

# 人红细胞钠泵活性及大黄浸液对其影响

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**内容提要** 用 $^{86}\text{Rb}^+$ 摄入法测定了70例健康人红细胞钠泵活性和大黄浸液对其抑制作用。结果：70例健康人红细胞钠泵活性为 $0.449 \pm 0.007 \text{ mmol/L RBC} \cdot \text{h}$ 。大黄浸液浓度 $10^{-4} \text{ mg/ml}$ 时，表现出抑制作用，抑制率为1.1%；浓度 $10^{-2} \text{ mg/ml}$ 时，抑制率达100%。还对大黄抑制钠泵与其药性作用关系进行了探讨。

近年来临床上应用以大黄为主的方剂及单味大黄治疗急、重症感染等疾病取得较满意疗效，临床上已证实大黄具有明显的清热降温作用<sup>①</sup>。由于细胞产热增加与钠泵活性紧密相关<sup>②</sup>，为此，我们采用 $^{86}\text{Rb}^+$ 摄入法测定健康人完整红细胞钠泵活性，并观察大黄对其影响。

## 资料和方法

### 一、资料

1. 大黄浸液为 $1.0 \text{ g/ml}$ <sup>③</sup>。实验前用等渗反应液(含 $150 \text{ mM NaCl}$ 、 $10 \text{ mM}$  葡萄糖、 $10 \text{ mM Tris-HCl}$ 、 $1 \text{ mM Rb}_2\text{CO}_3$ ， $\text{pH} 7.4$ )按需要稀释。

2. 完整人红细胞制备：取健康献血员(70例)静脉血 $4 \text{ ml}$ ，置于含有肝素 $0.1 \text{ ml}$ 的试管中，迅速摇匀，冷却， $2500 \text{ rpm}$ 离心5分钟，吸除血浆及红细胞的绒毛状表层。加入4倍体积的 $4^\circ\text{C}$ 生理盐水，在快速混匀器上混匀， $3000 \text{ rpm}$ 离心5分钟。同样操作重复两次。洗后的压积红细胞，用等渗液(不含 $1 \text{ mM Rb}_2\text{CO}_3$ 的等渗反应液)配成1:10红细胞悬液。

二、方法：人红细胞Na-K泵活性测定方法，采用 $^{86}\text{Rb}^+$ 摄入法<sup>④,⑤</sup>。取1:10红细胞悬液 $1 \text{ ml}$ 于试管内， $3000 \text{ rpm}$ 离心5分钟。吸出上清，沉淀为 $100 \mu\text{l}$ 压积红细胞。管内加入 $200 \mu\text{l}$ 等渗反应液。与此同时，作一哇巴因等渗反应液管(内含 $100 \mu\text{l}$ 压积红细胞及 $200 \mu\text{l}$ 反应液)为对照管。于各管中加入 $10 \mu\text{l}$   $^{86}\text{Rb}_2\text{CO}_3$ 液(大约

$10\,000 \text{ cpm}$ )，充分摇匀后放 $37^\circ\text{C}$ 水浴保温60分钟(每隔15分钟摇动一次)。保温结束后放冰箱冷却5分钟。 $4000 \text{ rpm}$ 离心5分钟以终止反应。在 $\gamma$ -能谱仪上计数放射性。吸出上清，以 $4^\circ\text{C}$ 等渗液 $2 \text{ ml}$ 洗两次，测沉淀中放射性cpm。以上操作均在冰浴中进行。依实验中各种用量，计算红细胞钠泵活性。红细胞钠泵对 $\text{Rb}^+$ 的转运活性以 $\text{mmol/L RBC} \cdot \text{h}$ 表示。

