中南大学学报(医学版) 2011, 36(1) 1- DOI: 10.3969/j.issn.1672 7347.2011. ISSN: 1672-7347 CN: 43-1427/R

论著

湖北钉螺人工感染、室内传代与模拟野外饲养的研究

夏英定， 汪世平， 刘雪琴， 高冬梅， 李庆华， 吴平， 陈秀春， 冯其梅， 周云飞， 张树菊

中南大学湘雅医学院寄生虫学系，血吸虫病免疫与传播控制湖南省重点实验室，长沙 410078

摘要：

目的：通过人工方法将湖北钉螺制备成血吸虫感染性钉螺，确定钉螺感染的最佳条件，建立钉螺人工感染传代的室内株，为研究其感染活性、遗传变异和疫苗等提供实验室依据。方法：用尼龙绢筛集卵法收集日本血吸虫成熟虫卵，常规法孵化毛蚴。将钉螺与毛蚴按不同比例进行感染，感染方式分为个体感染和集体感染。个体感染随机分6组(Ⅰ～Ⅵ组)，每组200只钉螺，每只钉螺置单孔内分别感染，钉螺感染毛蚴比例分别为1∶0，1∶5，1∶10，1∶15，1∶20，1∶25；集体感染随机分6组（Ⅶ～Ⅻ组），每组200只钉螺，按组别集中感染，Ⅶ～Ⅻ组钉螺感染毛蚴比例分别同Ⅰ～Ⅵ组。然后对每组钉螺的感染数、死亡数及尾蚴逸出量进行比较，确定最佳感染方法和比例。以第1代人工感染性钉螺逸出的尾蚴感染实验动物，获取成熟虫卵并孵化毛蚴，然后采用个体感染方式，以1∶15的比例继续感染钉螺，获得第2代人工感染性钉螺。比较第1代与第2代人工感染性钉螺的感染数、死亡数及尾蚴逸出数。通过动物感染实验，比较人工第1代、第2代感染性钉螺与自然感染性钉螺日本血吸虫成虫发育率、每克粪卵数（fecal eggs per gram, FEPG）及每克肝卵数（liver eggs per gram, LEPG）。 结果：个体感染Ⅰ～Ⅵ组的钉螺感染数分别为0±0，22.7±4.2，31.7±4.5，53.0±5.3，39.3±5.9，32.7±4.7；钉螺死亡数分别为21.7±3.1，25.0±3.6，31.3±4.9，44.7±6.5，78.3±9.5，89.7±13.6；钉螺平均逸蚴量为0±0，308.0±96.6，428.1±146.2，527.0±171.1，571.4±148.9，602.9±356.3。集体感染Ⅶ～Ⅻ组，钉螺感染数分别为0±0，12.3±2.5，18.7±4.7，28.3±4.2，33.3±4.7，29.3±5.5；钉螺死亡数分别为22.7±3.8，23.7±4.5，28.3±5.5，47.0±9.5，75.7±8.5，86.3±12.2；钉螺平均逸蚴量为0±0，244.5±57.3，292.3±74.8，347.1±100.8，477.2±142.1，447.3±161.4。用人工制备的第1代感染性钉螺对血吸虫进行人工传代研究，成功获得了人工第2代感染性钉螺，感染率为24.65%，钉螺死亡率为24.50%；与人工第1代钉螺26.65%的感染率及22.35%的死亡率差异均无统计学意义（P>0.05）。在尾蚴感染动物试验中，人工第1代、第2代感染性钉螺与自然感染性钉螺的成虫发育率分别为68.50%,73.50%,71.00%，3组间差异无统计学意义（P>0.05）；自然感染性钉螺和人工第1代、第2代感染性钉螺的FEPG分别为1 503±269，1 683±233，1 541±117；LEPG分别为6 641±1 819，6 272±1 419，7 263±1 643，3组间比较差异无统计学意义（P>0.05）。 结论：通过用人工感染的方法可以获得日本血吸虫感染性钉螺。个体感染方式优于集体感染方式，感染时钉螺与毛蚴的最佳比例为1∶15。人工感染性钉螺经传代后，第1代与第2代钉螺在感染数、死亡数及尾蚴逸出数等方面无明显差别。比较人工第1代、第2代感染性钉螺与自然感染性钉螺的成虫发育率、FEPG及LEPG，差异也无统计学意义,证明人工传代的血吸虫尾蚴（室内株）能达到自然野生株尾蚴的感染效果。

