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## A randomized controlled trial of sequential cardiac auscultation for medical students

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### Abstract

**Objectives:** Cardiac auscultation is an important technique for assessing the heart. However, because of recent technological developments, stethoscopes have gradually been abandoned and the ability of young physicians to perform auscultation has declined. We aimed to compare a sequential cardiac auscultation teaching method with the traditional teaching method and to determine whether the new method can improve the cardiac auscultation ability of medical students.

**Methods:** A randomized controlled trial for cardiac auscultation was performed, involving 180 junior-year medical undergraduates. Sequential cardiac auscultation was used as an experimental intervention and was compared with the traditional method used in a control group. Evaluation of all participants was conducted through cardiac auscultation on a simulated patient. Each test was composed of 20 standard cardiac auscultation problems delivered immediately after and 1 month and 6 months after teaching intervention (T2 and T3). A self-administered questionnaire was used to evaluate the students' attitudes, total learning times, and conditions under each teaching method.

**Results:** There was no significant difference in evaluation grade between the two groups immediately after teaching intervention. Later, the grades of the two groups decreased to some extent. The decrease in the experimental group was less sudden than that in the control group. At 1 month and 6 months after teaching intervention, the grades in the experimental group were significantly better than those in the control group (T2:  $16.80 \pm 1.79$  vs.  $15.53 \pm 2.19$ ,  $t = 2.451$ ,  $p = 0.017$ ; T3:  $15.40 \pm 2.22$  vs.  $13.37 \pm 3.09$ ,  $t = 2.926$ ,  $p = 0.005$ ). Most students in the experimental group held positive attitudes toward the sequential cardiac auscultation teaching method.

**Conclusions:** Cardiac auscultation is a necessary skill for physicians. The sequential cardiac auscultation teaching method can allow young medical students to simply and effectively grasp aspects relevant to cardiac auscultation.

### Article summary

Strengths and limitations of this study the SR2SMP sequential cardiac auscultation method simplifies many complicated points of knowledge into a text description through highly generalized knowledge and summaries. The SR2SMP sequential cardiac auscultation method reduces the absolute amount of material to be memorized and divides the material into modules for memorization. The SR2SMP sequential cardiac auscultation method provides the student with even more familiarity and confidence when they eventually deal with real patients. Due to the limited teaching hours, the results should be generalized with caution. Further large-scale and more in-depth studies are needed to verify these results

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## 1. Introduction

Cardiac auscultation is an extremely important medical technique for students and established physicians. The necessity of cardiac auscultation can be best demonstrated through a representative case. A 46-year-old pre-menopausal woman was admitted to hospital after experiencing chest pain for 3 hours. Her symptoms presented as pinprick-like chest pain that resolved in seconds and was unrelated to activity. Electrocardiography revealed a T wave peak in leads  $V_1$ – $V_3$ . Because this electrocardiographic change is common among women and often a normal variation, the patient's chest pain did not meet the typical characteristics of ischemic chest pain. In addition, the patient had no risk factors for coronary heart disease, so the young physician who admitted the patient initially allowed her to be discharged for observation. However, the initial cardiac physical examination of the patient revealed a very high pitch in the  $S_2$  aortic auscultation area similar to "the sound produced by a taut string of a musical instrument." The physician was curious and concerned about this finding, so ordered a precautionary troponin test. The results showed a significant increase in troponin level and follow-up coronary angiography confirmed occlusion of the distal left anterior descending artery. Interestingly, when the patient was re-examined by the night-shift physician, the abnormal sounds in the aortic auscultation area had disappeared. The possible reason for the late appearance of high  $S_2$  pitch is early-stage papillary muscle dysfunction following myocardial infarction. These cases are common in the course of clinical practice and have one aspect in common: special findings upon cardiac auscultation. These findings can be very subtle but can explain certain other disorders when clinical data cannot.

