

血瘀证患者红细胞膜流动性的观察

附属中山医院 胡庆福 陈泽霖 陈依萍
上海医科大学 药 学 院 郑瑶函 崔志平 庄庆祺
仪器分析中心实验室 戎大海 朱志强
附属华山医院 陈健民
上海香山医院 刘春堂 屠 森

内容提要 采用荧光偏振度测量方法,对健康人、冠心病及恶性肿瘤的血瘀证患者红细胞膜流动性作了检测。结果:冠心病和恶性肿瘤血瘀证患者的红细胞膜荧光偏振度数据和平均微粘度数据,均高于健康人(P 均 <0.05)。表明冠心病和恶性肿瘤患者红细胞膜的流动性降低,使红细胞的变形能力减弱,从而影响血液在血管网中的流动性,促使微循环障碍的发生,这与中医的血瘀理论相符。

活血化瘀研究是中医治则研究中最为活跃的一个领域。近年来从生化、生物物理、病理生理、血流动力学、血液流变学、微循环等方面对活血化瘀的原理作了研究。为了进一步阐明血瘀证的本质,本文探讨了红细胞膜流动性与血瘀证的关系。

资料与方法

一、观察对象:选择冠心病患者21例,诊断均符合中西医结合防治冠心病、心绞痛、心律失常研究座谈会修订冠心病诊断标准。

选择恶性肿瘤患者14例,其中乳腺癌7例,胃癌2例,结肠癌4例,口腔恶性肿瘤1例,诊断均经病理检查证实,以上患者中医辨证均属血瘀证^①。

健康人14名,为志愿献血者和本院工作人员,均符合以下条件:近期内X线、心电图检查无异常发现,血尿常规、肝肾功能测定均正常,各系统无器质性疾病史;近1个月内无急性病史;舌质淡红,苔薄白。

二、方法

1. 红细胞悬液的制备:取静脉血2ml,肝素抗凝,800r离心5min,用磷酸缓冲液(PBS)洗涤3次,以高倍显微镜下找不到血小板为度。配成 10^{10} 个红细胞/ml悬液备用。

2. 荧光偏振度测量及微粘度计算:实验中

用荧光探针标记红细胞膜脂区,通过测量荧光偏振度,求得平均微粘度。采用文献报道方法^(2~7)。

结 果

血瘀证患者与健康人的红细胞膜荧光偏振度和微粘度对比:见附表。

附表 患者与健康人、红细胞膜的荧光偏振度和微粘度 ($M \pm SE$)

组 别	例数	荧光偏振度	微粘度(π)
健康人	14	0.294 ± 0.007	3.656 ± 0.237
冠心病	21	$0.328 \pm 0.013^* \Delta$	$6.787 \pm 1.028^* \Delta$
恶性肿瘤	14	$0.333 \pm 0.011^*$	$6.698 \pm 1.357^*$

*与健康人组相比, $P < 0.05$; Δ 患者组间对比, $P > 0.05$

冠心病、恶性肿瘤血瘀证患者的红细胞膜荧光偏振度和微粘度均明显升高,与健康人相比差异均显著(P 均 <0.05),但患者组间比较,差异无显著性意义($P > 0.05$)。

讨 论

细胞膜的流动性是细胞膜的一个重要特征,细胞膜的流动性与细胞的多种功能直接相关。红细胞膜的流动性对细胞的通透性、变形性、脆性、物质运输、酶的活性都有影响,因此研究细胞膜流动性是了解膜结构与功能关系的

重要环节。目前有多种方法从不同角度对细胞膜流动性进行研究,其中荧光偏振方法比较简便⁽⁴⁾。1,6-二苯基-1,3,5-己三烯是标记完整红细胞的有效探针,可在生理条件下直接标记完整红细胞作荧光偏振测量,所得参数——荧光偏振度能定量说明膜脂分子的运动性及其微粘度,因而得到比较广泛的应用。

