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Basic evidence for the efficacy of Japanese Kampo medicine

Antitussive principles of *Glycyrrhizae radix (licorice)*, a main component of the Kampo preparations Bakumondoto (Mai-men-dong-tang)

Junzo Kamei, Ph.D.
Professor
Department of Pathophysiology and Therapeutics
School of Pharmacy and Pharmaceutical Sciences
Hoshi University, Tokyo Japan.

星薬科大學
HOSHI UNIVERSITY

Introduction

Kampo treatment in daily clinics at present can be classified into 3 major forms:

1) the use of Kampo prescriptions according to the methodology of traditional medicine.
2) the administration of Kampo medicines based on the concept of diseases/pathological conditions in Western medicine and concept of elementary Kampo medicine.
3) the evidence-based administration of Kampo medicines.

However, Kampo medicines have other paradigms that differ from Western medicine. Therefore, the characteristics and specificity of Kampo medicine must be comprehended based on basic and clinical evidence. The clinical evidences concerning “Kampo” has been accumulated year by year over two decades.

However, the mechanisms of action of Kampo is not well established.

To improve the reliability of the Kampo medicine, integration of the basic evidences, such as mechanism of action of Kampo, is important.

As an example of basic evidence for the efficacy of Kampo, the antitussive constituents of *Glycyrrhizae radix* (licorice), a main component of the Kampo preparations Bakumondoto will discuss.

Components of the Kampo preparations Bakumondoto

Bakumondoto has been used to treat severe dry cough in patients with bronchitis and pharyngitis, etc.

Bakumondoto consists of six herbs

Ophiopogonis tuber
Glycyrrhizae radix (licorice)
Pinelliae tuber
Zisyphi fructus
Ginseng radix
Oryzaeae fructus

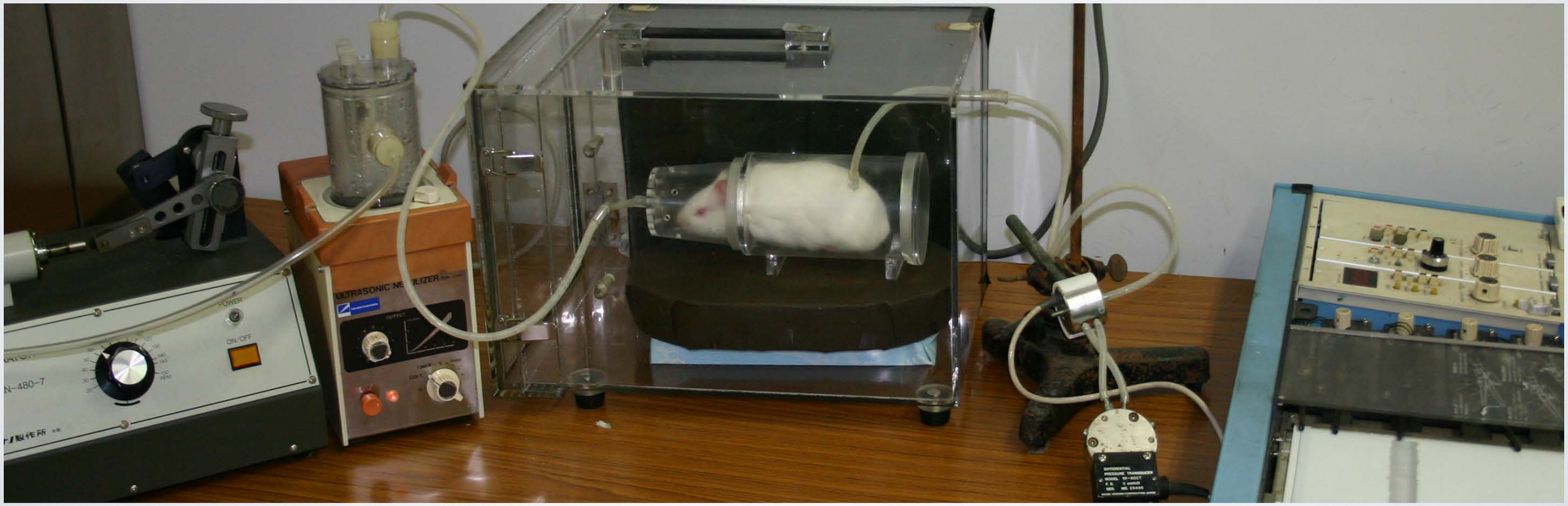
Glycyrrhizae radix (licorice) is a main component of Bakumondoto. It has been used since ancient Egyptian times as a drug for the respiratory organs, and has long been used as a flavoring and sweetening agent as well as a drug for analgesic and expectorant in Europe. Furthermore, licorice has been used as a very important crude drug in many Kampo preparations and acts as an antispasmodic, carminative and antidote, and is also taken for bronchial problems, coughs, mucous congestion, stomach problems, such as peptic ulcers, and for bladder and kidney ailments.

There has been no previous detailed study on the antitussive principle, since previous studies on Licorice have mostly focused on glycyrrhizin, one of its components. In the present study, we attempted to elucidate the antitussive constituents of licorice.

Experimental setup for cough measurement

Male Hartley guinea pigs weighing about 300-350 g.

Animals were exposed to a nebulized solution of capsaicin under conscious and identical conditions using a body plethysmograph. The transducer was connected to a respiratory amplifier and polygraph to record a respiratory pattern. The transducer was also connected to respiratory analyzer and to a personal computer which was used for on-line breath-by-breath measurement. The coughs produced during 7-min exposure period were counted.

