2-2. 2 国間ワークショップ「日本・モンゴル、日本・インドネシア：生物遺伝資源とその取扱い」

(1) 日本・モンゴル 2 国間ワークショップ「モンゴルにおける生物遺伝資源とその取扱い」

2011 年 9 月 30 日、JBA は、モンゴルよりモンゴル国自然・環境・ツーリズム省/持続可能な開発及び戦略計画部の Banzragch Tsesed 局長と、植物の専門家であるモンゴル国立大学の Javzan Batkhuu 教授を招聘し、野村コンファレンスプラザ日本橋（東京）において 2 国間ワークショップを開催した。Banzragch 局長にはモンゴルの生物遺伝資源に関する現状と将来について（講演 1)、Batkhuu 教授にはモンゴルの薬用植物の利用について（講演 2）お話いただいた。

講演 1：「Convention on Biodiversity in Mongolia: Implementation of Nagoya Protocol and future approaches」（発表資料 1 参照）

モンゴル国内には多様なエコシステムがある。モンゴルは 1993 年に CBD を批准し、1996 年には国の生物多様性戦略、行動計画を決めた。絶滅危機にある動植物を自然環境で保護し、現在約 14%が保護地域（保護地域を 4 つのレベルに設定し、70 カ所を指定）となっている。2030 年にはこれを 30%にしたい。現地住民を保護活動に参加させることが重要で、130 万 ha を現地住民の組合に委譲し保護活動を委任している。モンゴルは社会主義から自由主義の経済成長の時代に移行し、地下資源の開発が盛んになってきた。これを自然環境の保護とどのように対処させるかが重要である。2009 年、河川の源流での地下資源開発を禁止した。これに対し開発会社が反対活動を続けているので、法の実現は非常に難しい。モンゴルの生物多様性は年々減少している。この要因は、野生生物を狩猟し海外へ輸出する、また、遊牧民による森林資源の燃料化によるものである。

遺伝資源は非常に豊かであるが、国の最大関心事は地下資源・家畜であり、遺伝資源の経済的な評価はされていない。種の多様性も豊かであるが、あまり科学的研究もされていない。限られた研究の中でも、ほとんどの動物は登録されている。海外との共同研究により、新種発見も増えている。

<u>モンゴルにおける遺伝資源に関する法律</u>

- ✓ 憲法（1992）：条項 6 では、資源は国家の財産であるとされた。
- ✓ 環境保護法：条項 15 では、保護機関は自然環境省とされた。
- ✓ 動物相に関する法、植物相に関する法、狩猟に関する法：資源の状況により用途を決め、制限する。

遺伝資源に関する法律（Future goal）

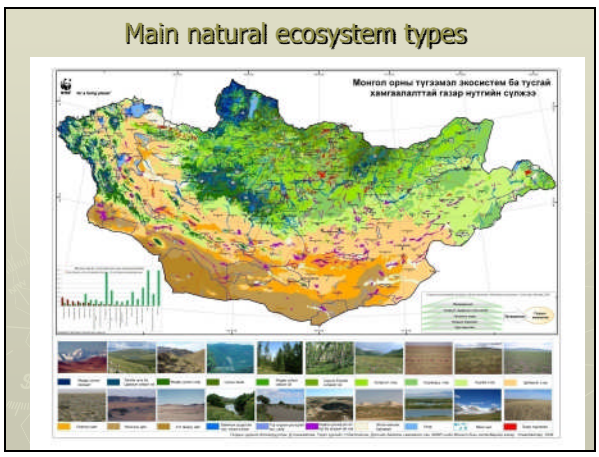
- ✓ 「名古屋議定書」は内閣の承認を得て、国会に提出中。本年中に署名したい。
- ✓ その後、モンゴルの遺伝資源法（案）を国会に提出する予定。
- ✓ 遺伝資源法（案）に含めること：中央政府の権限、地方政府の権限、利用許可システムの在り方、支払いシステムの在り方、伝統的知識の扱い方（モンゴル自然環境省が担当）

遺伝資源法（案）に関し、Banzragch 局長は、日本から意見提案をして欲しい旨述べた。また、彼は「利益配分など、モンゴルにとってどのようなシステムが良いのか一緒に考えて欲しい。教育、人材育成の分野でも協力してほしい。モンゴルの遺伝資源データ・ベース構築を大きなプロジェクトとしたい」と日本への期待を述べた。

