

参芪注射液对血小板表面活性和超微结构的影响

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内容提要 本文应用电镜观察了一次静脉注射参芪液 60ml 前后对冠心病心气虚患者血小板聚集的影响。结果提示用药前冠心病心气虚患者与正常相比血小板扩聚型和聚集数都明显增加, 用药后一小时上述异常现象明显好转。用家兔血小板体外实验表明参芪液对 ADP 诱导的血小板聚集也有抑制作用。血小板超微结构的初步观察提示对血小板的伪足形成及颗粒释放也可能有一定抑制作用。

北京中医学院东直门医院气血研究室用 Born 比浊法研究表明⁽¹⁾, 补气中药党参、黄芪对二磷酸腺苷 (ADP) 所诱导的血小板聚集有一定的抑制作用。为了进一步观察党参、黄芪对血小板聚集的影响, 我们采用电镜对血小板的表面形态和超微结构进行了研究。

材料和方法

一、试剂: 1. 参芪注射液: 将党参 (*Radix Codonopsis Pilosulae*, 产地山西) 和黄芪 (*Radix Astragali Seu Hedysaxi* 产地内蒙) 由我院制剂室制备成可供静脉用的药液, 批号 820520, 每毫升药液含相当生药党参、黄芪各 0.5g (制剂不含吐温)。

2. ADP: 北京中医学院生化教研室提供, 使用时以生理盐水稀释, 终浓度每毫升 $5\mu\text{g}$ 。

二、富血小板血浆制备: 硅化注射器采血, 以 3.8% 枸橼酸钠抗凝 (V:V=9:1)。血盛于硅化刻度离心管中, 1000 转/分离心 5 分钟, 取上清即为富血小板血浆 (PRP)。

三、血小板表面形态观察及标本制备: 根据 Schatz 法⁽²⁾并做了重要修改。预先制备、漂取 0.2% Formvar 膜, 膜上均匀放置电镜小铜网 4~5 个, 用干净玻片捞取之, 自然干燥。将上述 PRP 迅速涂于此玻片上, 37°C 温育 10 分钟, 室温下以改良台氏液轻轻冲掉血浆, 稍干即将该标本浸入 1% 四氧化锇固定液中。10 分钟后取出标本, 用双蒸馏水冲洗干净, 自然干燥。用 H-500 电镜观察, 顺序计数 100 个血小板。参照有关资料, 将血小板形态分为圆球型, 扩聚型, 进行分类计数, 并同时计数扩大型血小板所聚集的血小板个数

四、血小板超微结构观察及标本制备: 参照中国医学科学院基础所细胞生物室报道方法⁽³⁾。

观察对象及分组

一、冠心病心气虚患者: 根据虚证辨证标准, 心气虚患者有心悸、气短、神疲乏力、自汗, 脉细弱或结代, 舌淡胖或边有齿痕, 苔薄白等见证。全部病例符合 WHO 所规定的缺血性心脏病诊断标准。

所观察病例停药 5 天以上, 实验当天禁高脂饮食。晨 8 时用硅化注射器从肘静脉采血, 此为药前对照。然后, 缓慢静注参芪液 60ml。注射完后一小时再次取血。采血后制备电镜标本并观察之。本组共观察冠心病心气虚患者 9 例, 其中男 3 例, 女 6 例, 平均年龄 60 ± 9.5 岁。

二、正常人: 由本院健康医护人员和体检选取健康成人。本组共 10 例, 其中男 7 例, 女 3 例, 平均年龄 47.5 ± 15.4 岁。取血及标本制备同上。

三、动物: 健康雄性家兔, 体重 3 kg 左右。将制备的 PRP 各取 0.40ml, 一组不加任何药物, 加缓冲液对照, 一组加入 ADP 诱导聚集, 一组先加入参芪液再用 ADP 诱导聚集。加 ADP 前, 各组在 37°C 水浴中温育 10 分钟, 然后在搅拌情况下加入 ADP。2 分钟内迅速将此血浆制备成电镜标本进行观察。ADP 终浓度 $5\mu\text{g}/\text{ml}$, 参芪液 $40\text{mg}/\text{ml}$ 。

结 果

一、药物对人血小板表面形态影响, 见表 1。

冠心病心气虚患者在用参芪液之前和正常人相比血小板扩聚型和聚集数都增加。一次静注参芪液后, 血小板扩聚型和聚集数都明显下降, 用药前后差异有显著性, 且 9 例冠心病心气虚患者, 用参芪液后每例