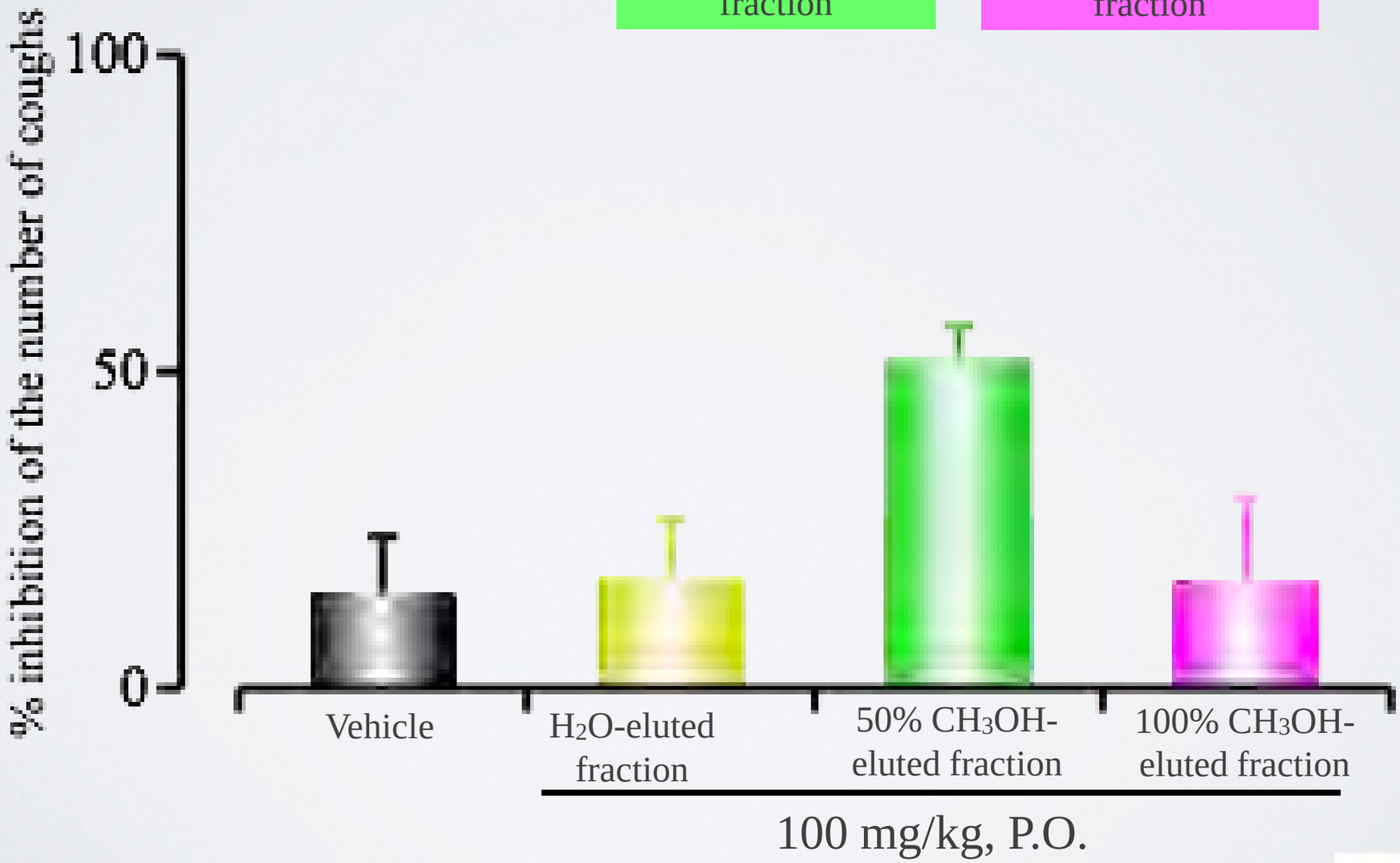


Active components of Glycyrrhizae radix (licorice)

Antitussive effect of the extract of water fraction of Glycyrrhizae radix (licorice)

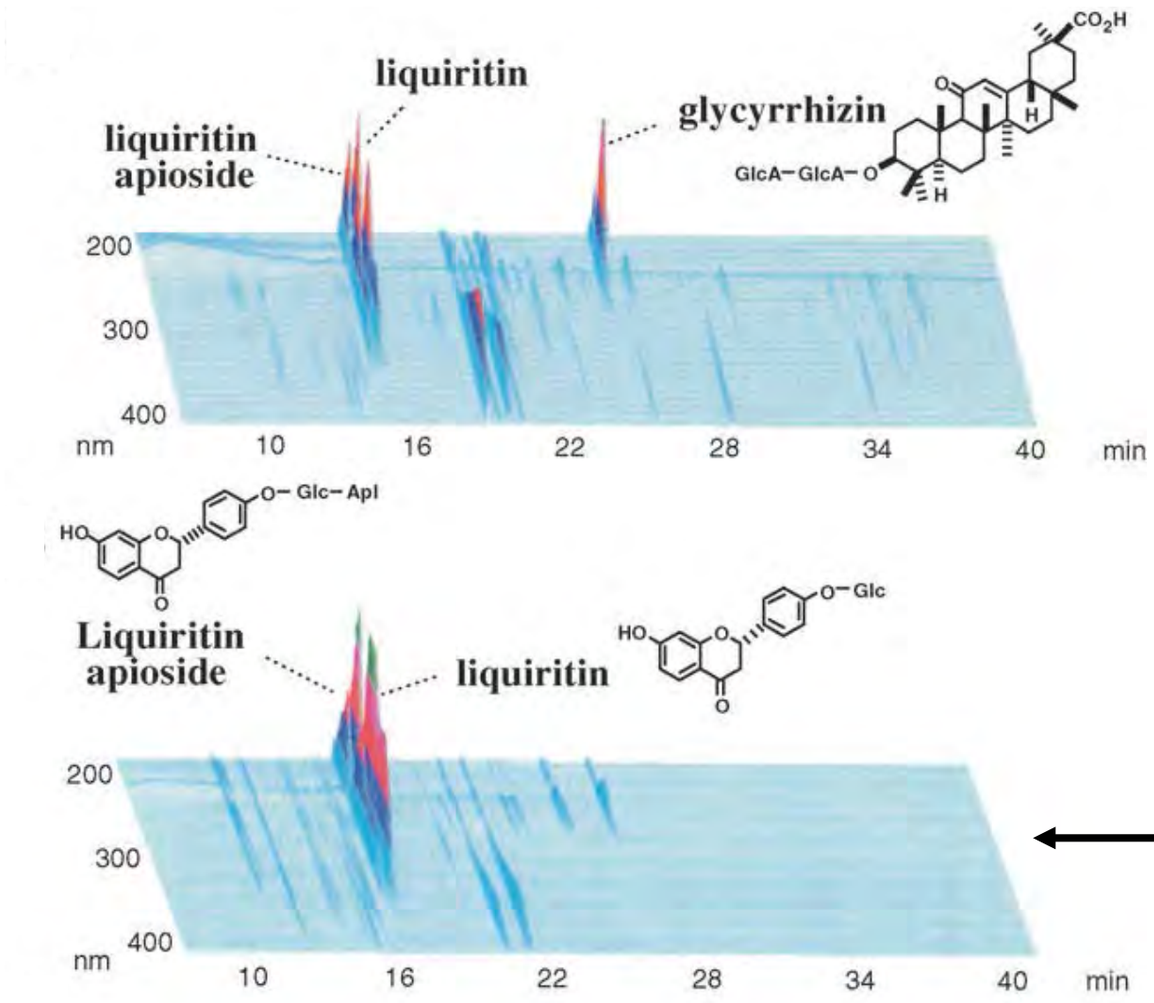
Water fraction of Glycyrrhizae radix (licorice)

- H_2O-eluted fraction
- 50% CH_3OH-eluted fraction
- 100% CH_3OH-eluted fraction



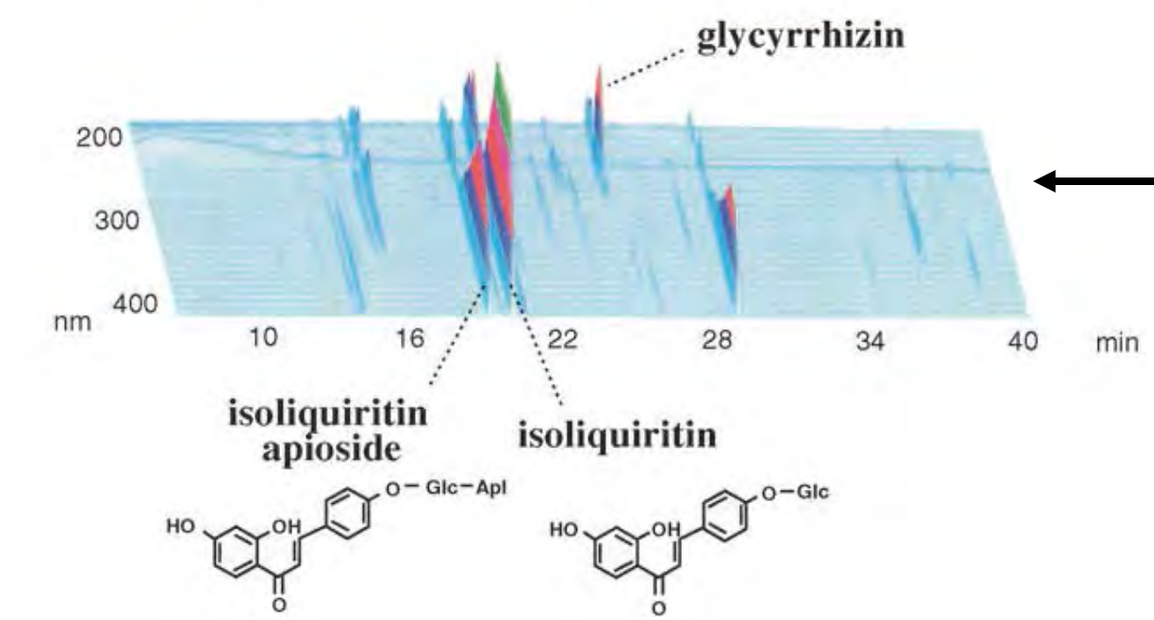
3D-HPLC analysis of the water fraction of Glycyrrhizae radix (licorice), 50% methanol-eluted fraction and 100% methanol-eluted fraction of the water fraction of Glycyrrhizae radix (licorice).

