QUORUM SENSING SCREENING OF SOME MEDICINAL PLANTS FROM MONGOLIA

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ABSTRACT

The purpose of this study was to search for a novel quorum sensing inhibitor and analyse its inhibitory activity of medicinal plants of Mongolia. This study investigated the effects of some plant extracts on the bacterial communication system, expressed as quorum sensing activity. Quorum sensing has directly propotional effect on the amount of certain compound such as pigments, produced by the bacteria. Alcohol extracts of 103 extracts of 66 medicinal plants were tested for anti-quorum sensing activity by the Chromobacterium violaceum assay using the standard disc-diffusion method. The screening revealed the anti-quorum sensing activity of 18 extracts of 13 plants; particularly aerial part of Hedysarum alpinum L., Spongiocarpella grubovii Ulzii and Goniolimon speciosum (L.) Boiss.

KEYWORDS: Quorum sensing screening, *N*-hexanoyl-L-homoserine lactone, *Chromobacterium violaceum* CV026, medicinal plant

INTRODUCTION

The bacteria use signaling molecules which are released into the environment. The term of quorum sensing to describe the phenomenon in which bacteria converse via secreted signaling molecules called autoinducers, which regulated the expression of particular genes. In the natural environment, there are many different bacteria living together which use various classes of signaling molecules [1]. Quorum sensing allows bacteria to co-ordinate their behavior. As environmental conditions often change rapidly, bacteria need to respond quickly in order to survive. These responses include adaptation to availability of nutrients, defense against other microorganisms which may enter for the same nutrients and the avoidance of toxic compounds potentially dangerous for the bacteria. And quorum sensing inhibitors using various bacterial models, such as *C.violaceum* CV026, which is a violacein-gram negative double mini Tn5 mutant from *C.violaceum* ATCC 31532 [2]. Quorum sensing produce a natural antibiotic called violacein, which is a water-insoluble, purple pigment with antibacterial activity [3]. This research introduces quorum sensing screening of some medicinal plants from Mongolia.

MATERIAL AND METHODS

Bacterial strain and culture condition. *C.violaceum* strain CV026 was used to determine quorum sensing inhibitory activity, which controls production of violacein (a purple pigment) due to *N*-hexanoyl-L-homoserine lactone (HHL, SIGMA) called autoinducer [4].

The strain was grown in 5ml of Luria-Bertani broth (LB-1% tryptone, 0.5% yeast extract, 0.5% NaCl) solidified 0.5% agar when required and supplemented with kanamycin (WAKO, final concentration 50µg/mL) at 27°C for overnight.

Plant materials and extraction. About 100 of plant extracts in our natural products library were used for this assay. All air-dried plant materials were separately extracted 3-5 times at room temperature with 80% (w/v) acetone [5] and extracts were filtered using filter paper (Whatman). Then the solvents were evaporated under reduced pressure on a rotary evaporator and redissolved in appropriate concentration (30mg/mL) of 96% ethanol for further experiments [6].

Quorum sensing screening assay. Standard disc-diffusion method was employed to detect anti-quorum sensing activity of the plant extracts. Each extracts (50µL) were loaded into sterile discs [4] (8mm diameter) placed onto prepared LA plates (LA=LB+1.6% agar) spread with 3mL soft LA (soft LA=LB+0.5% agar) added 20µL HHL working solution and 100µL overnight culture of CV026. Plates were incubated at 27°C for 24 hours and quorum sensing inhibition was detected by ring formation on purple background suggested that plant extracts exhibited anti-quorum sensing [7].

RESULTS AND DISCUSSION

Medicinal use of extracts obtained from plants in general have recently gained popularity, inducing scientific interest exemplified in screening programs for novel and new components and uses pertaining microbial growth or bacterial quorum sensing inhibition.

To validate some aspects of the traditional uses of the tested plants and also their antipathogenic potential was checked by examininig the anti-quorum sensing activity of such extracts using *C.violaceum* assays.

Antipathogenic anti-quorum sensing activities were observed (Table 1) with extracts of *Spongiocarpella grubovii* Ulzii aerial part (36.5mm) *Goniolimon speciosum* (L.) Boiss. flower, leaves, stem (27.5, 23.5, 26mm, respectively) and *Hedysarum alpinum* L. aerial part (24.5mm) in Figure 1. Weak antiquorum sensing activities were observed with extracts of *Artemisia dracunculus* L. aerial part (9.7mm) and *Amethystea coerulea* L. root (9.8mm).

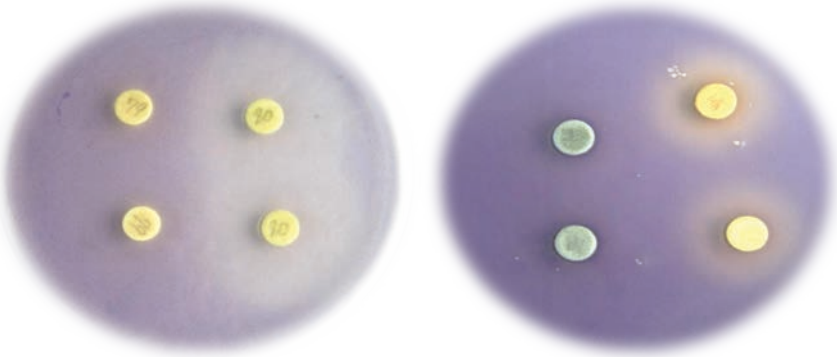
Table 1

The anti-quorum sensing activity of some plant extracts

№	Plant name	Part	Anti-quorum sensing activity (mm)		
			r1	r2	(r2-r1)
1	*Aconitum barbatum* Pers.	flower		8.7	8.7
2		leaves		8.1	8.1
3	*Ajania trifida* (Turcz.) Tzvel.	aerial part		8.3	8.3
4	*Amethystea coerulea* L.	root	9.8		
5	*Artemisia dracunculus* L.	aerial part	9.7	11.8	2.1
6	*Artemisia rutifolia* Stephex	aerial part		8.6	8.6
7	*Astragalus junafovii* Sancz.	aerial part	25	10	15
8		root	19	5.5	13.5
9	*Goniolimon speciosum* (L.) Boiss.	flower	27.5	10	17.5
10		leaves	23.5	9.5	14
11		stem	26	10	16

12	*Hedysarum alpinum L.*	aerial part	24.5	8.5	16
13	*Ledum palustre* L.	leaves	28	11	17
14		stem	19		
15	*Lophanthus chinensis* (Raf.) Benth.	aerial part	22	8.5	13.5
16	*Lophanthus krylovii* Lipsky.	leaves, flower	20.5	9	11.5
17	*Spiraea media* F. Schmidt	leaves, flower	18		
18	*Spongiocarpella grubovii* Ulzii	aerial part	36.5	19	17.5

paper disk D=8mm,
r1 = bacterial growth inhibition (antibacterial),
r2 = pigment inhibition, quorum sensing inhibition = (r2-r1)



a. b.

Figure 1. Anti-quorum sensing activity of some medicinal plants.
a. *Spongiocarpella grubovii* Ulzii. (aerial part), b. *Hedysarum alpinum* L. (aerial part)

CONCLUSIONS

In short, results presented here suggest that Mongolian medicinal plants have interesting anti-quorum sensing activities which may make them targets for the development of our further research.

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