表1 参芪对冠心病心气虚患者血小板表面形

态影响(M±SE)

组 别	圆、树型%	扩聚型%	聚集数(个)
正常人(10例)	87.0±2.5	12.9±2.5	34.6±5.8
心气虚、药前(9例)	72.7±4.3	27.3±4.3	66.9±9.6
心气虚、药后(9例)	89.2±2.9	10.8±2.9	24.6±6.1

扩聚型, 正常人与心气虚药前比 $P<0.05$ 心气虚药前、后比 $P<0.01$ 聚集数, 正常人与心气虚药前比 $P<0.01$ 心气虚药前、后比 $P<0.01$

血小板扩聚型和聚集数皆下降。说明参芪液能降低患者血小板表面活性, 对聚集的血小板有一定解聚作用。

二、参芪液对家兔血小板表面形态影响, 见表2。ADP诱导组血小板扩聚型和聚集个数比正常家兔对照组明显增高; 而用参芪液后再加ADP诱导聚集与ADP组相比, 扩聚型和聚集个数均明显下降。以市售丹参注射液为对照组, 表明丹参的作用略优于参芪组。参芪加丹参的作用与单用丹参近似。

表2 参芪、丹参对家兔血小板表面形态影

响(M±SE)

组 别	圆、树型(%)	扩聚型(%)	聚集数(个)
正常家兔	93.6±0.8	6.4±0.8	13.2±2.1
ADP组	78.0±2.9	22.0±2.9	55.8±9.1
ADP+参芪组	91.0±1.1	9.0±2.1	20.2±2.9
ADP+丹参组	94.4±0.7	5.6±0.7	12.8±1.7
ADP+参芪+丹参组	94.0±1.6	5.8±1.6	12.4±2.9

各组动物数=5 扩聚型组间 $F=18.1, P<0.01$;聚集数组间 $F=14.1, P<0.01$

三、血小板超微结构观察所见: 电镜观察5份动物标本表明, ADP诱导组家兔血小板伪足明显增多变长, 细胞内颗粒释放, 尤其致密颗粒的释放更加明显。用参芪后再用ADP诱导, 则血小板伪足皆有不同程度减少, 细胞内颗粒大部分存在。经统计学处理, 前者每个血小板超薄切面平均含 α 颗粒 2.7 ± 0.6 个, 致密颗粒 0.7 ± 0.2 个, 伪足 1.6 ± 0.6 条; 后者则每个血小板超薄切面平均含 α 颗粒 5.3 ± 0.6 个, 致密颗粒 1.1 ± 0.2 个, 伪足 0.1 ± 0.1 条; 两组相比有显著性差别($P<0.05$)。

电镜观察5份人的标本表明, 健康人和心气虚者以及心气虚用参芪液前后, 血小板超微结构变化不明显。

讨 论

1975年 Schatz 用电镜对缺血性心脏病患者的血小板进行了研究。在14例正常人中, 扩大型血小板占 $13.4\pm9.1\%$ ($M\pm SD$), 而14例缺血性心脏病患者, 扩大型占 $69.0\pm22.2\%$ 。而用此方法对补气中药抗血小板聚集的研究则未见报道。我们用改良 Schatz 法观察表明, 冠心病心气虚患者扩聚型和聚集数比正常人明显增加, 为气虚而致血瘀提供了初步的实验依据。

冠心病心气虚患者用参芪液后血小板扩聚型和聚集个数和用药前相比均明显下降。动物实验也表明, 用ADP诱导组和加参芪后再用ADP诱导组相比扩聚型和聚集数差别明显。这都说明党参黄芪能降低血小板的表面活性, 抑制血小板聚集。这和我们以往用血小板聚集仪观察参芪对血小板聚集具有抑制作用的结果是一致的。两种方法, 实验手段不同, 但结果一致, 更证实参芪确有抑制血小板聚集的作用。从血小板超微结构观察提示, 补气中药党参黄芪还能在一定程度上抑制血小板聚集时的颗粒释放和伪足形成, 提示对血小板的收缩也有一定的抑制作用。对预防和治疗血栓形成等病理改变是有意义的, 很值得进一步研究。