Water fraction of Glycyrrhizae radix (Licorice)

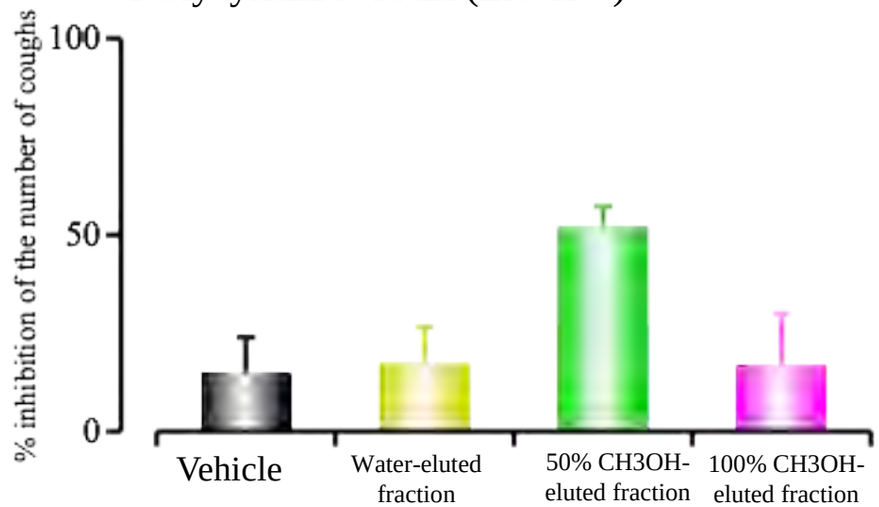


50% methanol-eluted fraction

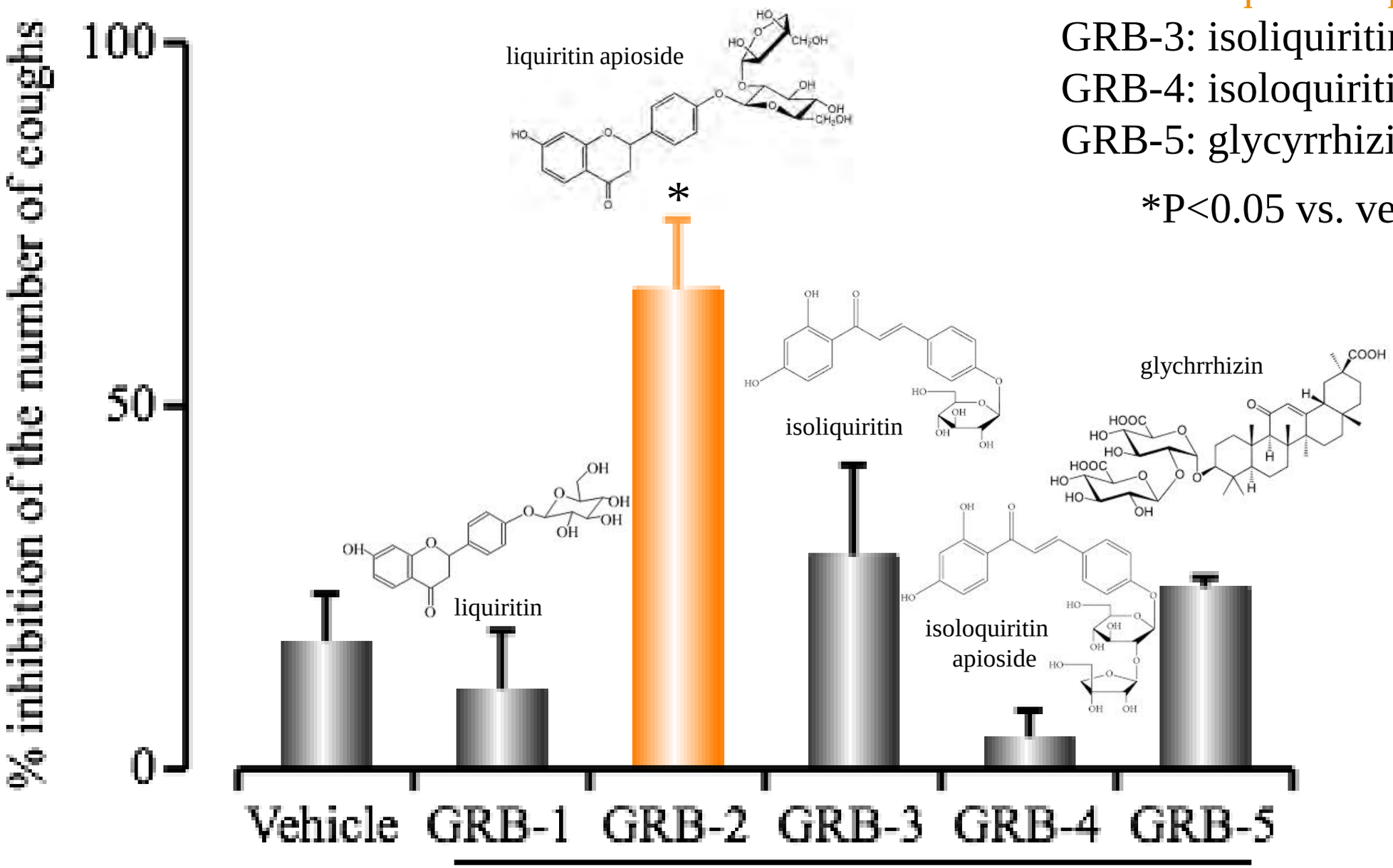
100% methanol-eluted fraction



Antitussive effect of the extract of water fraction of Glycyrrhizae radix (Licorice)



