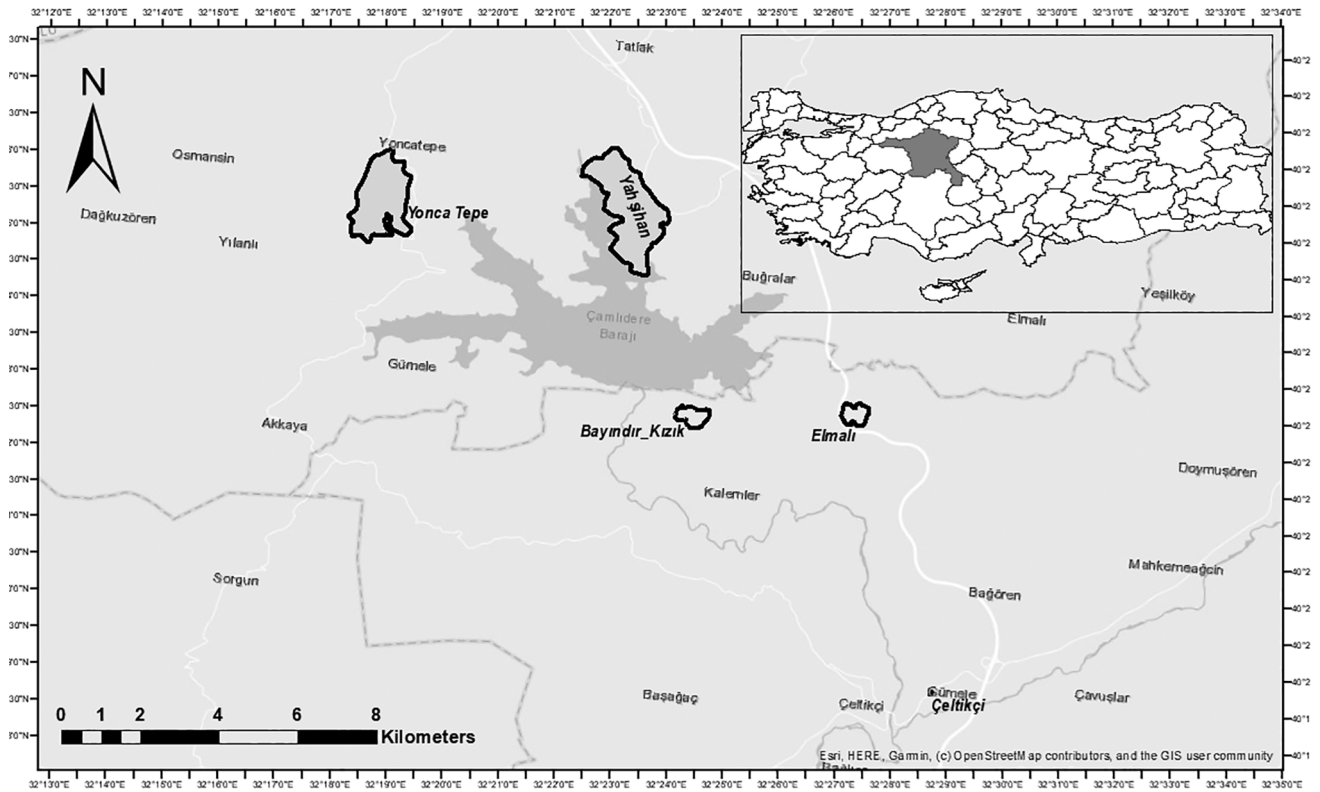


Figure 2. Distribution of *Astragalus bozakmanii* Podlech (Çamlidere, Ankara, Turkey).



Table 1. The four sites where *Astragalus bozakmanii* Podlech located, with altitude range, identified threats (IUCN 2012a), and habitats classification scheme coded (IUCN 2012b).