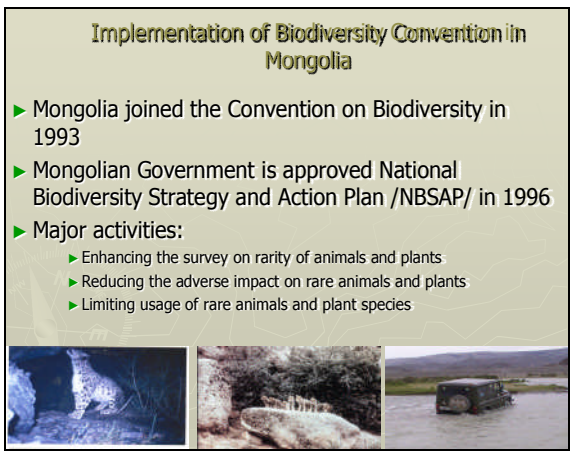
発表資料 1

1

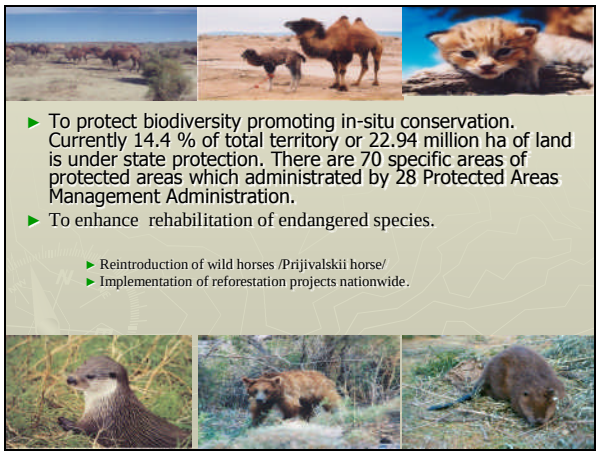
2



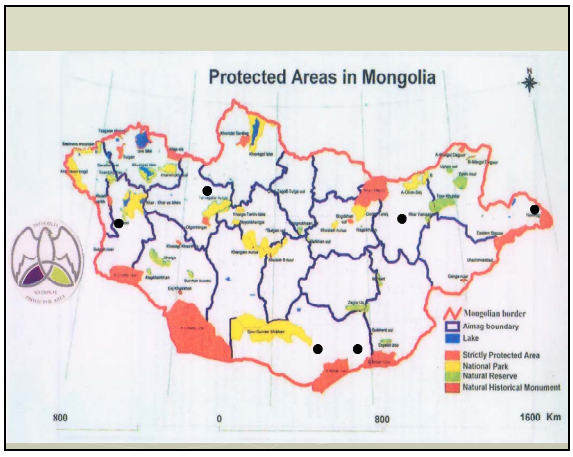
3



4


To protect biodiversity promoting in-situ conservation. Currently 14.4 % of total territory or 22.94 million ha of land is under state protection. There are 70 specific areas of protected areas which administrated by 28 Protected Areas Management Administration.
To enhance rehabilitation of endangered species.
Reintroduction of wild horses /Prijivalskii horse/
Implementation of reforestation projects nationwide.

5


Protected Areas in Mongolia
Mongolian border
Aimag boundary
Lake
Strictly Protected Area
National Park
Natural Reserve
Natural Historical Monument
800
0
800
1600 Km

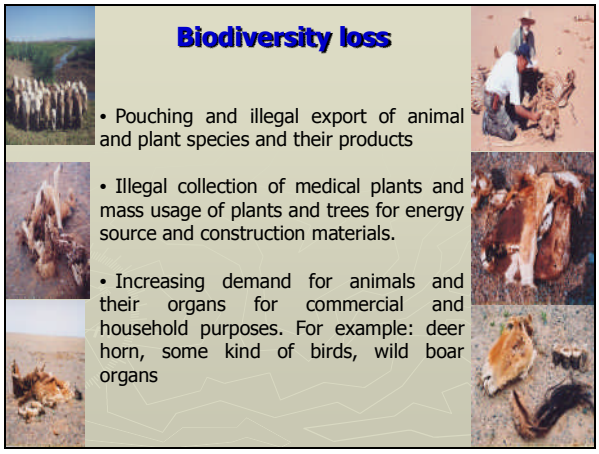
6


• To introduce community based natural resources management.
- 1.3 million ha forest protected and their plants, animals and pastureland preserved
- 541 forest local communities are existing

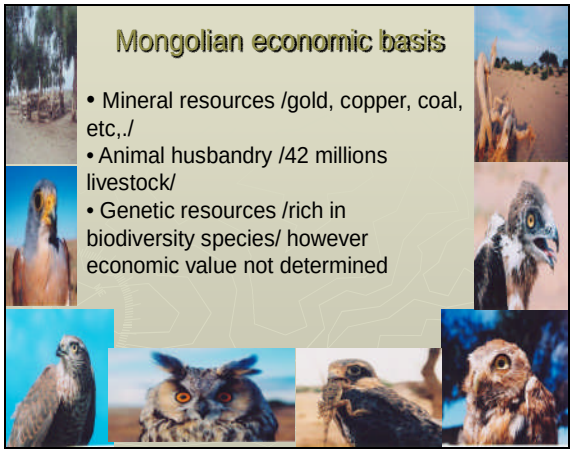
7


• In 2009, Mongolian Parliament approved the Law on restricting mining in the river basin and forest areas.
•The law provides legal base to protect biodiversity in river basin and forested areas.

8


Biodiversity loss
• Pouching and illegal export of animal and plant species and their products
• Illegal collection of medical plants and mass usage of plants and trees for energy source and construction materials.
• Increasing demand for animals and their organs for commercial and household purposes. For example: deer horn, some kind of birds, wild boar organs

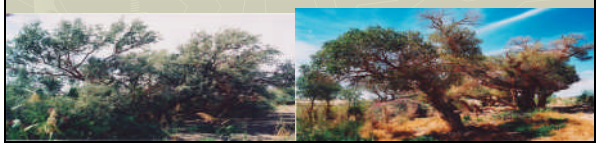
9


Mongolian economic basis
• Mineral resources /gold, copper, coal, etc,./
• Animal husbandry /42 millions livestock/
• Genetic resources /rich in biodiversity species/ however economic value not determined

10

Mongolian biological resources

- Mongolian land is about 1% of world total land territory
- 19000 species of biodiversity is recorded in Mongolia.



11

Mongolian biodiversity species compared to others

Biological Class	Number of species		
	Mongolia	Russia	World
Algae	1574	9500	40.000
Fungi	900	?	100.000
Lichen	980	3000	25.000
Moss	445	1370	12.000
High Plants	2946	11.400	270.000
Insects	615	3000	32.000
Worm	4	1000	10.000
Snake	36	2000	70.000
Crab	16	2000	40.000
Spider	451	10.000	75.000
Insects	13.000	100.000	950.000
Fish	76	669	30.000
Amphibians	8	27	5000
Reptiles	22	75	7300
Birds	468	732	9500
Mammals	132	320	4500

12

However limited studies undertaken, large animals mostly registered :

- For example:
- parasite of insects:

592 species registered in 1990
615 species registered in 2008

- Butterfly:

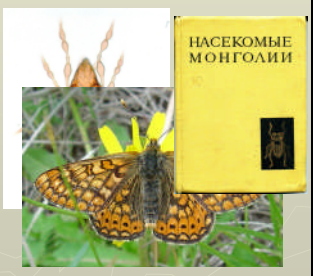
153 species registered in 1990
257 species registered in 2008

- Ant:

40 species registered in 1990
72 species registered in 2008

- soil acarus:

43 species registered in 1990
335 species registered in 2008



13

Percentage of export by main natural and genetic resources

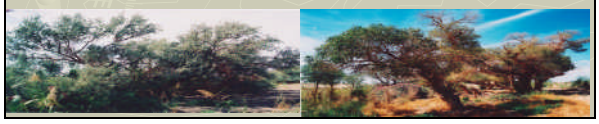
Total export:

- **copper 27.9%,**
- **coal 27.7%,**
- **Iron 8.2,**
- **gold 7.1%,**
- **crude oil 6.1%,**
- **raw cashmere 7.1%,**
- **other export 6.4 % but genetic resource is not registered.**

14

Use of biological resources

- Use for domestic household consumption
- Sport and special purpose hunting by foreign hunters
- Use plant species for medicine
- Use of plants and animals for scientific research purposes



15

Legal basis for Genetic resources

National Constitution of 1992:

Clause 6:

1. Mongolian land, its underground resources, forest, water, animal, plants and other natural resources are national property of people and subject to protection by National Government.