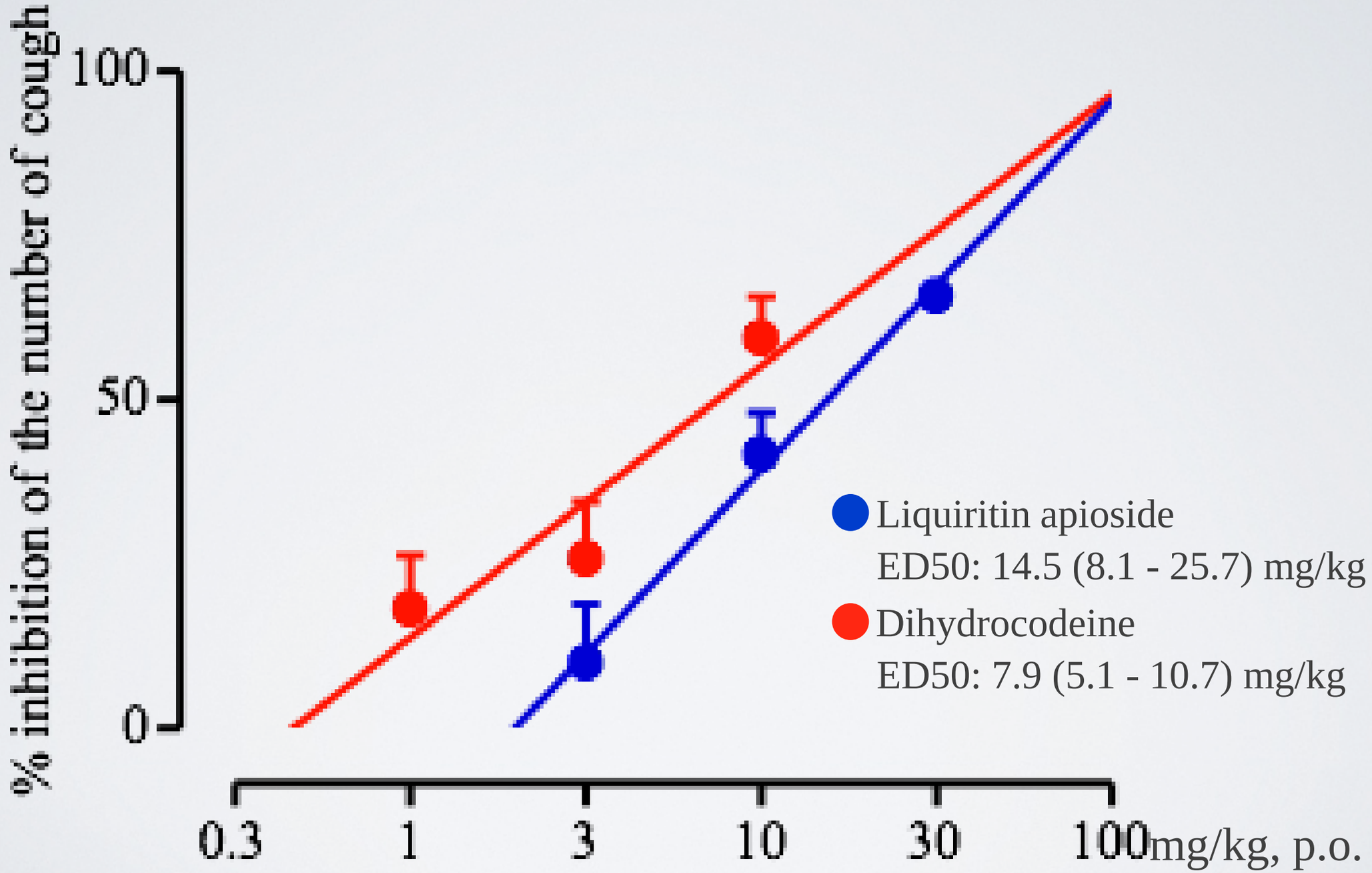
Antitussive effects of the bioactive compounds from Glycyrrhiza radix (licorice)



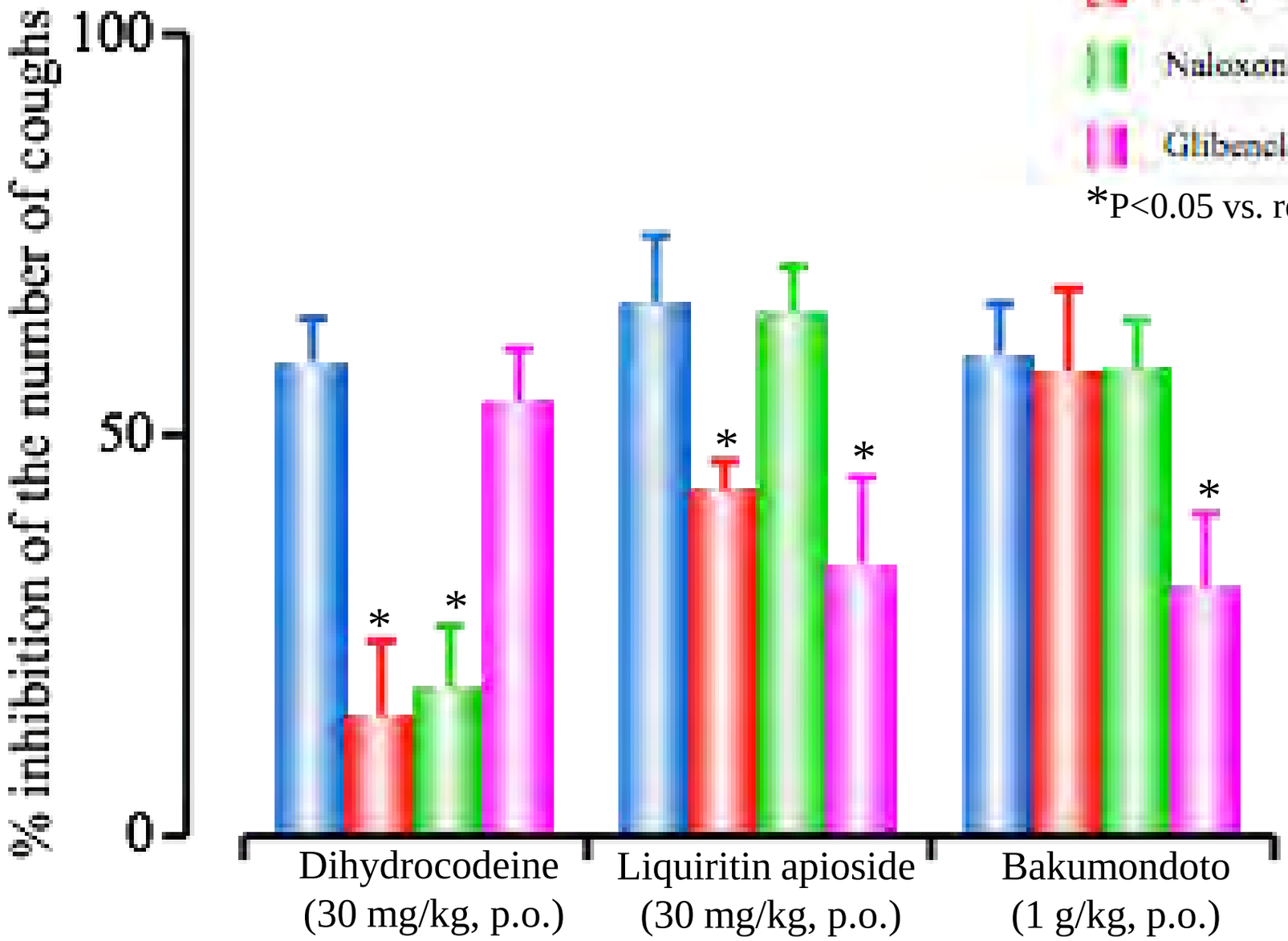
GRB-1: liquiritin
GRB-2: liquiritin apioside
GRB-3: isoliquiritin
GRB-4: isoloquiritin apioside
GRB-5: glycyrrhizin

*P<0.05 vs. vehicle

Dose-response curve for the antitussive effects of liquiritin apioside and dihydrocodeine