Sites Number	Name Sites	Estimated Population Size	Area of sites (km <sup>2</sup> )	Elevation (m above sea level)	Habitat (IUCN habitats classification scheme)	Assessed threats (IUCN threat classification scheme)
1	Yahşıhan	32.835	3.31	965-1030	14.1 Arable land 14.2 Pastureland	2.1 Annual & Perennial Non-Timber Crops 2.1.1 Shifting agriculture 2.3 Livestock Farming & Ranching; 2.3.2 Small-holder Grazing, Ranching or Farming 4.1 Roads & Railroads 7.2 Dams & water management/use; 7.2.9 Small dams 8.2 Problematic native species/diseases; 8.2.1 Unspecified species
2	Yoncatepe	9.600	2.40	1040-1100	14.1 Arable land 14.2 Pastureland	2.1 Annual & Perennial Non-Timber Crops 2.1.1 Shifting agriculture 2.3 Livestock Farming & Ranching; 2.3.2 Small-holder Grazing, Ranching or Farming 4.1 Roads & Railroads 7.2 Dams & water management/use; 7.2.9 Small dams 8.2 Problematic native species/diseases; 8.2.1 Unspecified species
3	Bayındır	6.832	0.70	1020-1100	14.1 Arable land 14.2 Pastureland	2.1 Annual & Perennial Non-Timber Crops 2.1.1 Shifting agriculture 2.3 Livestock Farming & Ranching; 2.3.2 Small-holder Grazing, Ranching or Farming 4.1 Roads & Railroads 7.2 Dams & water management/use; 7.2.9 Small dams 8.2 Problematic native species/diseases; 8.2.1 Unspecified species
4	Elmalı	1.690	0.33	1165-1185	14.1 Arable land 14.2 Pastureland	2.1 Annual & Perennial Non-Timber Crops 2.1.1 Shifting agriculture 2.3 Livestock Farming & Ranching; 2.3.2 Small-holder Grazing, Ranching or Farming 4.1 Roads & Railroads 7.2 Dams & water management/use; 7.2.9 Small dams 8.2 Problematic native species/diseases; 8.2.1 Unspecified species
Total		50.957	6.74			

Table 2. Phenology stages of *Astragalus bozakmanii* Podlech.

Stages	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr
Vegetative												
Flower Budding												
Generative												
Flowering												
Seeding												
Temperature average (°C)	12.25	16.3	20	20.54	16.66	10.04	5.3	-0.66	-1.96	1.86	4.48	8.56
Precipitation average (mm)	55.35	65.27	6.975	26.2	19.3	25.3	17.88	41.62	46.4	21.62	42.9	43.78
Humidity average (%)	64.14	67.06	54.26	55.18	52.25	65.44	66.78	76.27	86.26	59.58	66.35	59.38

Table 3. Soil characteristics in natural habitats of *Astragalus bozakmanii* Podlech.

Sites (Value = Mean ± SD)	Elmalı		Yahşıhan		Yoncatepe		Bayındır	
pH	8.06±0.19	7.92- 8.2	7.85±0.20	7.71-7.99	7.92±0.10	7.85-7.99	7.91±0.60	7.48-8.33
% CaCO <sub>3</sub>	15.18±11.9	6.7-23.65	5.98±4.84	2.56-9.4	1.78±0.30	1.57-1.99	13.82±6.04	9.55-18.09
% Salt	0.03± 0.00	0.0273-0.0276	0.04±0.00	0.0395-0.0405	0.03±0.01	0.0288-0.0381	0.04±0.02	0.0314-0.0564
% Organic matter	1.10± 0.38	0.83-1.37	1.39±0.66	1.86-0.93	1.03±0.55	0.63-1.42	1.17±1.10	0.39-1.95
% N	0.05±0.02	0.04-0.07	0.07±0.03	0.05-0.09	0.05±0.03	0.03-0.07	0.06±0.06	0.1-0.02
% Saturation	68.64±12.45	59.84-77.44	90.04±1.17	89.21-90.86	72.33±2.10	70.84-73.81	91.03±25.12	73.26-108.79
P (kg/da)	1.87± 0.73	1.35-2.39	2.62±0.67	3.1-2.15	1.71±0.39	1.43-1.99	1.71±0.06	1.67-1.75
K (kg/da)	67.97±39.28	40.2-95.75	187.77±31.90	165.21-210.32	174.79±14.11	164.81-184.77	77.88±10.95	70.14-85.62

& al. (Martínez-Fernández et al., 2014). Results from this study have not so high if it is compared with data of *A. bozakmanii*.

Conservation status - The current total estimated population size was calculated here to be 50.957 individuals: 32.835 in Yahşıhan, 9.600 in Yoncatepe, 6.832 in the Bayındır, and 1.690 in Elmalı, and estimated population size of each site ranged between 1690 and 32985 individuals. Based on the extent of occurrence (38.105 km<sup>2</sup>), area of occupancy (20 km<sup>2</sup>), number of locations (IUCN, 2019), and decline of the original population due to habitat loss, we assessed the species as Endangered (EN) status and B1ab (i, ii, iii) + 2ab (i, ii, iii) in the IUCN Red List of threatened categories and criteria.

*Piezodorus lituratus* (F.), an insect pest classified under Pentatomidae (Heteroptera), was observed in most of the mature legume pericarps of *A. bozakmanii*. Additionally, the beetle species *Oxythyrea cinctella* (S.), usually feeds on flowers of the host plant, is distributed in South Caucasus, Iran, Afghanistan, Pakistan, Middle Asia and Caucasus (Hurpin, 1962; Smetana, 2006). It was observed that this insect species caused significant damage to mature