关键词： 日本血吸虫；湖北钉螺；毛蚴；人工感染；传代

Artificial preparation, indoor passage, and nature breed of Oncomelania hupensis infected with Schistosoma Japonicum

XIA Yingding， WANG Shiping， LIU Xueqin， GAO Dongmei， LI Qinghua, WU Ping, CHEN Xiuchun, FENG Qimei, ZHOU Yunfei, ZHANG Shuju

Department of Parasitology, Medicine School of Xiangya, Central South University; Key Laboratory of Immune and Control Schistosomiasis of Hunan Province, Changsha 410078， China

Abstract:

ObjectiveTo prepare the infected Oncomelania hupensis by artificial method for the research on the activity, vaccine, and genetic variation of Schistosoma Japonicum (S. Japonicum).MethodsThe mature eggs of S. Japonicum were collected by Nylon silk method and the miracidia were incubated under appropriate conditions. Negative snails were infected with miracidia in different proportion by means of individual or collective infection to seek the best method and proportion of infection between miracidia and snails. Infected snails were divided into 12 groups in total. Ⅰ-Ⅵ groups were for individual infection and Ⅶ-Ⅻ groups were for collective infection. There were 200 snails in each group. The infection ratios between snails and miracidia in Group Ⅰ-Ⅵ or Ⅶ-Ⅻ were 1∶0，1∶5，1∶10，1∶15，1∶20，1∶25, respectively. The infected snails were screened, numbered, and reared singly. The amount of cercariae was calculated once every 10 days until the infected snails died. Then cercariae shedding quantity, infection quantity, and mortality of infected snails in every group were compared to find the best infection method and the best infection proportion between miracidia and snails. The cercariae were collected from the first generation of infected snails and were used to infect experimental animals. The mature eggs of S. Japonicum were saved from the infected experimental animals and incubated to get

miracidia. The snails were artificially infected by miracidium to get the second generation of infected snails. The developmental rates of adult worms, the egg density in fecal and liver were compared between artificially and naturally infected snails. ResultsIn individual infection Group Ⅰ-Ⅵ，the average infection value of snails were 0±0，22.7±4.2，31.7±4.5，53.0±5.3，39.3±5.9，32.7±4.7，the average fatality of snails were 21.7±3.1，25.0±3.6，31.3±4.9，44.7±6.5，78.3±9.5，89.7±13.6, and the average value of cercariae shedding from infected snails were 0.0±0.0，308.0±96.6，428.1±146.2，527.0±171.1，571.4±148.9，602.9±356.3, respectively. In collective infection Group Ⅶ-Ⅻ，the average infection value of snails were 0±0，12.3±2.5，18.7±4.7，28.3±4.2，33.3±4.7，29.3±5.5，and the average fatality of snails were 22.7±3.8，23.7±4.5，28.3±5.5，47.0±9.5，75.7±8.5，86.3±12.2, and the average value of cercariae shedding from infected snails were 0±0，244.5±57.3，292.3±74.8，347.1±100.8，477.2±142.1，447.3±161.4, respectively. The second generation of artificially infected snails was obtained successfully. The average infection rate and fatality rate for the second generation of artificially infected snails were 24.65% and 24.50%, both of which were not obviously different from that of the first generation of artificially infected snails (P>0.05). In the animal experiment, the worm growth rate for the naturally infected snails, the first or second generation of artificially infected snails were 68.50%，73.50% or 71.00%. There was no obvious difference among them (P>0.05). The fecal (or liver) eggs per gram for the naturally infected snails, the first or the second generation of artificially infected snails were 1 503±269，1 683±233， or 1 541±117 (or 6 641±1 819，6 272±1 419， or 7 263±1 643). There was no significant difference among the 3 groups (P>0.05). ConclusionInfected snails can be obtained through the artificial method by using S. Japonicum miracidia to infect snails. Individual infection has the advantage over collective infection. The optimal proportion of infection between snails and miracidia is 1∶15. There was no significant difference between the first and the second generation of artificially infected snails in the average of cercariae shedding, infection, and fatality average of snails. There was no significant difference between artificially and naturally infected snails in the developmental rate of adult worms, fecal and liver eggs per gram.

Keywords: Schistosoma Japonicum;Oncomelania hupensis;miracidia;artificially infected;passage

收稿日期 2010-09-03 修回日期 网络版发布日期

DOI: 10.3969/j.issn.1672 7347.2011.

基金项目:

通讯作者: 汪世平

作者简介:

作者Email: spwang@126.com

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