一、红细胞膜流动性变化与血瘀的关系:膜的流动性用微粘度表示,其值越小,细胞膜脂区的流动性越高⁽⁷⁾。本文的观察结果表明,冠心病血瘀证患者和恶性肿瘤血瘀证患者的红细胞膜微粘度明显高于健康人,即红细胞膜的流动性降低。变形是红细胞膜的一种特殊功能,在血液循环过程中,特别是在微循环过程中,红细胞的形状经常发生改变,由双凹盘形变为折叠状,以便顺利通过口径小于其直径的毛细血管网,红细胞膜的流动性和红细胞的变形能力有关⁽⁸⁾,所以它也是影响血液流动性的一个重要因素之一。冠心病和恶性肿瘤血瘀证患者红细胞膜的流动性减小,可使红细胞的变形性降低,从而导致微循环障碍,并容易形成微血栓。

微循环淤滞是血瘀证发生的病理基础。本文的观察结果表明,血瘀型冠心病、恶性肿瘤患者的红细胞膜流动性降低。这提示,血瘀证患者微循环淤滞的发生除与血流动力学紊乱、血液粘滞性增高等病变有关外,红细胞膜微粘度增大、膜流动性和细胞变形性降低可能也是一个重要病理因素。

二、影响血瘀证患者红细胞膜流动性的因素:红细胞膜流动性在很大程度上受血清脂蛋白或脂肪成份的影响,血清脂蛋白中胆固醇含量或游离胆固醇总量或胆固醇/磷脂(C/PL)比值升高,可导致红细胞膜胆固醇含量增加或C/PL比值升高,而使红细胞膜的流动性降低。本文观察的冠心病血瘀证患者的红细胞膜微粘度增大,可能就该组患者的血脂偏高有关,21例患者中有9例患者的血浆胆固醇含量升高。

肿瘤细胞膜的流动性研究表明,一些肿瘤细胞膜的流动性明显高于正常细胞^(9~11),有的

学者甚至提出可将肿瘤细胞的这一变化作为肿瘤的辅助诊断指标⁽¹¹⁾。但肿瘤细胞膜流动性升高的病理机理尚不清楚。导致恶性肿瘤血瘀证患者红细胞膜微粘度增加的病理因素也有待进一步探讨。

参 考 文 献

1. 中国中西医结合研究会活血化瘀专业委员会。血瘀证诊断标准。中西医结合杂志 1987; 7(3):129.
2. Cooper RA, et al. Factors influencing the lipid composition and fluidity of red cell membranes in vitro: Production of red cells possessing more than two cholesterol per phospholipid. Biochemistry 1978; 17:327.
3. Azumi T, et al. Polarization of the luminescence of phenanthrene. J Chem Phys 1962; 37:2413.
4. 聂松青,等。蜂王精对大鼠红细胞膜流动性的影响。北京医学院学报 1983; 15:249.
5. Shinitzky M, et al. Fluidity parameters of lipid regions determined by fluorescence polarization. Biochim Biophys Acta 1978; 515:367.
6. 林克椿,等。用荧光探针DPH研究腹水癌细胞流动性。生物化学与生物物理进展 1981; 42:32.
7. Owen J, et al. Decreased erythrocyte membrane fluidity and altered lipid composition in human liver disease. J Lipid Res 1982; 23:124.
8. Cooper RA. Abnormalities of cell-membrane fluidity in the pathogenesis of disease. N Engl J Med 1977; 297:371.
9. 董伟志,等。健康人、非癌患者及癌患者外周血淋巴细胞荧光偏振度的测定。生物化学及生物物理进展 1983; 3:34.
10. 赵保路,等。用自旋标记研究抗癌药物对中国地鼠肺正常细胞V79和癌变细胞V79-B1膜流动性的影响。生物化学与生物物理学报 1984; 1:43.
11. Inbar M, et al. A method for the quantitative detection of human acute Lymphatic leukemia. Europ J Cancer 1977; 13:1235.