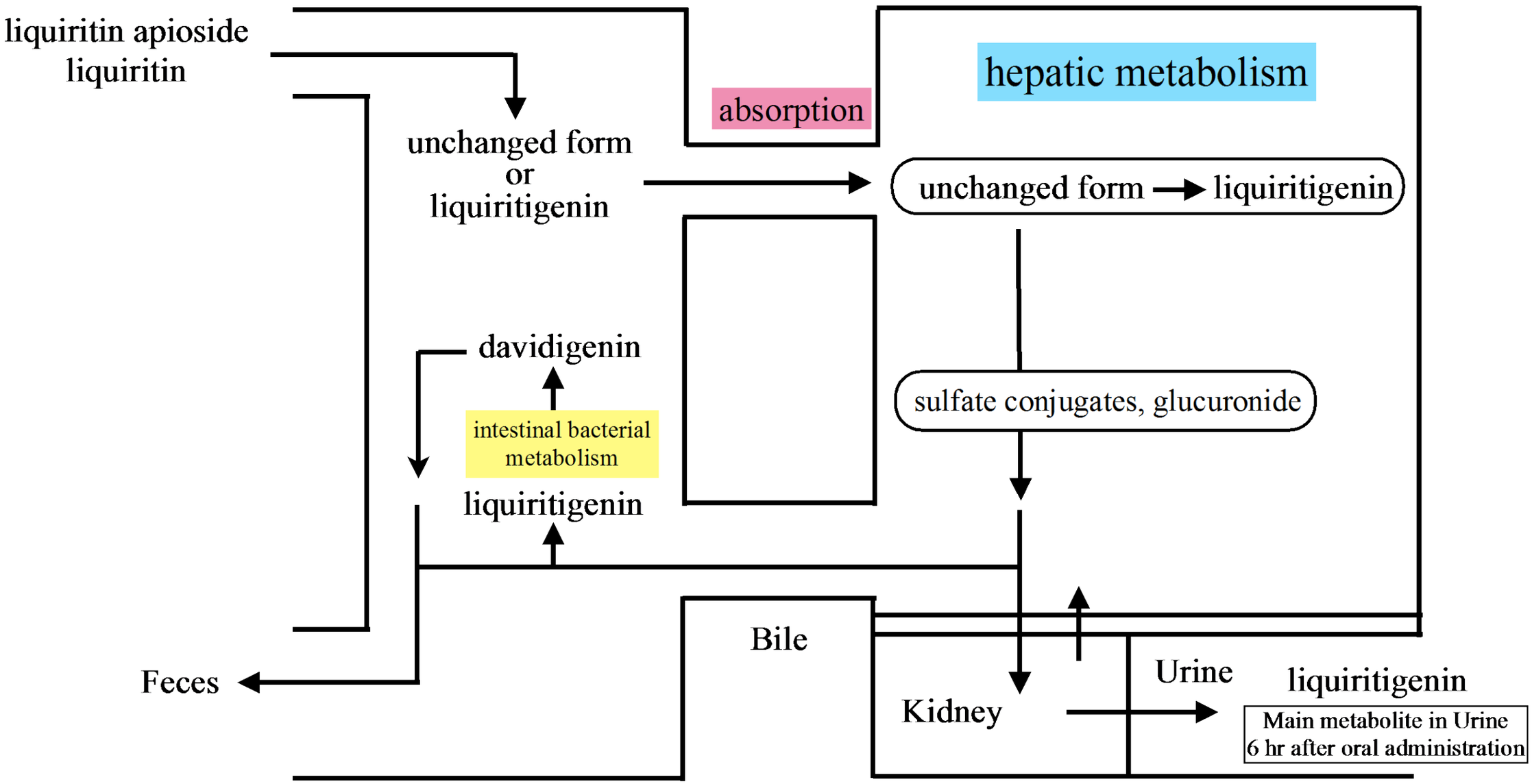


Effects of methysergide, 5-HT receptor antagonist, Naloxone, opioid receptor antagonist, and glibenclamide, ATP-sensitive K+ channel blocker, on the antitussive effect of dihydrocodeine, liquiritin apioside and Bakumondoto

