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酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的结果观察*

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摘要 目的:比较酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的结果,以探讨酶联免疫法和胶体金法在检验与病理科乙肝表面抗原检测中的应用价值。**方法:**选择2017年1月~2019年12月我院进行胃镜检查的100例上消化道疾病患者,均在胃镜检查前检测乙肝表面抗原,对照组使用胶体金法进行检测,观察组使用酶联免疫法进行检测。比较酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率、敏感度以及特异度。**结果:**酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率分别为35.00%(35/100)和33.00%(33/100),两种方法相比较没有明显的差异($P>0.05$);酶联免疫法在胃镜检查前检测乙肝表面抗原的敏感度(97.34%)与特异度(98.12%)均明显高于胶体金法($P<0.05$)。**结论:**酶联免疫法和胶体金法在胃镜检查前对乙肝表面抗原均有比较高的阳性检测率,但是酶联免疫法具有更高的敏感度以及特异度,仍然是现今检测乙肝表面抗原的主要方法,胶体金法由于检测方法更加简便和直观、方便携带、结果更加快捷,而且不需要特殊的仪器设备,更加适用于临床上紧急状态下患者的检测以及感染者初筛等情况。

关键词:酶联免疫法;胶体金法;胃镜检查;乙肝表面抗原;检测结果

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Observation on Results of ELISA and Colloidal Gold Method in Detection of Hepatitis B Surface Antigen Before Gastroscopy*

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ABSTRACT Objective: To compare the results of enzyme-linked immunosorbent assay and colloidal gold method in the detection of hepatitis B surface antigen before gastroscopy. **Methods:** A total of 100 patients with upper gastrointestinal diseases, who underwent gastroscopy in Ningxia General Hospital of Armed Police Force from January 2017 to December 2019, were selected and were detected HBsAg before gastroscopy. The control group was detected by colloidal gold method, and the observation group was detected by ELISA. The positive rate, sensitivity and specificity of ELISA and colloidal gold method in detecting HBsAg before gastroscopy were compared. **Results:** The positive rates of ELISA and colloidal gold in detecting HBsAg before gastroscopy were 35.00% (35/100) and 33.00% (33/100), respectively. There was no significant difference between the two methods ($P>0.05$). The sensitivity (97.34%) and specificity (98.12%) of ELISA in detecting HBsAg before gastroscopy were significantly higher than those of colloidal gold method ($P<0.05$). **Conclusion:** Two enzyme linked immunosorbent assay and colloidal gold method have high positive detection rate for HBsAg before gastroscopy. However, ELISA is still the main method to detect HBsAg due to its higher sensitivity and specificity. Colloidal gold method is more simple and intuitive, convenient to carry, faster results, and does not need special equipment, which is more suitable for the detection of patients in clinical emergency and the initial screening of infected persons.

Key words: Enzyme linked immunosorbent assay; Colloidal gold method; Gastroscopy; Hepatitis B surface antigen; Detection results

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前言

乙型肝炎属于全球范围内感染人数最多的一种慢性传染性疾病。随着乙型肝炎在全球范围内的日益流行,如何有效的

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预防乙型肝炎已经成为全球范围内一个极为重要的公共卫生问题^[1-3]。临床研究发现,乙肝病毒表面抗原是引起乙肝的致病菌的一种重要血清学标志物^[4]。乙型肝炎检测在临床上主要是用于胃镜检查前以及血液检测。由于患者在进行胃镜检查的过程中,可能会引起胃黏膜破损和出血的不良情况,如果是乙型肝炎患者,必须采取专用的检查仪器给予检查,不然可能会引起交叉感染现象。因而,在胃镜检查前需要常规检测乙型肝炎表面抗原,以保护自己和患者的安全。常见的检测手段主要包含酶联免疫法与胶体金两种^[5,6]。酶联免疫法将免疫酶技术作为基础,具有非常高的特异度以及敏感度,但是同时也存在操作过程比较复杂以及检测所需的时间比较长等缺点,对检测人员、检测环境和检测设备均有比较高的要求^[7,8]。胶体金主要是由氯金酸以及还原剂等发生反应进而聚合形成的颗粒,其容易受静电的影响,在弱碱的状态下,胶体金会产生负电荷,可以与蛋白质分子的正电荷发生结合,而且胶体金具有快捷性、方便性和稳定性等优点,但敏感度较低^[9]。本研究创新性地分析了酶联免疫法和胶体金法在检验与病理科乙肝表面抗原检测中的应用价值。