本刊启事 《中西医结合杂志》由北京新华印刷厂承担印刷、装订。如有残缺、错页等,请直接寄往北京市车公庄新华印刷厂质量检验科调换。

更正 本刊“昆布粉对实验性高脂蛋白血症及其血液流变学的影响”一文(1989; 9(4):223)第二作者为沈士芳,特此更正。

were less satisfactory. By using EYP for treating symptoms of aging the total effective rate was 96.8%, nootropil 71.7% and placebo 41.2%. There were noticeable differences in three groups. Those results suggested that EYP was one of the effective therapies for decreased fluid intelligence with aging.

(Original article on page 398)

Clinical and Experimental Research of Kidney-Tonifying Prescription in Prevention and Treatment of Children's Hearing-Loss Induced by Aminoglycosid Antibiotic Ototoxicity

Lin Wensen(林文森), Song Xuezheng*(宋学政), et al

Tianjin ENT section of Integrated Traditional and Western Medicine, Nankai Hospital, Tianjin

**Academy of Military Medical Sciences of PLA, Beijing*

This paper reports 30 cases of hearing-loss induced by aminoglycosid antibiotic ototoxicity treated mainly with Kidney-tonifying herbs such as Herba Epimedii, Rhizoma Drynariae, Rhizoma Polygonati, Radix Polygoni Multiflori, Magnetitum and Rhizoma Acori Graminci, etc. After three month's treatment, the authors found that 19 cases had improved their hearing more than 15 db, among which there are 8 cases who had improved 15 db and 11 cases 30 db. The patients' hearing of pre-treatment and post-treatment were determined by BAR V-wave hearing threshold.

In order to further prove that Kidney-tonifying prescription can antagonize the aminoglycosid antibiotic ototoxicity, animal experiments had been made. The authors chose guinea pigs for experimental animals, which were randomly divided into three groups: Kanamycin group, kanamycin with Kidney-tonifying herbs group, and control group. The indexes of the experiments were the animals' helix reflection index, BAR and observation of animals' cochlear under electronmicroscope. As a result, the damage to hair cell of Cortis organ has marked for the animals of kanamycin group, while for kayamycin with herbs group, it was obviously slighter ($P < 0.01$). There was significant difference between the groups. The result indicates that Kidney-tonifying herbs had the effect of protecting the animals' hair cells of cortis organ against aminoglycosid antibiotic ototoxicity and thus protected the animals' hearing.

(Original article on page 402)

Observation on the Fluidity of Human Erythrocyte Membrane in Patients with Blood Stasis Syndrome

Hu Qingfu(胡庆福), Chen Zelin(陈泽霖), et al

Zhong Shan Hospital, Shanghai Medcial University, Shanghai

In order to approach the relationship between the TCM typology of blood stasis syndrome and the fluidity of human erythrocyte membrane, the fluidity of human erythrocyte membrane was examined in 14 healthy volunteers, 21 patients with coronary heart disease and 14 patients with cancer by measuring the degree of fluorescence polarization using DPH as a fluorescent probe. The patients with coronary heart disease and the cancer patients were all of the TCM typology of blood stasis syndrome. The results showed: (1) The fluorescence polarization (P) of normal group was 0.294 ± 0.007 , and the microviscosity (π) was 3.656 ± 0.237 . (2) The P of the group of coronary heart disease with blood stasis syndrome was 0.328 ± 0.013 , and the π was 6.787 ± 1.028 . Both were significantly higher than those of the normal group ($P < 0.05$). (3) The P of the group of cancer with blood stasis syndrome was 0.333 ± 0.011 , and the π was 6.698 ± 1.357 . Both were significantly higher than those of the normal group ($P < 0.05$). (4) Yet no significant difference was noticed between the group of coronary heart disease and the group of cancer ($P > 0.05$).

The higher the π of erythrocyte membrane, the lower the fluidity of erythrocyte membrane. The lower fluidity of erythrocyte membrane in patients with blood stasis syndrome can result in the declined ability of erythrocytic deform which can accelerate the disturbance of microcirculation. This accords with the TCM theory about blood stasis syndrome.

(Original article on page 405)