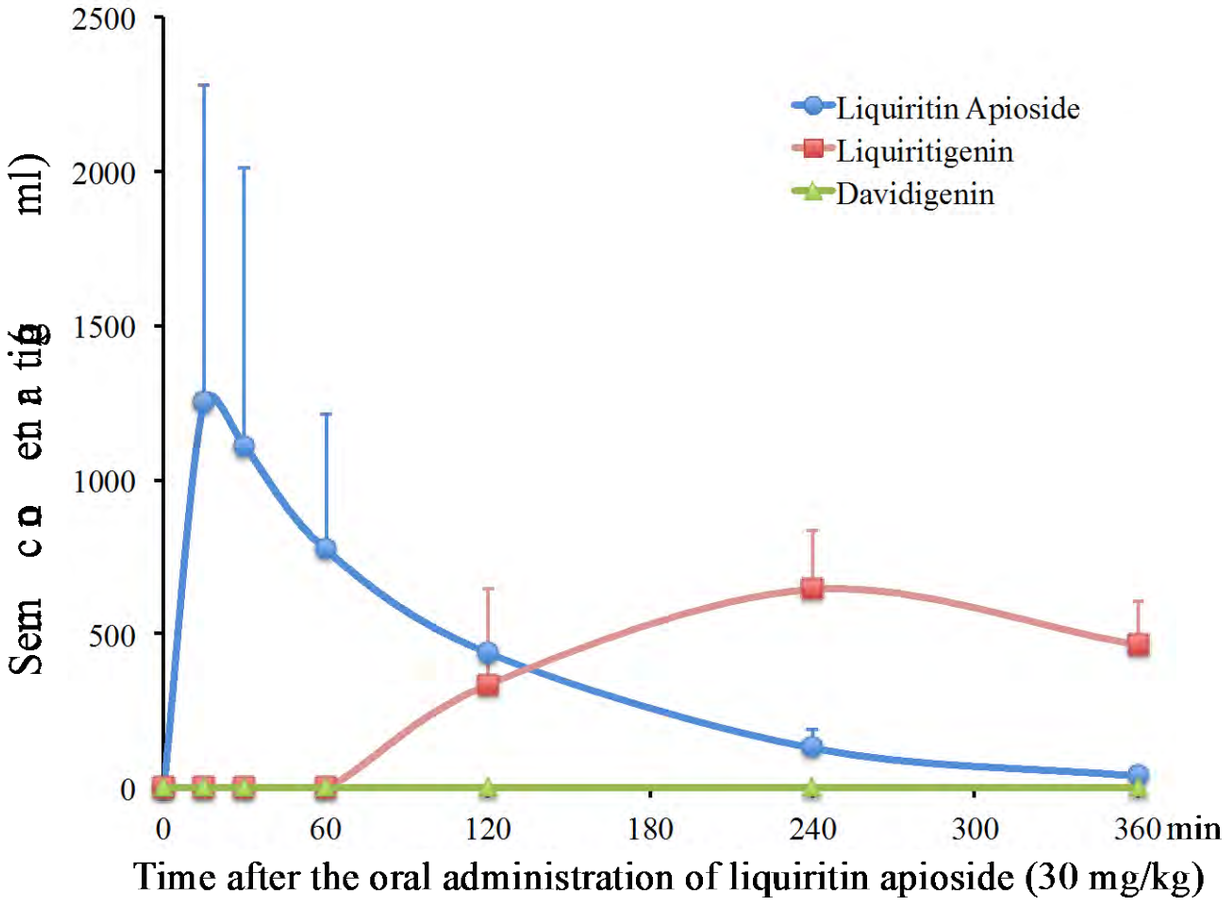


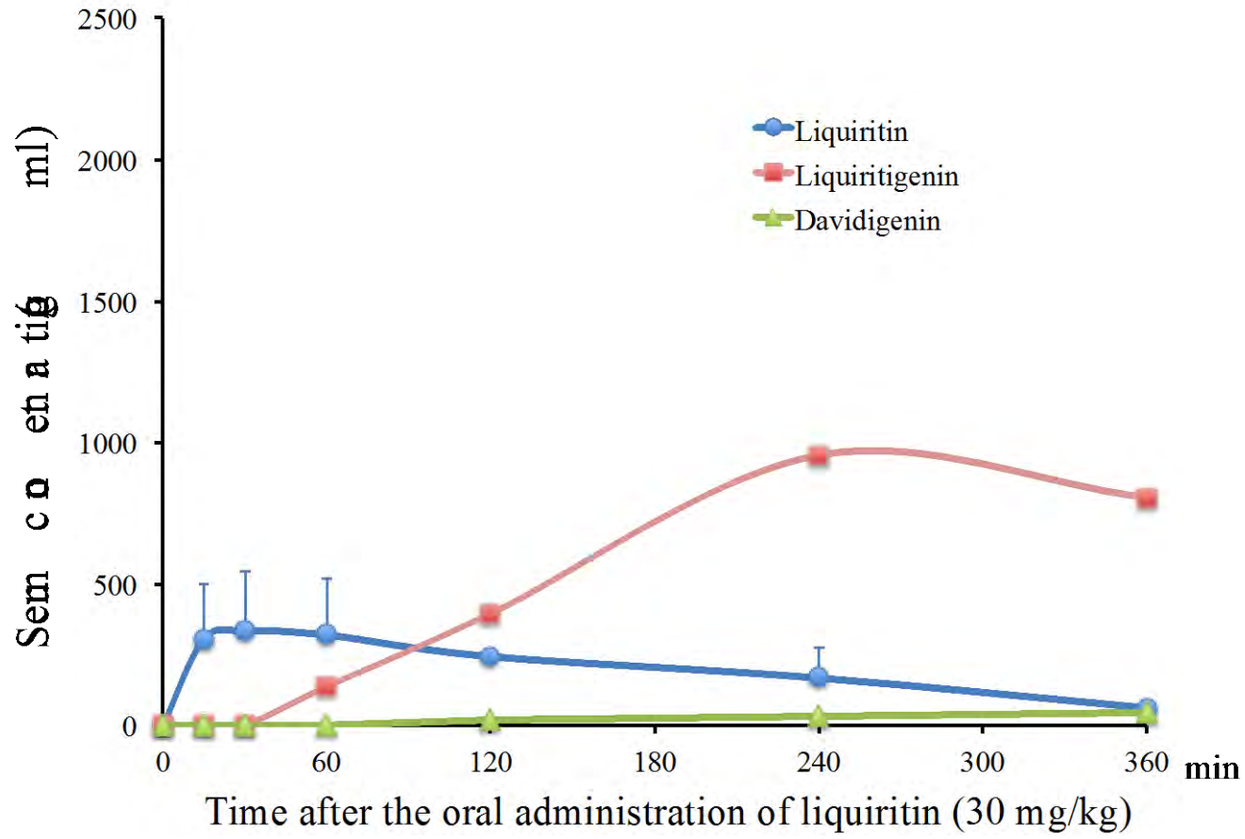
*P<0.05 vs. respective saline-treated group

Estimated biotransformation of liquiritin apioside and liquiritin, active components of Glycyrrhizae radix (licorice)



Plasma concentration–time curves of unchanged forms and their metabolites after liquiritin apioside and liquiritine administration





Time of C_{max} (t_{max}) of liquiritin, liquiritin apioside and liquiritigenin in guinea pigs after oral administration of liquiritin and its glycosides

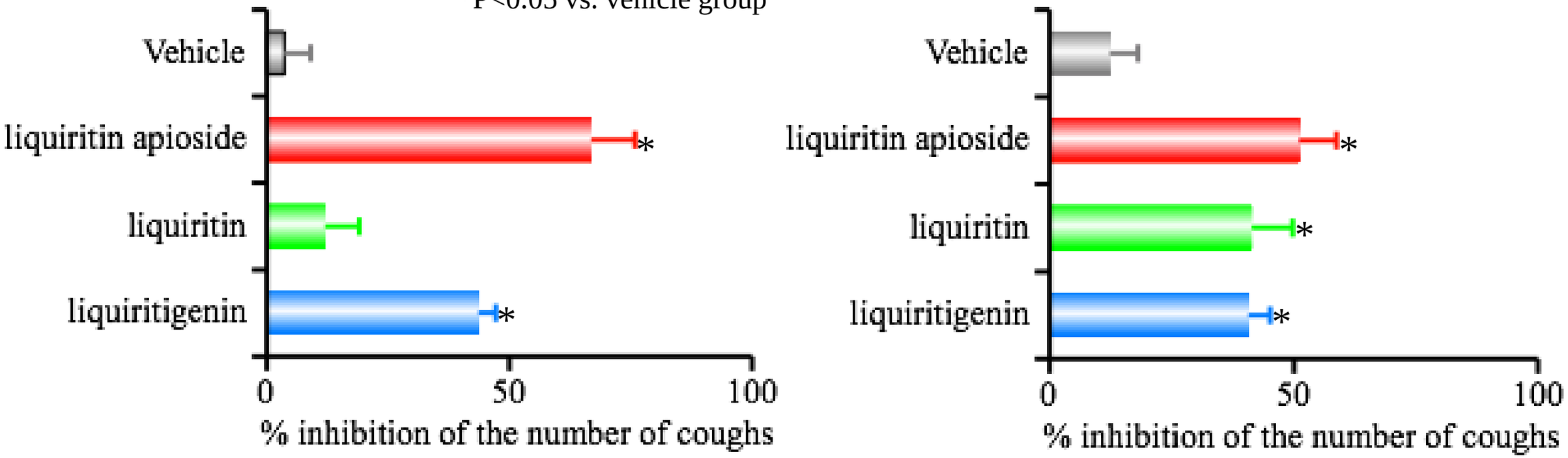
Compound	liquiritin apioside		liquiritin	
t_{max}	liquiritin apioside	0.5 h	liquiritin	1.3 h
	liquiritigenin	3.1 h	liquiritigenin	5.5 h