2. Land except owned by Mongolian citizen, its underground resources and all natural resources, forest, water resources, animals are common property of Mongolian State.

16

Law on Environmental protection:

Clause 15:

Authority of Central Government:

Article 6.

To provide environmental information for citizen, companies and organizations, and to support process for sustainable usage of biological and genetic resources and its practice of knowledge and new idea.

17

Law on Fauna, Law on Flora, Law on Hunting:

The main regulations are as follow:

- Animals and plants are classified in 3 classes:
  - Very rare
  - Rare
  - Rich
- The usage of animal and plant species are classified in 3 prurposes:
  - Commercial
  - Household
  - Special purposes and medicine production
- Those law also legalized usage, protection, rehabilitation and selection process of animal and plants.

18

Goals for Future

- Convention on Biodiversity:

1/ To protect biological and genetic resources

2/ To provide sustainable use of biological and genetic resources

3/ To distribute economic and future efficiency of biological and genetic resources as a equal.

19

Future goal

- Nagoya Protocol:

1/ To prepare process to join Nagoya protocol

2/ To complete the process of joining the Nagoya protocol this year

3/ To develop a new law on Genetic resources

20

Future goal

- Basic issue of the Law on Genetic resources:

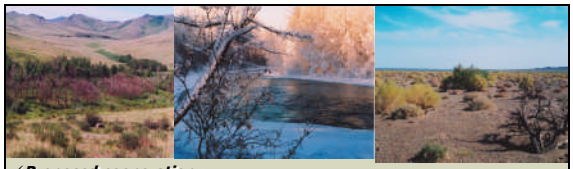
1/ To determine management power of Government and local administrations for genetic resources

2/ To establish provisional and usage permission system

3/ To establish usage payment system of genetic resources

4/ To establish legal condition of traditional knowledge of genetic resources

21



✓ ***Proposed cooperation:***

- Enhance the cooperation on capacity building for distribution of the economic and future efficiency of biological and genetic resources as a equal.
- Establish database for genetic resources and its traditional knowledge
- Participation in "SLEEPING MICROBIAL BEAUTIES " project

22

講演 2：「Access to Mongolian plants: *In vitro* bioassay screening」（発表資料 2 参照）

モンゴルの薬用植物は古くから様々な病気の治療や予防に幅広く利用されている。今日では近代分析技術やインビトロ薬理試験により、植物の特性や有効性を明確にする必要がある。その活性評価のためのインビトロ試験では少量の抽出物、フラクション、あるいは化合物が必要となる。モンゴル植物相はまだ体系的に生物学的活性を選別されていない。そこで、演者等は 2006 年よりモンゴル植物の生理活性スクリーニング（抗菌・抗酸化・抗変異活性試験、アセチルコリンエステラーゼやリパーゼ阻害活性）を実施している。以下それらの試験結果を発表した。

モンゴル植物 130 種から 350 以上のメタノール抽出物を得、生理活性を調べた。

抗菌スクリーニング（*S. aureus*、*M. luteus*、*E.coli*、*E. faecalis*、*Ps. aeruginosa*、*S. epidermidis*）では、*Larix sibirica* Ldb. (stem)、*Juniperus sabina* (aerial parts)、*Comarum Salesovianum* (aerial parts)、*Caryopteris mongolica* (root)及び *Potentilla viscosa* (leaves)が強い活性を示した。その内、*Larix sibirica* Ldb からは活性物質としてイソピメリン酸を得た。さらに、抗菌物質として、*Comarum Salesovianum*(aerial parts)からは 2 つの化合物、2-hydroxy-6-nonil benzoic acid, 2-hydroxy-6-non-8-enil benzoic acid（新規物質）を得た。

モンゴルでは家畜の炭疽菌感染（*Bacillus anthracis*）がしばしば起こり問題となっているので、炭疽菌に対する活性も調べている。*Larix sibirica* Ldb.は、抗炭疽菌活性を有している。遊牧民は昔、この木の樹脂を家畜に与えていた。

Chamaerhodos erecta (L.) Bge (aerial parts)、*Dasiphora parvifolia* (aerial parts)、*Nymphaea candida* (aerial parts)及び *Geranium pseudosibiricum* (aerial parts)は、高い抗酸化活性を有する植物であることがわかった。*Chamaerhodos erecta* (L.) (aerial parts)からは、1 新規化合物と 11 の既知化合物が単離された。

Leptopyrum fumaroides (whole plant)、*Juniperus sibirica* Burgsd (aerial parts)、及び *Pulsatilla flavescens* (leaves)は、高い抗変異活性を有していた。*Pulsatilla flavescens* (leaves)からは、12 の化合物（2 つの新規物質を含む）を得た。*Juniperus sibirica* と *Leptopyrum fumaroides* の抗酸化活性の活性成分はクロロフォルム画分の cetylic alcohol であった。

Carduus crispus L. (aerial parts)、*Bergenia crassifolia* (L.) (leaves and root)、*Juniperus communis* L. var *saxatilis* Pall. (aerial parts)及び *Patrinia rupestris* (aerial part)は、アセチルコリンエステラーゼ阻害活性を有していた。*Carduus crispus* からはいくつかの phenolic acids を単離同定した。

Agrimonia pilosa (aerial parts)、*Cotoneaster melanocarpa* (stem)及び *Pteridium aquilinum* (stem)は、強い抗リパーゼ活性を示した。

スクリーニングの結果に基づき、ハーブティーの商品化の可能性も検討した。

Access to Mongolian plants: *In vitro* bioassay screening

J.Batkhuu

Ts.Dash Memorial Laboratory of Bioorganic Chemistry and Pharmacognosy
Department of Biochemistry and Bioorganic Chemistry
School of Biology and Biotechnology, National University of Mongolia (NUM)