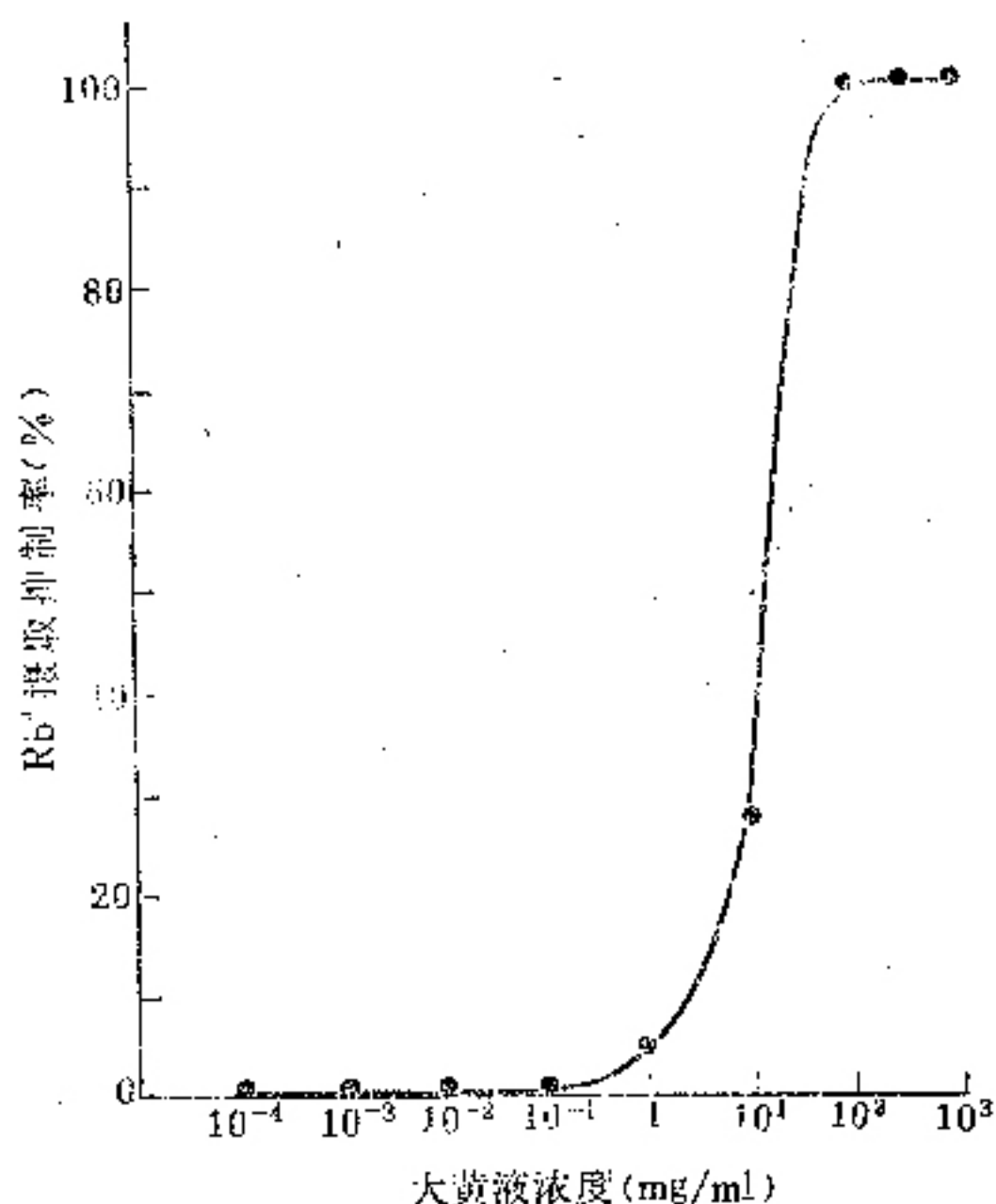
大黄浸液以等渗反应液按10稀释，使成为每毫升含1000、100、10、1、 $10^{-1}$ 、 $10^{-2}$ 、 $10^{-3}$ 、 $10^{-4} \text{ mg}$ 的不同浓度。实验用上述各浓度的大黄浸液 $200 \mu\text{l}$ 、压积红细胞 $100 \mu\text{l}$ 及 $^{86}\text{Rb}_2\text{CO}_3$   $10 \mu\text{l}$ ， $37^\circ\text{C}$ 保温60分钟，同上述操作，计算红细胞在不同浓度大黄浸液作用下的钠泵活性，由 $^{86}\text{Rb}^+$ 摄入量换算出大黄浸液对其的抑制率。

经预实验选择出最佳实验条件：保温时间60分钟。哇巴因浓度 $1 \text{ mM}$ ，红细胞体积 $0.1 \text{ ml}$ ， $^{86}\text{Rb}^+$   $10 \sim 40 \mu\text{l}$ ， $^{86}\text{Rb}^+$ 浓度 $1 \text{ mM}$ 。

## 结 果

一、70例健康人红细胞钠泵活性为 $0.449 \pm 0.007 \text{ mmol/L RBC} \cdot \text{h}$ ( $\text{M} \pm \text{SE}$ )。

二、大黄浸液对健康人红细胞钠泵活性影响：见附图。大黄浸液浓度 $10^{-4} \sim 10^{-2} \text{ mg/ml}$ 时，对红细胞钠泵活性无抑制作用； $10^{-1} \text{ mg/ml}$ 时，表现出抑制作用，抑制率为1.1%；浓度为1、 $10^1$ 、 $10^2$ 、 $10^3 \text{ mg/ml}$ 时，抑制率分别为4.1%、28.4%、100%、100%。



附图 大黄浸液对健康人红细胞钠泵活性的影响

## 讨 论

细胞内高钾低钠是依赖于细胞膜钠泵不断地维持浓度梯度差,这是一耗能的主动运输离子的过程。每分解一分子ATP,伴随有3个 $\text{Na}^+$ 向外及2个 $\text{K}^+$ 向内转运。凡具有 $\text{Na}^+$ 、 $\text{K}^+$ 运输系统的生物组织均可测出 $\text{Na}^+$ 、 $\text{K}^+$ -ATP酶活性。人类红细胞膜 $\text{Na}^+$ 、 $\text{K}^+$ -ATP酶与完整红细胞阳离子主动转运系统存在许多共同特点。细胞膜钠泵是细胞主动转运 $\text{Na}^+$ 、 $\text{K}^+$ 的主要途径,细胞一系列生理生化活动均与钠泵活动相关。某些疾病可能是由于钠泵异常<sup>(6~9)</sup>。研究钠泵是了解细胞膜功能完整性的重要内容,也是和疾病发生、发展和转归以及药物治疗密切相关的。