In 1816, the stethoscope was invented by Laennec. After the introduction of "indirect auscultation," the practice of "direct auscultation"

by placing the ear directly onto the chest of the patient was abandoned, ushering in an era of quality in the practice of auscultation. Cardiac auscultation is the important technique of using a stethoscope to obtain clinical information about the heart (Fang JC. & O'Gara PT., 2015). It has many advantages, including being simple, direct, and rapid, and has important significance for the diagnosis of heart disorders. However, because of recent technological developments, especially the widespread use of echocardiography, stethoscopes have gradually been abandoned and the ability of young physicians to perform auscultation has declined. In fact, the stethoscope has now become something of a decorative symbol worn around the neck of physicians (Sztajzel J.M. et al., 2010)(Mangione S. et al., 1997). A recent training survey found that the auscultation ability of medical students and young physicians was a cause for concern, and some researchers have even called for a "defense" of the stethoscope (Vukanovic-Criley J.M. et al., 2006)(Mangione S., 2001)(Shindler D. , 2007).

Cardiac auscultation is an important and difficult aspect of cardiac physical diagnostics education. Important information that can serve as a basis for diagnosis can be obtained by performing proper auscultation. Thus, cardiac auscultation is a basic practical skill that medical students and clinical physicians must master<sup>6</sup>. However, because cardiac auscultation involves a large body of knowledge, the time available for clinical education is limited, and there is a lack of active, direct teaching methods, cardiac auscultation has become a difficult aspect of medical education that students find challenging to master (Shaver J.A. et al., 1995).

In addition, although standardized electronic patient simulations have solved the problem of few opportunities for clinical auscultation practice and a lack of typical cases, they are associated with their own limitations and disadvantages. For example,

simulations do not allow the student to interact with a real patient and the audio signal transmitted through the simulation equipment is often distorted, which often leads to a lack of three-dimensionality in understanding auscultation sounds. Finally, simulation-based education is a supportive teaching procedure or tool and often must be combined with many other teaching procedures in order to be effective (Cook D.A. et al., 2011)(Issenberg S.B. et al., 1999)(Fraser K. et al., 2009)(Butter J. et al., 2010)(Sverdrup Ø. et al., 2010)(Fraser K. et al., 2011)(Norman G. et al., 2012)(McKinney J. et al., 2013)(Chen R. et al., 2015)(Takashina T. et al., 1997)(Issenberg S.B. et al., 1999).

Based on the above and our many years of experience in teaching cardiac auscultation, we proposed a sequential auscultation method to aid in the acquisition of cardiac auscultation skills. In this method, cardiac auscultation training is conducted according to the sequence of heart sounds, heart rate, heart rhythm, extra heart sounds, murmurs,

## 2. Methods

### 2.1. Subjects

We performed a randomized controlled trial of 180 third-year clinical medical students who were selected from the 2015–2016 class at our university. Computerized randomization methods were used to randomly assign the participants into two groups; 90 participants were included in the experimental SR<sub>2</sub>SMP sequential cardiac auscultation-teaching group and 90 in the traditional teaching methods group.

### 2.2. Teaching methods

The two groups used the same teaching materials, course curriculum, and practical guidance. The qualifications and experience of the instructors, teaching progression, and examination format were consistent between the groups. During the cardiac auscultation section, the two groups used either the cardiac sequential auscultation method or the traditional teaching method. The cardiac auscultation section included a total of four

and pericardial friction sound (SR<sub>2</sub>SMP). Corresponding teaching content is arranged in each training module. For example, in the heart rhythm unit, cardiac auscultation is taught in the context of regular heart rhythms and typical irregular heart rhythms (sinus arrhythmia, premature ventricular contraction, and atrial fibrillation). When medical students hear irregular heart sounds, they associated them with the corresponding clinical pathology, which allows cardiac auscultation practice to become a procedural, sequential, dynamic process that can be followed and closely combined with exposure to clinical situations.

The purpose of the present study was to compare the SR<sub>2</sub>SMP approach and the traditional teaching methods with respect to compatibility with the teaching curriculum, educational simulation patients, and actual patients. We aimed to assess whether this new method can improve the cardiac auscultation ability of medical students and whether this teaching method helps students in the long term.

classroom units and was arranged to be conducted over 4 consecutive weeks (Table 1). The specific teaching methods were as follows.