1

Our research interest

- Field survey on distribution and natural resources of Mongolian medicinal plants (collection and identification of plants, also educate young people in the field…)
- Preparation of extracts (Bank of Mongolian plant extracts)
- Primary screening for evaluation of biological activities by using in vitro assays (We are required to introduce assays demanding little money and big skill of young sensitive students, for example antimicrobial, antioxidant, enzyme inhibition and mutagenic)
- Selection of active plants (may be patent application for dietary supplement, cosmetics etc. with domestic and foreign companies , Monos, Monchemo, Kao, GIST, VTT…)

2

Our research interest (continued)

- Activity-guided isolation of active compounds by using simplest methods as CC and TLC
- Structure determination – NMR, MS (if we obtained pure active compounds, structure determination is possible with foreign colleagues , then may be patent application for functional food, cosmetics etc. with domestic and foreign companies , Monos, Monchemo, Kao, GIST, VTT…)
- In vitro and clinical studies → patent application for drug → production and marketing with pharmaceutical company
- Cultivation of active plants by classic method and tissue culture (cell suspension culture)
- To search ways to use plants on the basis of traditional medicine and experiment results

3

Screening methods employed in the Lab

Antibacterial Activity	Disc Diffusion Method	*S.aureus*, *M.luteus* *E.coli*, *E.faecalis*
Lipase Inhibition Assay	BALB-DTNB method	Pancreatic lipase, BALB, DTNB, Spectrometry-412nm
Anti-mutagenic Activity	Ames test	*S.typhimurium* TA1537 (his$^-$) Standart mutagen: 9-aminoacridine
Antioxidant Activity	DPPH radical scavenging method	DPPH, rutin Spectrometry-517nm
	β-carotene bleaching method	Linoleic acid, trolox, Twin40, β-carotene Spectrometry-470nm
AChE Inhibition assay	Ellman's method	AChE, ACh, DTNB, Spectrometry-412nm

4

Screening on antibacterial activity of Mongolian plants

- Since 2004
- Method: Disc diffusion assay
- Test microorganisms:
  1st screening: *S. aureus*, *M. luteus*, *E. coli*, *Ps. aeruginosa*, *E, faecalis*
  2nd screening: *S. epidermidis*, *B. anthracis*, *H. pilory*
- Positive control: Kanamycin
- 594 extracts of 226 plant species were tested. Briefly, 125 samples of 76 plants showed antibacterial active at least against one bacterial strain.

High active plants:
***Comarum Salesovianum* (aerial part)**, *Caryopteris mongolica* (root), *Agrimonia pilosa* (root), *Larix sibirca* (stem), *Potentilla viscosa* (stem)

5

Comarum Salesovianum (Steph.) Aschers. et Gr.

Diagram 1 Extraction and isolation procedure of *C. Salesovianum*

6

Antibacterial activity of *Comarum Salesovianum* (Steph.) Aschers. et Gr.

Table 1. Antibacterial activity of fractions and isolated compounds from *C. Salesovianum*

Name of sample	Inhibition zone (mm)						
	C (μg/disc)	*S. aureus*	*M. luteus*	*E. coli*	*E. faecalis*	*Ps. auruginsa*	*S. epidermidis*
80% EtOH	500	20,4±0,14	21,1±0,01	NA	12,2±0,18	NA	17,4±0,36
DCM	500	24.8±0.08	24.8±0.03	NA	16.5±1.2	NA	20.2±1
n-BuOH	500	10.9±0.01	NA	NA	NA	NA	NA
WR	500	NA	NA	NA	NA	NA	NA
Cs-1	500	33	35.2	11	28.3	9.7	37
Cs-1	50	22.1	22.6	NA	17.3	NA	12.2
KM		10	15	12,7	16,6	9	15,2

NA=no activity. Inhibition zone (in mm) including the diameter of disc (the diameter of disc = 8 mm); C=concentration; DCM=dichlorometane; n-BuOH=n-Butanol; WR=water residue; KM=Kanamycin

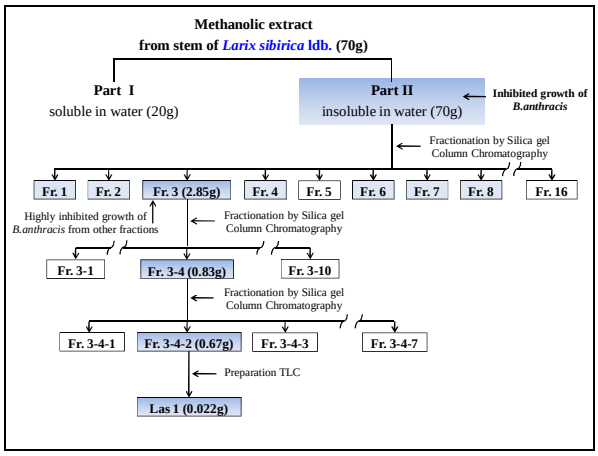
7

Anti-*Bacillus anthracis* activity of some Mongolian plants

- We tested totally 67 samples of 49 plant species on *Bacillus anthracis* strains by disc diffusion method.
- 25 samples of 19 plant species inhibited growth of *Bacillus anthracis.*
- Further we studied *Larix sibirica* Ldb.

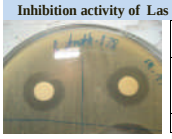
Plants		*Bacillus anthracis* strains						
Name	Parts	90	107	120	126	132	147	178
Artemisia frigida	leaf	—	10	10	8	10	—	—
	stem	—	15	8	14	10	—	—
Abies sibirica	leaf	12	—	12	—	—	13	12
	stem	14	—	13	—	—	12	12
Betula platyphylla	leaf	10	—	13	—	—	13	14
Caryopteris mongolica	root	20	—	18	—	—	18	19
Chelidonium majus	stem	12	—	13	—	—	13	13
Ephedra sinica	leaf	12	—	11	—	—	11	11
Ferula Bungeania	stem	13	—	14	—	—	10	11
Ferula feruloeoides	root	10	—	14	—	—	13	14
Juniperus sabina	leaf	—	15	18	13	12	—	—
	stem	—	16	13	15	15	—	—
Juniperus sibirica	leaf	—	15	16	16	13	—	—
	stem	—	16	0	20	15	—	—
Pinus sylvestris	leaf	—	16	0	21	16	—	—
	stem	—	10	14	18	17	—	—
Larix sibirica	**stem**	**11**	**0**	**10**	**11**	**12**	**11**	**11**
Pulsatilla flavescens	stem	10	—	10	—	—	0	13
	leaf	11	—	12	—	—	0	13
Polygonum divaricatum	areal part	10	—	10	—	—	10	10
Rhododendron dahuricum	leaf	-	0	14	0	15	—	—
Sanguisorba officinalis	root	—	13	11	12	12	—	—
Spiraea salicifolia	leaf	10	—	10	—	—	10	10
Trollius asiaticus	areal part	14	—	12	—	—	13	14
Vaccinum vitis-idaea	leaf	—	0	11	0	0	—	—