钠泵的研究方法有多种。在钠泵的运转中,细胞外 $\text{K}^+$ 结合位点可被多种一价阳离子所替代,其中 $\text{Rb}^+$ 对钠泵亲和力仅次于 $\text{K}^+$ ,且同位素标记的 $^{86}\text{Rb}^+$ 半衰期远较同位素标记的 $^{42}\text{K}^+$ 为长,因而细胞 $^{86}\text{Rb}^+$ 摄取率测定亦是钠泵功能研究的常用方法。此法灵敏度高、简便易行。

大黄在临床应用历史悠久。近代药理实验

证明具有多方面的作用,如泻下、利胆、解热、抗感染、清除内毒素等,正日益引起国内外学者的重视。动物实验和大量临床观察都证明了大黄可使感染性发热降温。大黄降温机理可能是通过多种因素起作用,例如大黄有抗菌作用,其机制主要是抑制糖及糖代谢中间产物的氧化和脱氢,抑制氨基氮的同化和氨基酸的氧化、脱氢和脱氨,从而抑制细菌体蛋白质和核酸的合成,抑菌生长,达到降温<sup>(2)</sup>。经实验证明了大黄是一个线粒体电子传递系统的抑制剂,能够显著地抑制还原型辅酶I氧化酶和还原型辅酶I-细胞色素C还原酶两个酶系统的活性,线粒体氧化磷酸化效率明显下降,能量代谢障碍,体温下降。我们的实验证明大黄对钠泵具有抑制作用,抑制人红细胞钠钾离子的主动转运。因此,大黄降温机理可能与大黄抑制 $\text{Na}^+$ 、 $\text{K}^+$ -ATP酶活性,从而使ATP分解减少有关。此研究工作尚待深入进行。

## 参 考 文 献

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plasma cAMP decreased, but cGMP in plasma and tissue significantly increased. There were significant difference in three types. After treatment, these indices in all three types normalized. The present study suggested that Zuojin pills could inhibit excretion of gastric acid, reduce plasma cAMP and increase plasma cGMP; Huangqi Jianzhong decoction could increase plasma cGMP level, and serum and tissue gastrin level; Shashen Maidong decoction could decrease cGMP levels in plasma and tissue as well as gastrin levels in serum and tissue, the cAMP/cGMP ratio in plasma and tissue was similar to that in control. It is believed that traditional Chinese medicine therapy is an essential principle regulating balance of physiological function in human body.

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### **Preliminary Study on Relationship between Yin(阴)Deficiency, Yang(阳)Deficiency in Chronic Pharyngitis and Function of Vegetative Nervous System**

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This paper reported the relationship between the Yin deficiency, Yang deficiency and the functional status of vegetative nervous system, based on 50 chronic pharyngitis patients and 30 normal subjects as control. The result showed that, compared with control, the Yin deficiency group belonged predominately to hyperfunction of the sympathetic nervous system, and the Yang deficiency group to hyperfunction of the parasympathetic nervous system ( $P < 0.005$ ). The observation indicated that the functional imbalance of the vegetative nervous system perhaps was one of the causes to influence the throat disease with imbalance of holistic Yin and Yang. The imbalance of internal organs of the body might cause the functional disorder of the cortical and subcortical centers through the vegetative nervous system, and also the pharyngeal pathologic changes might be aggravated. The chronic inflammation of pharynx might cause the imbalance of the cortical and subcortical centers through the vegetative nervous system too, and thus induced the symptoms of Yin or Yang deficiency to appear. It revealed that evaluating the functional status of vegetative nervous system might contribute to the treatment according to syndrome differentiation of chronic pharyngitis and other throat diseases.

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### **Studies of Erythrocyte Sodium Pump Activity in Human and Effect of *Rheum palmatum* on Its Activity**

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The cation-transport activity of the human erythrocyte sodium pump of 70 healthy persons was studied by  $^{86}\text{rubidium}$  uptake method to measure the experimental optimum conditions which was  $0.449 \pm 0.007$  mmol/ RBC/h. The inhibitory activity of *Rheum palmatum* on the human erythrocyte sodium pump in vitro was also observed. Our studies suggested that the abnormal sodium pump activity and cellular energy metabolism was possibly mutually influenced pathophysiologically.

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### **Effect of Moxa-Cone Moxibustion on Temperature and Microcirculation of Febrile Rabbits Caused by Colitoxin**

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In order to explore the role of moxibustion as antipyretics, this paper studied the effect of moxibustion on temperature and microcirculation in rabbit fever model caused by colitoxin which simulated fever model caused by exogenous pathogenic factors. The febrile rabbits were divided into two groups: one was moxibustion group(MG) which was moxibusted at Dazhui(GV14) point immediately after the fever was induced, the other was control group (CG) which was not moxibusted, but moxa-cone was placed on the Dazhui. The pyretic effect of these two groups was comparatively observed between MG and CG. Results showed that the fever incubation period in MG was shortened,