fruit and seeds of *A. bozakmanii* (figure 3). *Piezodorus lituratus* (F.), also known as Gorse Shield Bug (Heteroptera: Pentatomidae), is a widespread insect in many parts of Europe, and feeds on several plants, especially flowers and fruits of many Fabaceae family members (Schaefer and Panizzi 2000) (figure 3). Some researchers noted in their studies that adult beetles cause local damage to bud of ornamental plants such as *Rosa* L., *Prunus* L., *Cydonia* L., *Papaver* L., *Vitis* L., different cereals as well as other deciduous fruit trees (apricot, citrus, almond, apple, and etc.; Öztürk et al., 2004, Hurpin, 1962). Similar to other members of Cetoniinae, the scarab beetles can also damage the reproductive parts of flowers, and thus make them infertile (Hurpin, 1962; Homonnay and Homonnayne-Csehi, 1990; Alford, 1991). In accordance with these studies, we observed that the flowers of *A. bozakmanii* were visited by *Oxythyrea cinctella* (S.) to feed on their reproductive parts, thus rendering them infertile (figure 3).

The main threats of the entire population of *A. bozakmanii* were habitat fragmentation and habitat loss due to degraded/abandoned areas after agricultural activities and roads opened for access to agricultural lands and



Figure 3. Pests observed on reproductive organs of *Astragalus bozakmanii* Podlech. a. *Piezodorus lituratus* (Fabricius, 1794). b. *Oxythyrea cinctella* Poda.

infrastructure expansion. The individuals of *A. bozakmanii* were frequently encountered in the roadside and/or field edge habitats formed as a result of the anthropogenic influences. Although there were some differences between the observed threats on the localities, the most prominent threat was the altered habitat composition with agricultural activities. Agricultural expansion induces to significant habitat loss and fragmentation of natural and semi-natural habitats, which ultimately causes the loss of natural enemies of agricultural pests (Zhao et al., 2015; Schellhorn et al., 2015) and declines in biodiversity (Bianchi et al., 2006; Tscharntke et al., 2007). *Decreasing* plant diversity leads to changes in the community composition of the pest complex-herbivorous insects and their natural enemies (predators and parasites) (Power and Flecker, 1996). Increasing agricultural pests leads to decrease in plant population size. Consequently, serious problems were observed in the reproductive biology of *A. bozakmanii* due to the decrease in the natural enemies of agricultural pests; however, further research to reveal interaction between this plant species and its pests are needed.

Conclusions

Area of occupancy and extent of occurrence for the threatened plant taxa were calculated as 38.105 km<sup>2</sup> and 20 km<sup>2</sup>, respectively. According to IUCN (2019), the current status of the species can be evaluated as EN (Endangered) based on the B criterium in terms of EOO value <5000 km<sup>2</sup> (38105 km<sup>2</sup>) B1b (i, ii), AOO (area of occupancy) value <10 km<sup>2</sup> (20 km<sup>2</sup>) B2b (i, ii). However, the section

4.11 in the IUCN Guidelines (IUCN, 2019) describe the term ‘location’ as a geographically or ecologically distinct area in which a single threatening event can rapidly affect all individuals of the taxon present. All subpopulations of the taxon were admitted as a single location owing to the existence of a sole main threat in the habitats (Condition (a) under Criterion B1 and B2). In addition, the continuous decrease of the area, extent, and quality of the habitat fulfils the requirement (iii) under Criterion B1b and B2b. As a result of all these assessments, we assessed the Endangered (EN) global conservation status for *Astragalus bozakmanii*, according to the new IUCN Red List criteria (IUCN, 2019), B1ab (i, ii, iii) + 2ab (i, ii, iii).

The IUCN Red List recommends important conservation precautions and studies for *Astragalus bozakmanii* (table 4). Developing a long-term conservation action of *A. bozakmanii* is primarily necessary to study its population genetics. As this type of research requires long term studies. Thus, we suggest an integrated conservation plan for *A. bozakmanii* including both in-situ and ex-situ conservation to prevent the depletion of the present gene pool. In addition, the change of microclimate due to Çamlıdere dam on a local scale should be considered and appropriate protection actions need to be developed so that natural enemies of agricultural pests can re-enter their habitats. We propose that the cooperation between research institutes and local government should be to carried out in the same area every year for the restoration of natural processes of *Astragalus bozakmanii* habitat and for the long-range observation of the population and habitat trend.

Table 4. Conservation actions needed and research needed proposed and carried out, for *Astragalus bozakmanii* Podlech. Conservation actions coded following IUCN (2012c) and research needed are IUCN (2012d).

Conservation action	Proposed	Research needed	Adopted
3. Species management	3.4. Ex situ conservation	1. Research	1.6. Conservation action
2. Land /Water Management	2.3. Habitat Natural Processes restoration	3 Monitoring	3.1. Population Trend 3.4. Habitat Trend

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## Notes on contributors

Ümit Subaşı: all stages of this article, fieldwork, control of herbarium collections (Herbarium of Ankara University - ANK) and literature, evaluation of the conservation status, article writing phase.

## Conflicts of interest

The author not reported any potential conflict of interest.

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