8



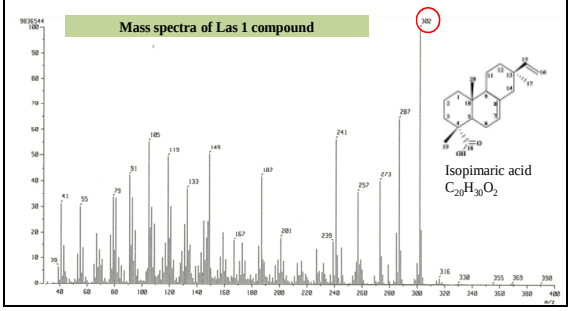
9

Inhibition activity of Las 1 compound of the *Bacillus anthracis* (clear zone, mm)

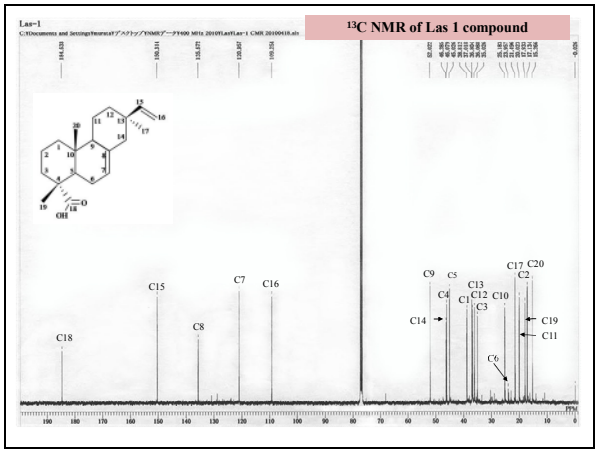


Sample	Conc	*Bacillus anthracis* strains			
		90	120	147	178
Las1	500μg/ disc	17	15	15	18
		17	15	17	17
		17	17	15	17
Control (amp)		36			

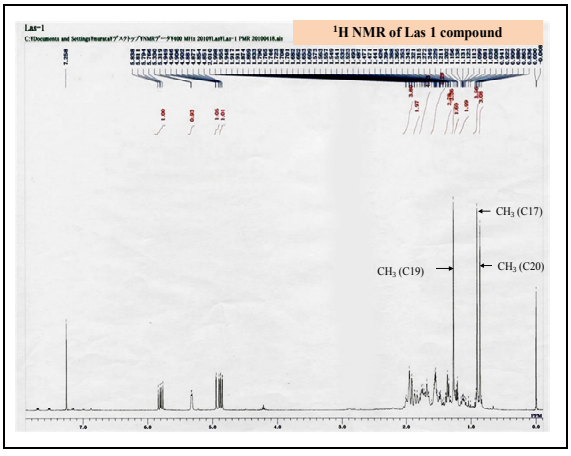
Mass spectra of Las 1 compound



10



11



12

Screening on anti-mutagenic activity of Mongolian Plant

1st screening for "Antimutagenic activity of Mongolian Medicinal Plants"

Since 2007
Method: Modified Ames test
Test strain: *Salmonella typhimurium* TA1537 (His$^-$)
Standart mutagen: 9-aminoacridine (20μg/plate)
Inhibition ratio: (IR)=A-B/C × 100

9-aminoacridine (9AA)

Screened samples: 141 different samples of 82 plant species.
(100μg/plate, 10%DMSO)

Chamaneron angustifolium (green leaves),
Rubus Sachalensis (stem)
***Pulsatilla flavescens* (leaves)**
Arabis pendula L. (stem)
***Leptoperum fumaroides* L. Reichb (A.part)**
***Juniperus sibirica* (leaves)**
Rhinanthus songaricus (leaves, stem)

Antimutagenic activities were IR>40%

13

Isolation of active compounds *Pulsatilla flavescens* leaves

N:48°05.159', E:106°18.433'
H:1374m (2nd June 2010)

Dry leaves 680g of P.flavescens

Methanol extract 215g

Use in The Mongolian Traditional Medicine:
Ulcer pus, Heals infected wounds, Neutralize poisoning, Gland tuberculosis (scrofula), Hemorrhoid

Hexane, (5.7g) | **EtOAc, (17.8g)** | **Water , (175g)**

EA (8g) | Water (20g)

MPLC, Hi-Flash silicagel column, 60x180mm, $CHCl_3$-MeOH

OCC , Diaion HP-20, Sol : H2O→EtOH

EA1.. EA5.. EA8.. EA10.. EA14

FR1 FR2 FR3 FR4 FR5

MPLC condition:
Column: silicagel column,
Sol: Hex-EtOAc or $CHCl_3$-MeOH
F.rate: 20ml/min
Det: UV 254nm

Comp 1 (73mg)
Comp 2 (20.2mg)
Comp 3 (18mg)
Comp 4 (78mg)
Comp 5 (17mg)
Comp 6 (4.5mg)

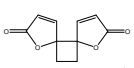
HPLC condition:
Column: L-column HB, ODS 10x250mm, 5μm
Sol: A - 0.1% HCOOH, B - CH_3CN
Isocratic 20%B
F.rate: 1.2ml/min (× 3)
Det: UV 254nm and IR-500
Inject: 10mg/400μl × 8

Comp 7 (176mg)
Comp 8 (53.7mg)
Comp 9 (41.6mg)
Comp 10 (5.4mg)
Comp 11 (12mg)
Comp 12 (29mg)

