EFFECT OF GINSENG EXTRACT ON OXYGEN CONSUMPTION IN RAT LIVER MITOCHONDRIA

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ABSTRACT

With microorganism as a single cell model to investigate the cellular effect of total extract of ginseng powder, it was found that ginseng affects cellular respiration biphasically (Tso and Fung, 1980; Tso, 1981). As ginseng is recognized to be a tonic medicinal herb, this finding suggests a possible role of ginseng in altering cellular energy metabolism. Along this line, the same effect on mitochondrial oxygen consumption was studied. It was found that under a controlled pH condition, a significant stimulation of the mitochondrial respiration was observed. This stimulation was ginseng dose-dependent. However, when ginseng was applied at an above threshold concentration, an inhibitory effect was noted. This confirms the previous observation with single cell organism and suggests a universal regulation of energy metabolism effect that transcends cell origin.

INTRODUCTION

Recent pharmacological studies with *Panax ginseng* root extract or the isolated ginsenosides have revealed the diversified properties of the saponins (Otsuka *et al,* 1977; Karzel, 1977). The extract enhances glucose consumption of the rat liver *in vitro* (Kang, 1977). Except ginsenoside Rb_1, all other isolated fractions have an increase of leucine incorporation and an increase of serum protein biosynthesis (Oura *et al,* 1975). A similar stimulatory effect on RNA precursor incorporation has also been reported (Oura *et al,* 1971). Subsequent physiological studies also indicate that ginseng extract potentiates the effect of nerve growing factor (Saito *et al,* 1977). We have recently also shown that Rd, one of ginsenosides, was specifically effective in reducing the uptake of neurotransmitters that can be significant in modulating the central nervous system function (Tsang *et al,* 1983). Moreover, the lipid soluble fraction of the crude extract has produced a significant depression in the aquisition of sound diuscrimination behaviour in rats (Saito *et al,* 1979). These and other reports taken together have indicated that the ginseng extract exhibits a complicated but significant effect on the physiology of living organisms. On the other hand, none of the findings so far reported has provided us with an unambiguous picture of the function of the extract in these diversified physiological systems.

Although the ginsenoside fractions produced a variety of physiological effects, it seems that in general it has a stimulatory function. With a simple prokaryotic system as a model, we have previously shown that ginseng has produced a strong stimulatory effect on the cellular respiratory system. This study reports an extension of our ginseng-on-energy metabolism project with a

subcellular fraction, the mitochondrial fraction.

MATERIALS AND METHODS

Korean Red ginseng powder was obtained from Korea Ginseng Centre Ltd., Hong Kong. The original crude extract was prepared by refluxing the powdered ginseng with glass distilled water (100mg/ml) for one hour. The filtrate was used.

Sprague-Dawley rats were used in this study. Mitochondrial fractions were prepared as described in Haldar *et al* (1979) for mouse liver. The liver was rinsed four times with 5 volumes of 0.25M sucrose containing 1 mM EDTA, pH 7.4 and the mitochondrial fractions were rinsed four times with 0.25M sucrose. Oxygen consumption at 37°C was measured directly using a biological oxygen monitor (Yellow Springs Instrument Co., Inc., Ohio, USA). For calibration, the air saturated glass distilled water was taken to be 5.0μl 02/ml water.

RESULTS

The presence of ginseng in the assay medium produced a notable stimulatory oxygen consumption as shown in Table 1 (Experiment 1). This stimulation was not observed with sugar moiety. At 0.1mg/ml ginseng concentration, hardly any stimulation was observed. The stimulation was approximately proportional to ginseng concentration up to 1mg/ml (data not shown). At a higher ginseng concentration, however, an inhibition was exhibited. Even though there was slight variations from batch to batch mitochondrial preparations, the optimal respiratory stimulating ginseng concentration seems to be at approximately 1mg/ml to 3mg/ml.

With either N,N'-dicyclohexylcarbodiimide (DCCD) or oligomycin as an ATPase inhibitor, the respiratory rate of the mitochondrial preparation was much reduced due to an accumulation of the phosphorylated products. The reduction was reversed by the addition of ginseng. This behaviour was again a biphasical one: effective at about 1mg/ml ginseng concentration and ineffective at concentrations higher than ten-fold, presumably due to inhibition.

DISCUSSION

Like prokaryotes, rat liver mitochondria respond to low concentrations of ginseng with a stimulation of respiration that cannot be ac-

Table 1. Effect of ginseng on oxygen consumption of washed rat liver mitochondria at 25°C.