1 资料与方法

1.1 一般资料

选择 2017 年 1 月~2019 年 12 月我院进行胃镜检查的 100 例上消化道疾病患者,男 57 例,女 43 例;年龄 19~73 岁,平均(51.29±13.78)岁;体重 34~103 kg,平均(57.43±15.92)kg;其中,食管炎患者 34 例,消化道肿瘤患者 35 例,慢性胃炎患者 31 例。

1.2 研究方法

胶体金法和酶联免疫法检测所使用的仪器主要包括:MW-12A 洗板机(深圳迈瑞公司生产)、MR-96A 酶标仪(深圳

迈瑞公司生产)、HR40-IIA2 生物安全柜(青岛海尔公司生产)、HH.W2.Cu600 型电热恒温水浴箱(上海医疗器械六厂生产)、BY-400C 型医用离心机(北京白洋医疗器械公司生产)、Thermo 单道移液器 10~100 μ L(上海热电公司生产)。检测试剂:胶体金法和酶联免疫法所使用的分别购自泉州市睿信生物公司和卡迈舒(上海)生物公司。检测方法:采取不含有抗凝剂的真空采血管,对本研究中进行胃镜检查的 100 例上消化道疾病患者进行样本采集,在采集前必须确保患者已经至少空腹 12 h,抽取 5 mL 的静脉血,在室温的条件下自然放置 1~2 h,当血液发生凝固、血块发生收缩后,按照 3000 r/min 的速度进行 15 min 的离心,将血清吸出,放在提前准备好的容器内,血清待检。胶体金法采取移液器将上述待检的血清滴加至试剂加样处,每次加样完成后需要更换一个吸头,20 min 后观察且记录胶体金法的检测结果。酶联免疫法需要首先将试剂在室温的环境下平衡 30 min 后再进行加样,经过温育、洗板、加酶以及显色等多个步骤,加终止液后的 10 min 内,酶标仪即可以显示检测结果。

1.3 观察指标

比较酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率、敏感度和特异度。

1.4 统计学分析

采用 SPSS 21.0,两组间计量资料对比用 t 检验,计数资料用 χ^2 检验,以 $P<0.05$ 为差异有统计学意义。

2 结果

2.1 酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率对比

酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率分别为 35.00%(35/100)和 33.00%(33/100),两种方法相比较没有明显的差异($P>0.05$),见表 1。

表 1 酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率对比[例(%)]

Table 1 Comparison of positive rate of HBsAg detected by enzyme linked immunosorbent assay and colloidal gold before gastroscopy [n(%)]

Testing Method	n	Negative	Positive	Positive rate
Enzyme linked immunosorbent assay	100	65	35	35(35.00)
Colloidal gold method	100	67	33	33(33.00)

2.2 酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的敏感度与特异度对比

酶联免疫法在胃镜检查前检测乙肝表面抗原的敏感度

(97.34%)与特异度(98.12%)均明显高于胶体金法($P<0.05$),见表 2。

表 2 酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的敏感度与特异度对比[例(%)]

Table 2 Comparison of sensitivity and specificity of ELISA and colloidal gold method in detection of HBsAg before gastroscopy[n(%)]

Testing Method	n	Sensitivity	Specificity
Enzyme linked immunosorbent assay	100	97.34*	98.12*
Colloidal gold method	100	89.13	90.12

Note: Compared with colloidal gold method, * $P<0.05$.

3 讨论

据病理学统计,全球 2010 年乙肝表面抗原阳性人数大约为 2.48 亿,其中,中国所占的比例高达 45%,乙型肝炎病毒感

染者中大约有 30%经由母婴传播途径获得^[10]。乙肝表面抗原属于乙型肝炎病毒外壳蛋白的一种主要成分,也是判断患者感染乙型肝炎病毒的重要标志,是目前临床上乙型肝炎血清标志物检测中极为重要的一项,可在机体感染乙肝病毒后的早期出