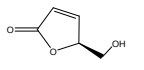
Antimutagenic activity of fraction were tested on *Salmonella typhimurium* TA98, B[a]P with S9, AF-2 with buffer, Average ±SD

14

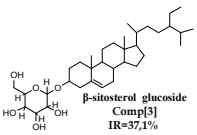
Molecular structure and anti-mutagenic activity of isolated compounds from *P.flavescens*



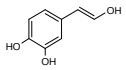
Anemonin
Comp[1]
IR=61,8%



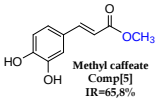
Dihydro-5-(hydroxymethyl)-2(3H)-furanone
Comp[2]
IR=9,3%



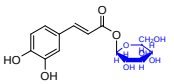
β-sitosterol glucoside
Comp[3]
IR=37,1%



Caffeic acid
Comp[4]
IR=55,6%



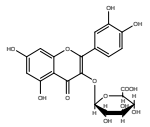
Methyl caffeate
Comp[5]
IR=65,8%



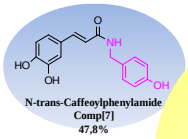
Caffeic acid β-glucopyranose
Comp[6]
IR=11,8%

15

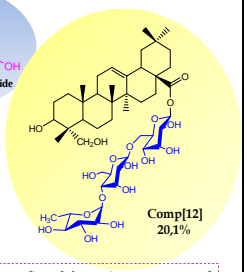
Molecular structure of isolated compounds



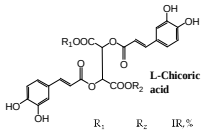
Quercetin 3-O-β-D-glucoronide
Comp[11]
IR=89,6%



N-trans-Caffeoylphenylamide
Comp[7]
47,8%



Comp[12]
20,1%



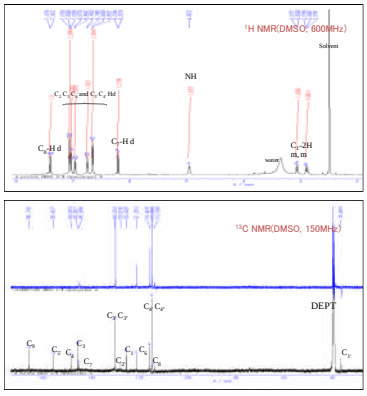
L-Chicoric acid

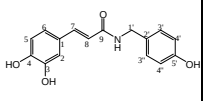
	R_1	R_2	IR,%
Comp[8]	H	H	44,2%
Comp[9]	CH_3	H	83,3%
Comp[10]	CH_3	CH_3	72,3%

Our study confirmed that major components of leaves of *Pulsatilla flavescens* were anemonin, caffeic acid related compounds and triterpene saponins which could be responsible for various biological activities of the plant.

16

^{1}H and ^{13}C NMR spectrum of N-trans-caffeoylphenylamide

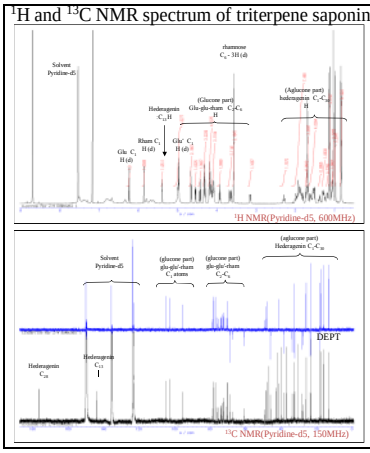


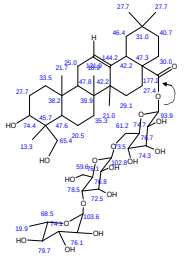


N-trans- caffeoylphenylamide

17

^{1}H and ^{13}C NMR spectrum of triterpene saponin

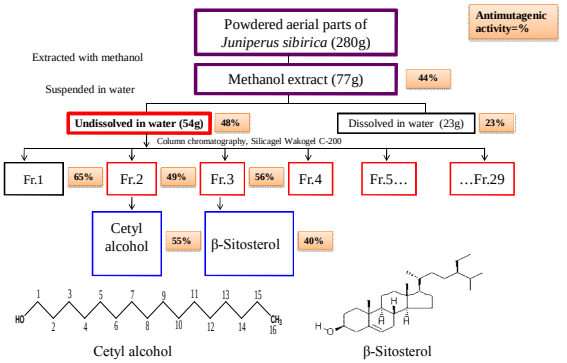




Hederagenin
28-O-α-L-rhamnopyranosyl
(1→4)β-D-glucopyranosyl(1→4)
β-D-glucopyranosyl ester
$C_{48}H_{78}O_{18}$

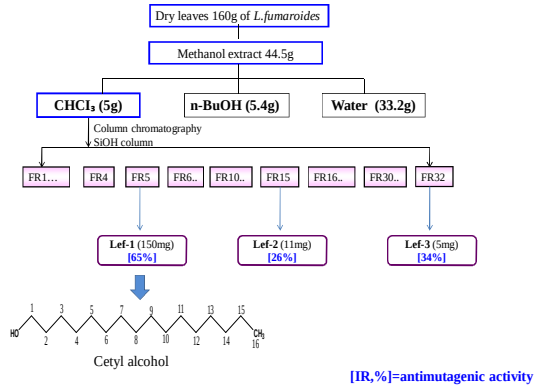
18

Extraction, fractionation and column chromatography separation of *Juniperus sibirica* aerial parts



19

Fractionation and Isolation of *Leptoferum fumaroides* (aerial part)



[IR,%]=antimutagenic activity

20

Screening on antioxidant effect of Mongolian plant extracts

DPPH radical scavenging method	β-carotene bleaching method
• Since 2007 • We studied 324 Mongolian plant methanol extracts belonging to 186 species and we evaluated their inhibitory effect against free radicals.	• Since 2010 • By Miller (1971) β-carotene bleaching method • Auto-oxidation of Linoleic acid • 60 plant extracts belonging 37 species of 18 families.