Antitussive effects of liquiritin apioside, liquiritin and liquiritigenin

1 hr after administration

4 hr after administration

*P<0.05 vs. vehicle group

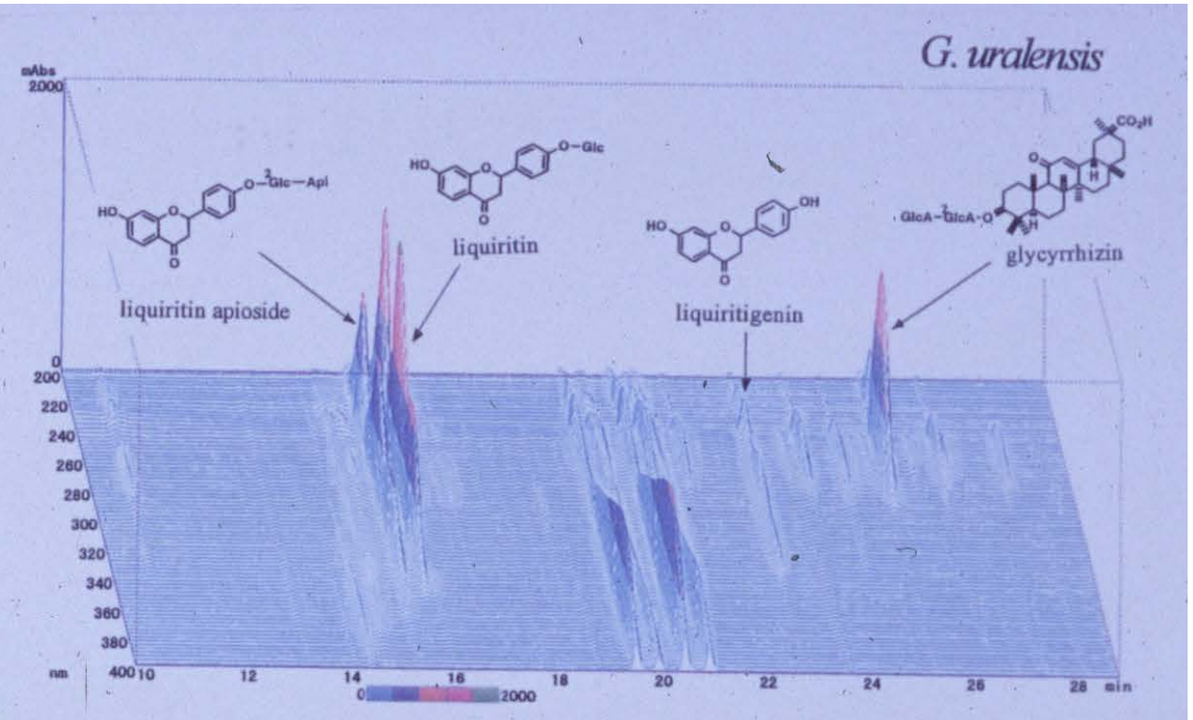


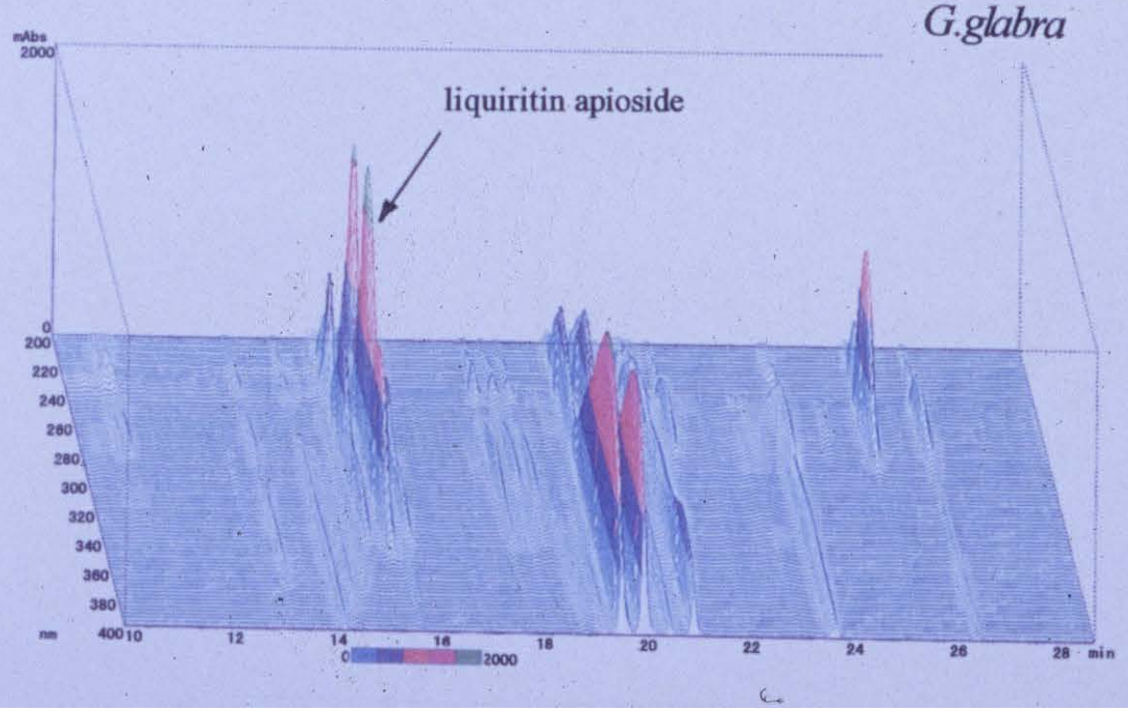
The antitussive effects of liquiritin apioside, liquiritin and liquiritigenin were assessed 1 and 4 h after p.o. administration of each drug.

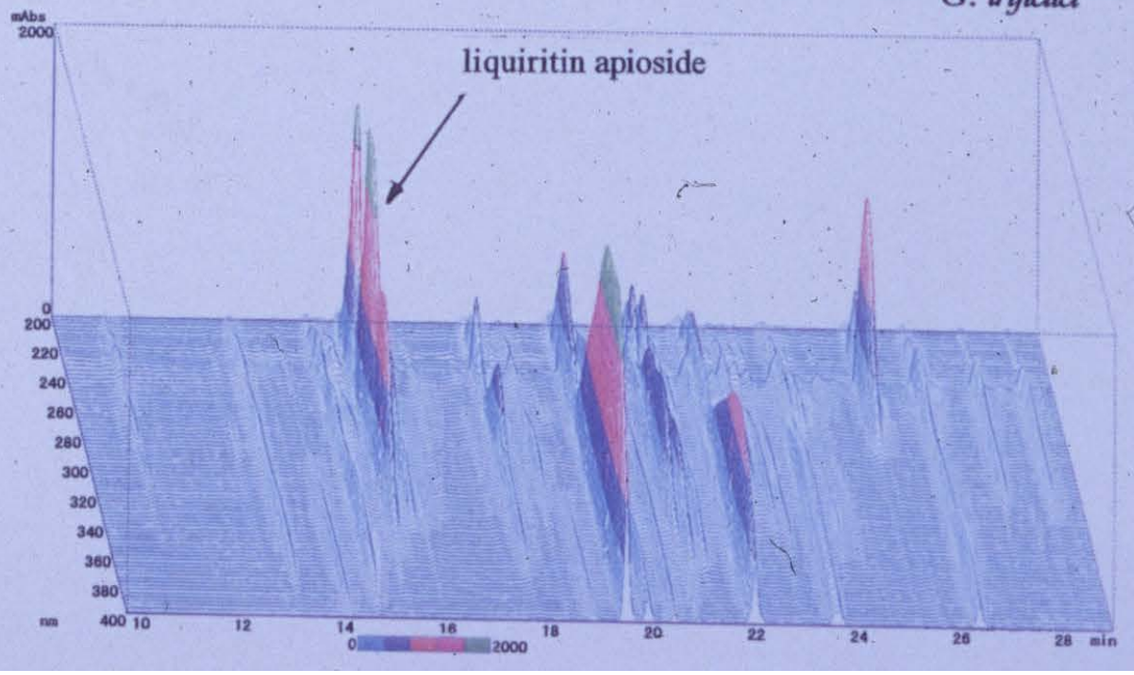
Glycyrrhizae radix (licorice) species used for Bakumondoto in the Japanese Kampo medicinal prescription that are often used as a antitussive medicine should be selected under consideration of the amount of liquiritin apioside, liquiritin, and liquiritigenin in their prescriptions.

Active constituents properties of Glycyrrhizae radix (licorice) for antitussive derived from different species

	Species	Main places of production
Dongbei Licorice (Tohoku-kanzo)	G. uralensis Fisch	Northeast of China, Hebei, Shanxi, Neimenggu
Xibei Licorice (Seihoku-kanzo)	G. Glabra Linne G. uralensis	Gansu, Quinghai, Neimenggu
Xinjiang Licorice (Sinkyo-kanzo)	G. inflta Batal, G. glabra Linne, G. uralensis Fisch, G. kashinski, etc.	Xinjiang, Gansu