现,且自身没有传染性,但乙肝表面抗原的出现往往会伴随乙肝病毒的存在,因此,临床上将其作为已感染乙肝病毒的重要标志物^[11-15]。胃镜由于具有形象直观、操作简便和准确率比较高等优点,而成为诊治上消化道系统疾病最为常用的一种手段^[16-18]。在胃镜检查前,通过乙型肝炎表面抗原检查对患者的情况进行准确和有效的鉴别,并且以检测的结果来判断是否需要对患者采取针对性的消毒以及必要的隔离手段,一旦出现乙型肝炎表面抗原阳性的结果,在胃镜检查的过程中,需要注重污染物以及医疗器械的处理,避免发生乙肝传播,提醒医院内相关的医疗人员做好周全的防护措施。

酶联免疫法因为比较容易实现自动化结果判读和实验操作,有利于大批量样本的结果追溯以及检测,因此被广泛应用于血站的乙型肝炎病毒检测中^[19]。酶联免疫法可以将酶对抗原以及抗体的特异性免疫反应进行结合,具有非常高的敏感性以及特异性,其原理主要是把抗原或抗体固定于载体的表面,将酶和抗体或抗原进行结合而形成酶标抗体或酶标抗原,同时具有免疫活性以及酶活性,然后采用酶标仪判断检测的结果^[21-24]。酶联免疫法是继酶免疫、荧光免疫和放射免疫之后的一种新型标记免疫技术,具有重复性好和灵敏度高的优点,可以定量检测或半定量检测,但是酶联免疫法容易受到多种因素的影响,如标本保存,血清分离,加样有无溅出和气泡,试剂在使用前是否平衡到室温,加样有没有加在孔壁部的非包被区,洗板情况,孵育的方式以及时间等,每一步的操作均会对酶联免疫法检测的结果造成一定程度的影响;而且,酶联免疫法由于需要特殊的设备,反应的时间比较长等,不适合应用于无偿献血以及门急诊患者的初筛工作中^[25,26]。胶体金法的检测原理主要是把特异性的抗体固定于膜特定区域后,加入待测样本血清,血清中的抗原可以与膜上的抗体发生特异的性结合,然后把胶体金作为示踪的标记物,通过观察试纸条颜色的变化,就可以判断检测的结果,一般情况下在 10 min 内可以获得结果,具有速度快和检测方便的优点,主要被应用在临床上的定性检测中,但其敏感度相对较低^[27,28]。因为胶体金显色必须有比较高浓度的标记物,使其灵敏度受到一定程度的限制,在浓度比较低时很容易产生假阴性,因此对于低滴度的早期乙型肝炎感染者很容易造成漏检^[29]。

本研究发现,酶联免疫法和胶体金法在胃镜检查前检测乙肝表面抗原的阳性率分别为 35.00 %和 33.00 %,两种方法相比没有明显的差异;表明酶联免疫法和胶体金法在乙肝表面抗原的检测方面具有较高的一致性。酶联免疫法以及胶体金法均能满足临床诊断乙型肝炎的需要,两种检测手段各有利弊,应当按照实际工作的需要选择采取适合的方法,例如当进行无偿献血以及门急诊患者的乙肝表面抗原初筛工作、紧急状态下患者的乙肝表面抗原检测时,需要优先考虑使用胶体金法;如果需要对乙型肝炎病人进行明确的诊断,或胶体金的试纸条法明确显示阳性的患者进行复诊时可以优先考虑采用酶联免疫法。酶联免疫法在胃镜检查前检测乙肝表面抗原的敏感度(97.34 %)与特异度(98.12 %)均明显高于胶体金法。表明酶联免疫法在胃镜检查前对乙肝表面抗原的检测具有更高的敏感度以及特异度。与周成等^[30]的研究结果基本一致。分析其原因

为,酶联免疫法可以把抗原抗体反应的特异性以及酶化学反应的敏感性进行有效的结合,因此该方法不但敏感,而且具有比较高的特异性;而且酶联免疫法具有比较高的催化反应转化率以及比较高的酶纯度,并且性质比较稳定,专一性比较强,对乙肝表面抗原的灵敏度明显高于凝集反应^[31,32]。

综上所述,酶联免疫法和胶体金法在胃镜检查前对乙肝表面抗原均有比较高的阳性检测率,但是酶联免疫法具有更高的敏感度以及特异度,仍然是现今检测乙肝表面抗原的主要方法,胶体金法由于检测方法更加简便和直观、方便携带、结果更加快捷,而且不需要特殊的仪器设备,更加适用于临床上紧急状态下患者的检测以及感染者初筛等。

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