High antioxidant active plants:
***Chamaerhodos erecta**, Agrimonia pilosa, Ephedra sinica, Cotoneaster melanocarpa Lodd, Nympaea candida, Dasiphora parvifolia (Fisch.) Juz.*

21

Some high antioxidant active plants

Chamaerhodos erecta
•Upper part IC50 (μg/ml) = 59.24±0.04

Ephedra sinica
•Leaf IC50 (μg/ml) = 52.38±1.91

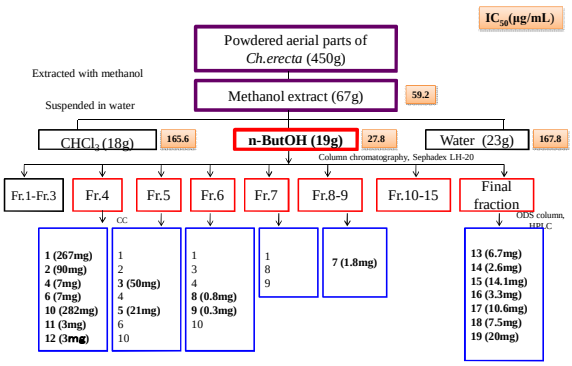
Geranium pseudosibiricum J. Mayer
•Leaf IC_{50} (μg/ml) = 51.18±0.2

Geranium pratense L.
•Upper part IC_{50} (μg/ml) = 52.12±0.69

Vaccinium vitis-idaea L.
•Stem IC_{50} (μg/ml) = 52.52±0.22

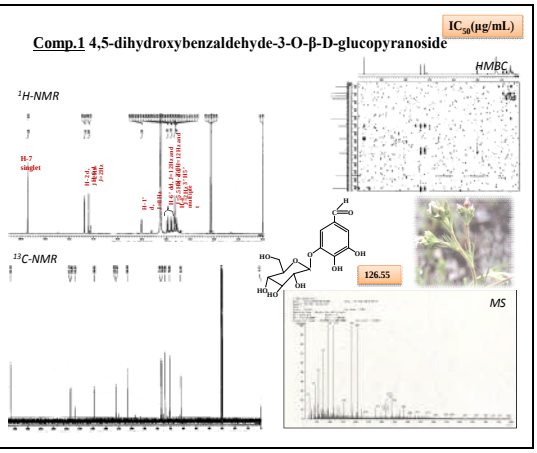
22

Isolation and chracterization of antioxidant compounds from aerial parts of *Chamaerhodos erecta*

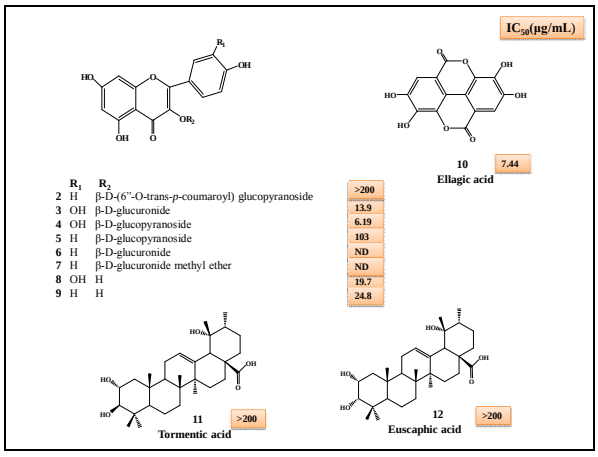


23

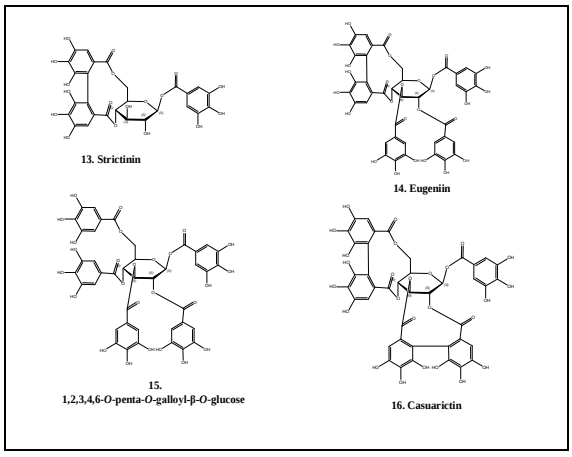
Comp.1 4,5-dihydroxybenzaldehyde-3-O-β-D-glucopyranoside



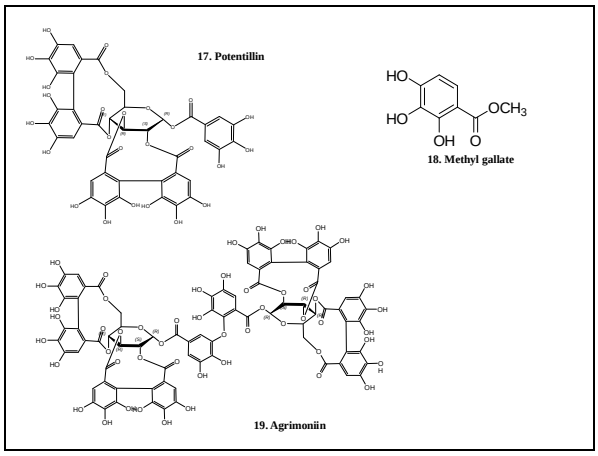
24



25



26

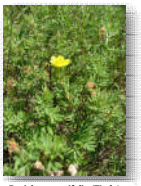


27

Extraction and fractionation of *Dasiphora parvifolia*

- We selected leaf extract of *Dasiphora parvifolia* (Fisch.) Juz. based on results.

Fig 1. Leaf of *Dasiphora parvifolia* (Fisch.) Juz. fractionated by liquid-liquid partition.



Dasiphora parvifolia (Fisch.) Juz.

Dasiphora parvifolia Fisch. Juz (Leaf)
with MeOH at room temperature (W/V, 1:10)
Extraction for 5 days
Filtration
Concentrated with a rotary evaporator
Methanol extract (37 g)
Dichloromethane (7 g) — IC_{50}=67.3±0.3
n-butanol (17 g) — IC_{50}= 33.4±0.5
Water residue (13 g) — IC_{50}=200<
Isolate active compounds

28

Screening on acetylcholinesterase inhibition activity

Since 2009
Method: Modified version of the colorimetric method of Ellman.
Screened samples: 134 different samples prepared from 82 plant species.