Expt.	Addition Sequence	Ginseng Concentration (mg/ml)	Oxygen Consumption (μmoles 02/hr/mg)	% of Control
1	Control		0.355	100
	Succinate		3.86	1087
	Ginseng#	2.20	5.11	1439
2	Control	1.20	0.351	100
	Succinate	4.70	1.10	314
	Ginseng	10.6	2.00	569
			1.39	396
			0.74	211

Rat liver mitochondria (0.22mg/ml) were incubated in a medium containing: 160mM sucrose, 3.8μM rotenone, 0.46mM ADP, 20mM KCl, 5mM MgCl2, 10mM Tris-Cl pH7.4. The final concentration of Na succinate was 4.70mM and 2.46mM in Experiment 1 and 2 respectively. The data was a typical record of three experiments. # A separate experiment with the addition of glucose instead of ginseng gave essentially equal oxygen consumption rate (3.82).

Table 2. Effect of ginseng on oxygen consumption of oligomycin suppressed rat liver mitochondria.

Addition Sequence	Ginseng Concentration (mg/ml)	Oxygen Consumption (umoles 02/hr/mg)	%of Control
Control		0.092	100
Succinate		0.63	685
Oligomycin#		0.136	148
Ginseng	0.85	0.92	1000

Experimental conditions were similar to that listed in Table 1. The concentrations of mitochondrial preparation, Na succinate and oligomycin added were 0.51mg/ml, and 4.65mM and 0.17mg/ml correspondingly. #Equal results were noted with DCCD instead of oligomycin.

counted for by the sugar moieties present in the molecule. The profile of the stimulation, however, resembles the action of a conventional uncoupler of oxidation phosphorylation and respiration that acts biphasically, with a stimulation at low concentrations and an inhibition at relatively higher concentrations. This is confirmed by the action of ginseng in accelerating the oxygen consumption rate which has been slowed down by an ATPase inhibitor, oligomycin. Apparently, ginsenosides are detergent-like molecules that associate readily with the membrane of the subcellular organelle to render the latter leaky. Crude ginseng extract contains a great variety of gensinosides. Obviously, there is a need to understand and identify which moiety in the ginseng extract is responsible for the uncoupling action.

Since the concentration of ginseng inside the subcellular fractions of either the mammalian species or the specific organ is lacking, any suggestion to the effect of ginseng on normal mitochondrial function will only be speculative. As the orignial 100mg/ml extract is an unusual concentrated preparation and that the cellular gensenoside uptake, either the kinetic or the structural specificity, is unknown, it is most likely that under ordinary conditions, the uppermost concentration of ginseng in the cell should be less than 1mg/ml. At this concentration range, only a slight uncoupling action can be expected. At a first glance, it seems that an uncoupling effect in the cellular power plant indicates a lack of efficiency in the cellular energy management. There is no clue at present on now a slight uncoupling effect may interfere with the well-being of the cell or the individual as a whole. Historically, a conventional uncoupler such as dinitrophenol has been shown to exhibit a remarkable stimulating effect on fat metabolism which has been tried extensively in the past for clinical reduction of obesity (Jenkins and Hartung, 1949). In this aspect, this biochemical acitivity of ginseng may then be of great value. Apparently, this ginseng regulated energy metabolism effect needs further exploration.

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인삼 추출물이 세포의 산소소모에 미치는 영향

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인삼이 강장제인 약용식물로 인식됨에 따라 세포에 대한 인삼 추출물의 효과를 연구하기 위해 미생물을 실험재료로 연구한 결과, 인삼이 두 단계로 세포 호흡에 영향을 미치는 것으로 관찰되었으며 (Tso

and Fung, Microbios, Lett. 13 : 7~12., Tso, Acta Microbiologica sinica 21 : 53~56)이것은 세포 에너지 대사 변화에 중요한 역할을 할 수 있다는 것을 시사해 주었다.

이와 같은 결과로 인삼이 세포의 mitochondria 에 의한 산소 소모에 미치는 효과를 검토한 결과, 일정한 pH 조건하에서 인삼은 호흡을 촉진시켰으며, 일정 농도 이상 투여하였을 경우 다소의 억제효과를 보였다.

위의 결과들은 세포의 mitochonbria 호흡에 미치는 인삼의 효과를 재확인하는 것으로 강장제로서의 인삼의 가치를 뒷받침해 주고 있다.

K.S. Lee: Do you base the uncoupling effect of ginseng only on one data showing that dinitrophenol has no effect after ginseng. If that is the only base you have, I think it is very weak. If not, what other data do you have?

Tso: Actually, I'm showing a typical one but all of these experiments have been repeated a couple of times.

That's not the only base but I'll describe it to you later. Because in the white blood cells, the motility is different and Chemotaxtic behavior is also different.

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