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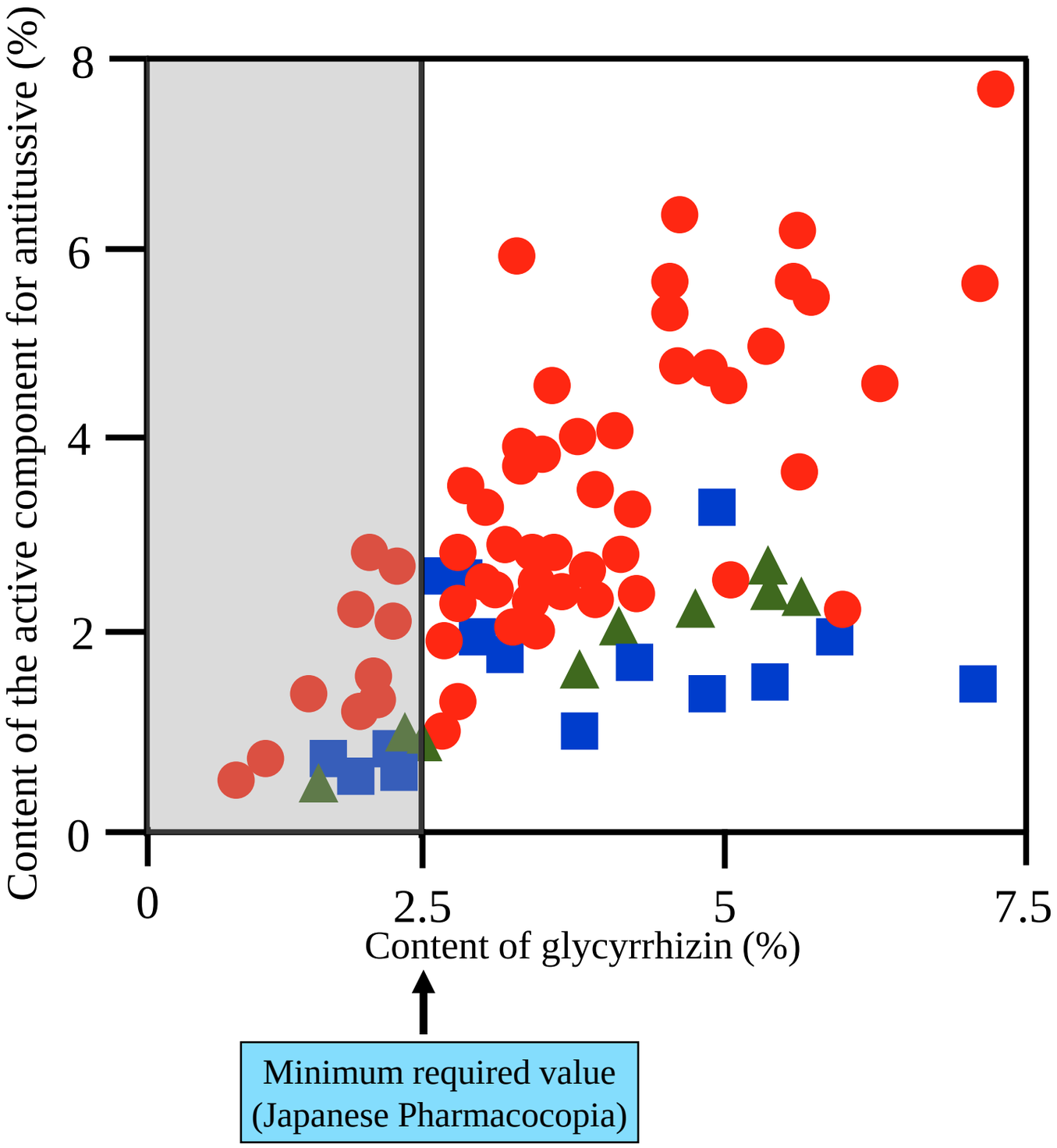
Active constituents properties for antitussive derived from the three species of Glycyrrhizae radix (licorice)

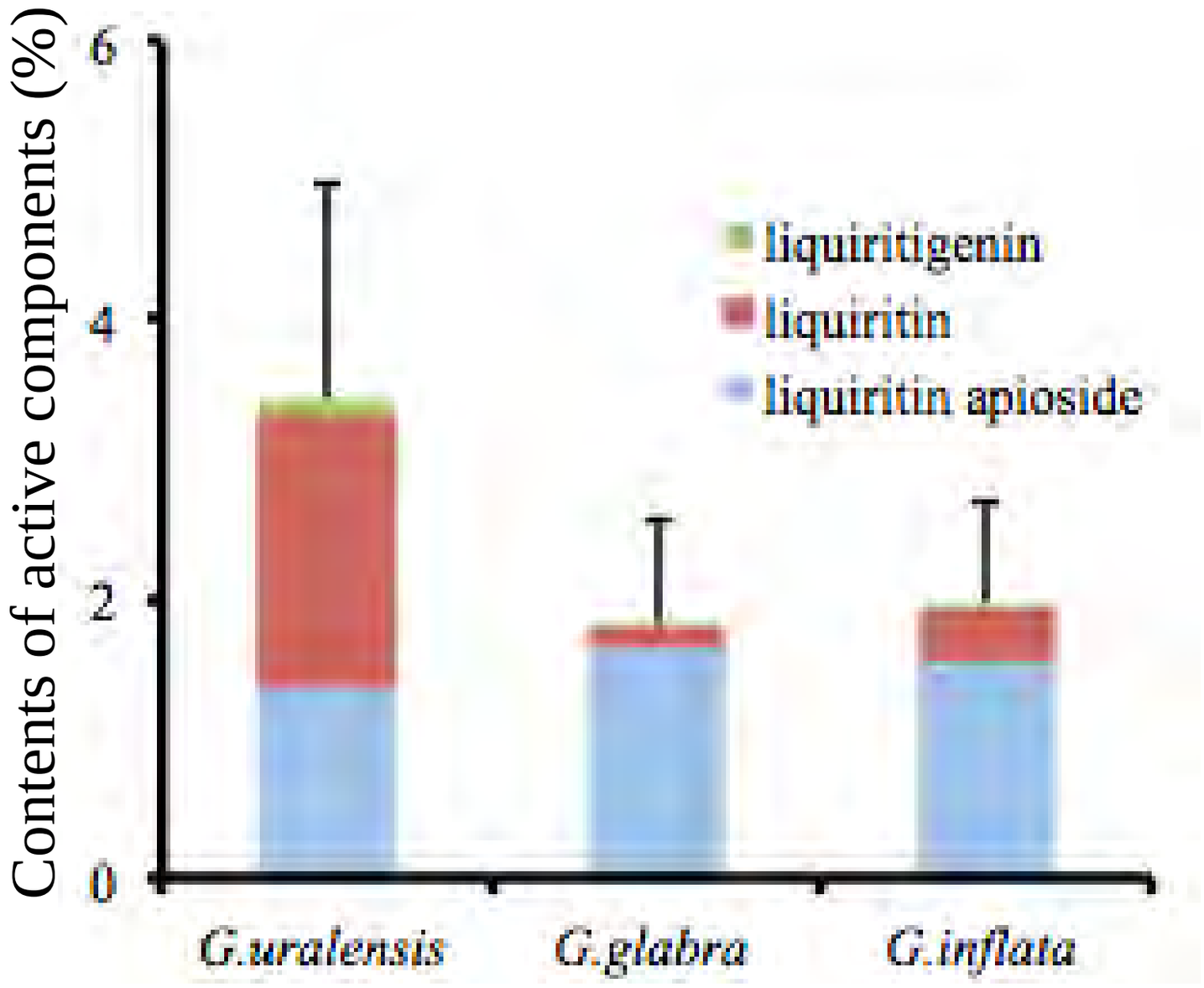


G. uralensis
G. glabra
G. inflata

In Japanese Pharmacopoeia 2011 (JP2011), two species of G. uralensis Fischer and G. glabra Linne are listed as licorice.

Furthemore, it is established the content standard of glycyrrhizin 2.5 % in JP2011

Active constituents properties for antitussive derived from the three species of Glycyrrhizae radix (licorice)



The mean contents of liquiritin, liquiritin apioside, and liquilitigenin in G. uralensis are significantly higher than those of G. glabra and G. inflata .

Conclusion

1. Glycyrrhizae radix (licorice*)*, one of the main components of a Bakumondoto, contains a potent antitussive compound: liquiritin apioside. The antitussive effect of liquiritin apioside depends on both peripheral and central mechanisms.

2. Glycyrrhizae radix (licorice) may produce a persistent antitussive effect and suggest that liquiritin apioside plays an important role in the earlier phase, while liquiritigenin, which is a metabolite of liquiritin apioside and liquiritin, plays an important role in the late phase.

3. When licorice is used for medicinal purposes, the licorice species should be selected with recognition of those constituent properties. Hence, licorice speciesused for Bakumondoto in the Japanese Kampo medicinal prescription that are often used as a antitussive medicine should be selected under consideration of the amount of liquiritin apioside, liquiritin and liquiritigenin in their prescriptions.

When the basic evidence for herbal medicine is accumulated, the specificity of the Kampo medicine will understood, and clinical evidence for Kampo medicine is expected to increase in near future.