据时其煌等报道, 活血化瘀的冠心 II 号可以抑制胶原诱导的血小板颗粒的释放, 但不能抑制伪足的形成^[3]。鉴于冠心病等心脏病的中医基本病机是气虚血瘀, 因而益气活血是一条重要的治疗途径。中医理论认为气可行血又可摄血。气虚血瘀则益气可以行血化瘀; 气虚血脱则益气可以摄血固脱。为此, 应用现代科学方法逐步阐明益气与行血、摄血的关系及其机理, 具有重要的理论和实践意义。本文对补气中药抗血小板功能所进行的初步研究表明补气药党参黄芪对血小板聚集以及释放、收缩功能具有一定的抑制作用, 从一个侧面反映了气血相关理论的实质, 为益气活血治则提供了一定的现代科学实验依据。

参 考 文 献

1. 廖家桢, 等. 参芪等益气中药治疗冠心病作用机理的研究. 中华医学会第二届全国心血管病学术会议论文摘要, 1983:29.
2. Irwin J Schatz. Blood platelet response to plasma from patients with ischemic heart disease. AM J Cardio 1975; 35(1):204.
3. 时其煌, 等. 冠心 II 号对家兔血小板释放 5-HT 含量和超微结构的影响. 中西医结合杂志, 1981; 1(2): 90.

cases were treated with hormone. In the former group, marked effect was observed in 32 cases (78%), improvement in 9 cases (22%). Twenty-four cases were followed up, among which 20 cases (83%) were found cured. The results obtained were much better than those obtained in the group treated with hormonal therapy.

In those patients treated with traditional Chinese medicine. We investigated cyclic changes of the ovary by measuring basal temperature in 11 cases, examining cornification of vaginal epithelium in 12 cases, examining cervical mucus crystals in 12 cases, and assaying blood progesterone level in 7 cases, all the findings indicating that the tonifying kidney method promoted ovarian function.

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The Role of Purgation Therapy in the Emergency Treatment of Acute and Severe Cases of Biliary Tract Troubles

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About 4 years' clinical practice of ours has proved the previous belief that once the biliary system is infected and gall stones are formed, and acute suppurative obstructive cholangitis, septicemia and infectious shock follow in the wake, surgical operation would be indispensable is no longer true. Under certain circumstances, it can be successfully treated non-operatively.

In our series there are 123 severe cases with biliary infection and gall stone formation. 70 cases were treated with combined traditional Chinese and western medicine chiefly with purgation therapy and 53 cases by operation. For reasons given above, the mortality in our series has dropped from 59% (in 60's) to 10.6% (by now).

The authors believe that the mechanism of purgation therapy is to ensure frequent loose stools, as a result of which clinical symptoms and signs are relieved or disappear. In combination with purgation therapy "Stone-Expelling Decoction" is given and proves very effective. Treatment with combined traditional Chinese and western medicine does not exclude surgical intervention. One has to choose the right therapy for individual cases. While employing purgation therapy, the doctors and medical workers must closely monitor the changes of the condition.

When operation is needed, the patient must be referred to surgery in due course of time, operative therapy is an important and effective measure to remove obstruction, drain purulent bile, and alleviate infection.

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Effects of Codonopsis Pilosulae-Astragalus Injection on Superficial Activity and Ultrastructure of Platelets

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Codonopsis Pilosulae and Astragalus (CP-A) are often used to treat coronary heart disease (CHD) in TCM. In order to investigate the effects of CP-A on platelet, the present study reports the effects of CP-A on the superficial activity and ultrastructure of platelets by means of electromicroscopy. The study was performed in patients with CHD in vivo (N=9), in which the superficial activity of platelets were lowered following a single dose of 60 ml CP-A injection (content of CP-A 30gm each) intravenously. The spread type platelets and number of aggregated platelets were reduced from $27.3 \pm 4.3\%$ to $10.8 \pm 2.9\%$ ($M \pm SE$, $P < 0.01$) 66.9 ± 9.6 to 24.6 ± 6.1 ($M \pm SE$, $P < 0.01$) respectively. The results of study performed in rabbits in vitro showed that the ADP-induced platelet aggregation was also inhibited by CP-A, the spread type platelets and the number of aggregated platelets were reduced from $22.0 \pm 2.9\%$ to $9.0 \pm 2.1\%$ ($P < 0.01$), 55.8 ± 9.1 to 20.2 ± 2.9 ($P < 0.01$) respectively. The observation of changes in the ultrastructure of aggregated platelets induced by ADP found that the formation of pseudopodias and the release of α -granules and dense-granules from these platelets were inhibited by CP-A to certain extent. It is well known that adhesion, release and aggregation of platelets play an important pathogenetic role in CHD, so that the effects of CP-A on platelet is of beneficial therapeutic value for treating CHD.

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