Active plants:
Carduus crispus (leaves) activity=81.6%
Patrina rupestris (aerial part) activity=49.6%
Artemisia annua (flowers) activity=52.1%
Filipendula palmata (seeds) activity=63.8%

MeOH extract
CHCl3 | n-BuOH | water
Isolation
Column chromatography
β-sitosterolsterol

29

Investigation on acetylcholinesterase inhibitor effect of *Carduus crispus*

Methanol extract 44.5g

Hexane	CHCl3	EtOAc	n-BuOH	Water
[34.5%]	[40.1%]	[48.0%]	[81.6%]	[45.7%]

Isolation
RP-HPLC

compound	R	R1	IC_{50}(μg/ml)
neochlorogenic acid	H		400
p-coumarylquinic acid	H		205
chlorogenic acid		H	>1000

Pure compounds:
1 neochlorogenic acid
2 p-coumarylquinic acid
3 chlorogenic acid

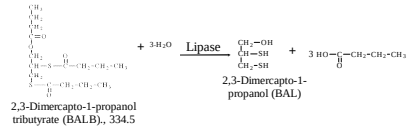
[AchE inhibitor effect,% at 1000μ/ml]

30

Evaluation of lipase inhibitor activity of Mongolian plant extracts

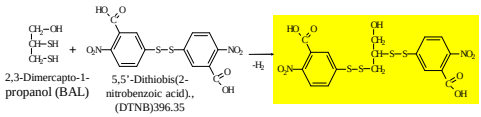
Method: BALB-DTNB method

Reaction mechanism:



2,3-Dimercapto-1-propanol tributyrate (BALB)., 334.5

2,3-Dimercapto-1-propanol (BAL)



2,3-Dimercapto-1-propanol (BAL)

5,5'-Dithiobis(2-nitrobenzoic acid)., (DTNB)396.35

31

Evaluation of lipase inhibitor activity of Mongolian plant extract

- Since 2006
- We screened124 plant extracts from prepared 81 plant species
- Active plants:
  Cotoneaster mongolica (stems 55%), *Pteridium aquilinum* (stems 36.8%), *Ephedra equistina* (leaves 68.3%), *Abies sibirica* (leaves 16%) showed higher anti-lipase activity than other extracts.

Active-guided isolation of active compounds from *Cotoneaster mongolica* (stems) is going on.

32

Possibilities of making product based on our screening results

Herbal tea

- Herbal teas have been used for daily needs since ancient times.
- All types of teas, including green, black, red and herbal teas, have antioxidant properties because of the polyphenols.
- These antioxidants in herbal tea are known to provide the body with protection against free radicals.
- Therefore, the aim of this study was to investigate antioxidant activity of herbal tea infusions prepared from some Mongolian medicinal plants using DPPH free radical scavenging method.

Preparation against smell of armpit sweat

Staphyllacoccus epidermidis

***Comarum Salesovianum* (aerial part) showed high activity against *S. epidermidis*.**

33

Composition of herbal tea infusion

We chose following plants based on their high antioxidant activity.

Fig. 1. *Chamaenerion angustifolium* (L.) Scop.

Fig. 2. *Dasiphora parvifolia* (Fisch.) Juz.

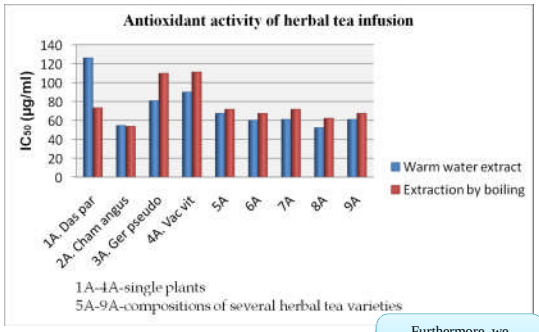
Fig. 4. *Vaccinium vitis-idaea* L.

Fig. 3. *Geranium pseudosibiricum* J. C. Mayer.

Herbal tea preparation:
We mixed these plants with various ratios
1.200 ml of hot water (95℃) was added to 2 g of dried herbs mixture and extracted it for 15 min at room temperature.
2.200ml water was added to 2g of dried herbs mixture and boiled it for 15 min.
Measure antioxidant activity

34

Fig 1. Antioxidant activity of herbal tea infusion



Furthermore, we selected 2A and 5A extracts based results.

35

Tab. 1. Antioxidant activity of 2A, 5A herbal tea infusion and other commercial herbal tea

№	IC_{50} values (μg/ml)
5A	54.06±0.4
2A	67.29±0.13
Seabuckthorn herbal tea	100.54±1.03
Chinggis Khaan herbal tea	200<
Huvsgul herbal tea	164.47±0.54
Karkade herbal tea	195.6±3.08

36

Introduction of Ts.Dash Memorial Laboratory of Bioorganic Chemistry and Pharmacognosy, NUM

1991	Tne laboratory was established with keen effort by prof . Ts.Dash as Laboratory of Protein Chemistry under the Institute of Chemistry, Mongolian Academy of Science
1997	moved to Faculty of Biology, NUM
1997	Investigation on biological active metabolites of Mongolian plants is started
2002	Screening of Mongolian plants for biological activities
2010	The lab named Ts.Dash Memorial Laboratory of Bioorganic Chemistry and Pharmacognosy
2011	Renewal open of Laboratory of Plant Biotechnology at this lab (Obtaining of secondary active metabolites by cell suspension culture)

37

Domestic collaboration:

- Department of Plant Taxonomy, Institute of Botany, Mongolian Academy of Science
- Laboratory of Natural Product Chemistry, Institute of Chemistry and Chemical Technology, Mongolian Academy of Science
- National Center for Infectious Diseases with Natural Foci, Ministry of Health
- Monos Co., Ltd
- Otoch Manaramba University

38

International collaboration:

Japan: Tohoku Pharmaceutical University
Toyama University
Toho University
Kao Co., Ltd
Regional Promotion Support Center, Japan

Austria: Graz University

China: Inner Mongolia University (Institute of Macromolecular Chemistry and Mongolian Medicine)
Peking University (Lab of Pharmacognosy, School of Pharmaceutical Sciences)

Republic of Korea: Gwangju Institute of Science and Technology

Finland: Sector of Plant Biotechnology, Technical Research Centre

Belarus: Institute of Biology, Academy of Sciences

39

Thank you for your attention

E-mail:bathuu@mobinet.mn
jbatkhuu@hotmail.com

40