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On the perspectives of the study of the anxiolytic effect of plant species, endemic to Caucasus

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Abstract

Present article is aimed to **w**elcome colleagues**,** interested in phytomedicine**,** to the collaborati**ve** study of the anxiolytic effects of the extracts of some plant species**,** endemic to Caucasus**.** Rat **g**rooming, as **w**ell as rat behavior in Elevated plus maze**,** forced swimming test, Open **f**ield test, Light and dark box test and Punished conflict test, are suggested animal models of anxiety for the study of anxiolytic effects of endemic plant extracts.

KEYWORDS: plant; endemic; Caucasus; anxiolytic; collaboration; research

Introduction

Earlier reports from the World Health Organization [1] suggested that anxiety and related disorders will become the second leading cause of disability in both developed and developing countries by the year 2020. Present situation (War in Ukraine) represents an immediate cause of elevation in the level of anxiety with subsequent negative effect on human well being. Taking into account the wide- spread use of plant extracts and their effectiveness in treating anxiety, as well as polyvalent treating capacity [2], absence of side effects and relatively low costs as compared to the use of prescription drugs, present article is aimed at promoting the study of the role of officinal plant species in treating anxiety in human beings.In particular, we would like to invite colleagues, interested in phytomedicine, to the collaborative study of the anti-anxious effects of the extracts of some plant species, endemic to Caucasus and to Georgia in particular.

Variety of plant extracts are known for their anxiolytic effect and use in stress management. However plant species endemic to Caucasus, and to Georgia in particular, are not studied with this respect. Endemism is the ecological state that influences the chemical content of the plant species thus making endemic plantsthe focus of interest for their use in medicine [3,4]. Although there are ethnograph-ical documents concerning healing capacity of plants endemic to Georgia [5,6], anxiolytic effect of these plant species should be confirmed (or denied) in special scientific trials.

Extracts of the four endemic species are suggested to be examined for theiranxiolytic capacity in animal models using the methods of translational medicine. The choice of these species is based on preliminary information about their treating efficacy. In particular, earlier (2016-2018) we collected information from local dwell-ers in the territory of the spread of this endemic species. Respondents witnessedthe use of the tinctures of the plant species of interest as sedatives. At the sametime, respondents provided information about which anatomical parts are used for medicinal purposes, when (season) they should be collected (see Table 1) and what is a method of preparation of the herbal medicine rat grooming, as well asrat behavior in Elevated plus in Elevated plus maze (EPM), Forced swimming test (FST), Open Field test (OFT), Light and dark box test (LDB) and Punished conflict test, are suggested animal models of anxiety for the study of anxiolytic effects of endemic plant extracts. Grooming consists of two principal patterns: A. Syntactic chain: distinct, predictable sequence of bouts such as 1. licking the paws, 2. strokes along the snout,
3. strokes over the top of the head and behind the ears by the forepaws, 4. scratching

body by hind paws and licking the body fur and 5. licking hind paws, tail and genitals. B. second pattern is represented by spontaneous, not sequential bouts, which may be repeated many times [7,8].

Table 1. Endemic Plant species, suggested for translation medical trial

PLANT SPECIES	GENUS/PHYLUM	SPREAD IN NATURE
Pulsatilla georgica Rupr.,	Pulsatilla/ Ranunculaceae/ Ranunculales	Eastern Georgia
Galanthus woronowii	Galanthus/ Amarillidaceae/ Asparagales	Western Georgia
Campanulla kachetica Kantsch	Campanulla/ Campanullaceae/Asterales	Eastern Georgia
Gymnospermium smirnowii (Trautv)Takht.	Gymnospermium/Berberidaceae/ Berberid-ales	Eastern Georgia

Grooming behavioral microstructure undergoes predictable changes in stressful situations [9,8]. Increase in the percentage of interrupted grooming bouts, incorrect transitions from one grooming phase to another is described as a reaction to the stressor. These changes in grooming microstructure are behavioral marker of stress in rats [10]. It is known, that some plant tinctures have an effect on grooming disorders, caused by stress [11,12,13].

The EPM apparatus consists of a “+”-shaped maze, elevated above the floor with two oppositely positioned closed arms and a center area. When animal, placed in the center, explores the maze, its behavior is recorded and the preference for being in open arms over closed arms (expressed as either as a percentage of entries and/ or a percentage of time spent in the open arms) is calculated to measure anxiety-like behavior. The study of rats’ behavior in EPM relies upon rodents’ unconditioned fear and avoidance of heights and open spaces. Thus, behavior in EPM reflects a conflict between the preference for protected areas and instinctive drive to explore novel en- vironment. An increase of the time and the proportion of the entrances into the open arms without a changed locomotor activity are regarded as a powerful marker for an anxiolytic substance effect in rats and mice [14,15,16,17,18,19,20,21,22,23,24].

In FST animal is placed in an inescapable transparent tank that is filled with water and escape related mobility behavior is measured. The FST involves placing a rat or mouse in a tank with enough water so that it cannot touch the bottom with its hind paws. A normal animal displays a burst of escape activity, and then, eventually, immo- bility occurs when the animal stopes swimming and floats, producing only small limb

movements necessary to keep its head above water [25]. Movements to keep head above water are not suggested to express escaping activity. The period of immobility is recorded. Anxiety has an effect on the duration of escape behavior in FST and this test is used to study the anxiolytic effect of plant extracts [26,27].

The OFT is a common measure of exploratory behavior and general activity in rats, where both the quality and quantity of the activity can be measured. The OFT apparatus represents an enclosure, with walls, preventing experimental animal form escape. Distance moved, time spent moving, rearing, and change in activity over time are among many measures that can be tabulated and reported. Some outcomes, particularly defecation, center time, and activity within the first 5 minutes, likely gauge some aspects of emotionality including anxiety [28].

In PCT (Vogel Conflict Test) rats deprived from water, are provided with a drinking spout that delivers a mild shock after every 20 licks [29]. Some plant derived anxiolytics are reported to attenuate the shock-induced suppression of drinking [30].

LDB is based on the innate aversion of rodents to brightly illuminated areas and on the spontaneous exploratory behavior in response to novel environment and light. The test apparatus consists of a small dark safe compartment, where animal finds comfortable space, and an illuminated compartment, about two times larger of the dark compartment. Anxiolytics are reported to facilitate exploratory activity of animals – movement from the dark space to illuminated compartment to explore the new environment [31].

Collaborative efforts of specialists are desirable to fulfill the gap in the knowledge of anxiolytic effects of Caucasian endemic plant species. Observation of rat grooming in response to the stressor, as well as study of rat behavior in EPM, FST, OFT, LDB and PCT, with further biochemical and anatomical study of the nervous circuits – tar- gets for plant anxiolytics, is promising in throwing more light to the treating capacity of herbal extracts.

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