### 2.3. Teaching preparation stage

(a) Instructor preparation: One year before beginning teaching with the experimental sequential auscultation method, the instructor broke down and refined knowledge of cardiac auscultation teaching content, mentioned that the cardiac sequential auscultation method was simpler and easier to memorize, and proposed it as a new method for cardiac auscultation technique training after approval by a panel of experts. The traditional methods instructor only performed conventional collective lesson preparation and other activities.

(b) Student preparation: Ten days before the study, educational administration instructors randomly divided the students involved in the study into two groups. The instructor explained the purpose and outline of the teaching program to all

students, described the teaching process and requirements to the two groups of students, and obtained informed consent forms from the students after hearing their opinions. One week before teaching began, the teaching assistant posted the

teaching materials on the course homepage, which included section information, PPT courseware, reference materials, references, practice problems, audio data, and so on.

**Table 1: Teaching information**

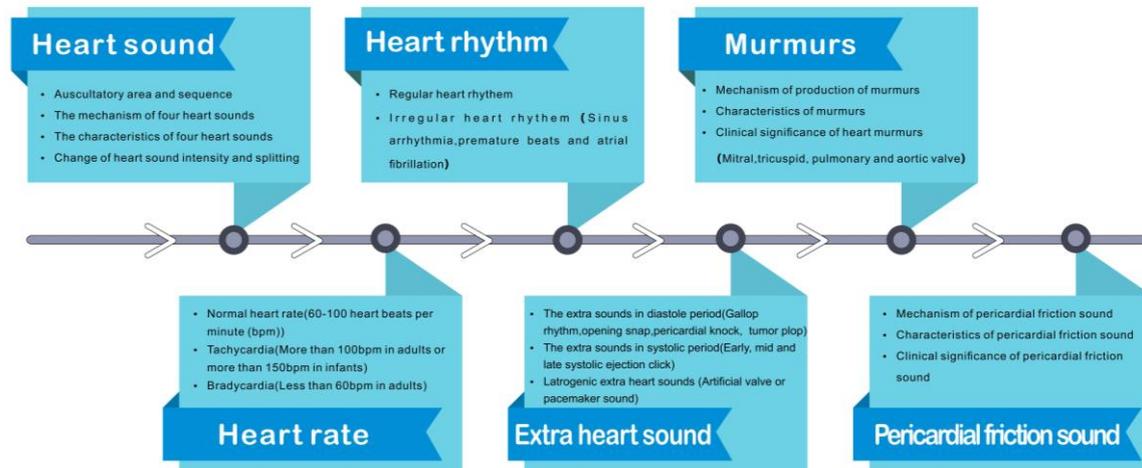
Teaching units	Teaching contents	Time (min)	Teaching methods
1	General information of cardiac auscultation, heart sound, heart rate, heart rhythm, extra heart sound	135	Lecture
2	Murmurs, pericardial friction sound	135	Lecture
3	Cardiac auscultation probation	135	Practice by simulator
4	Cardiac auscultation probation	135	Practice in ward

#### 2.4. Teaching stages

The teaching content was divided into four teaching units, with each unit taking 135 min (Table 1). Units 1 and 2 were lectures in which the instructor presented theoretical knowledge according to curriculum requirements and offered interaction with the students in the form of simplified quizzes, discussions, and question-and-answer sessions depending on the students' needs after presenting the most important points. Units 3 and 4 were practical courses. First, simulation education was provided, in which students practiced cardiac auscultation using an electronic human simulation system to diagnose different heart rates, heart rhythms, changes and interruptions in heart sounds, and extra-heart sounds. They also performed repeated auscultation and practiced identifying typical heart murmurs with guidance from the instructor. Next, ward rounds were conducted, during which students practiced bedside cardiac auscultation using multifunctional infrared

stethoscopes with guidance from the instructor to further consolidate the theoretical knowledge gained from working with the simulations and to experience the different challenges of cardiac auscultation in actual patients. The experimental and control groups used the sequential auscultation method and the traditional teaching method, respectively. Students' ability was tested at the end of each stage.

The SR<sub>2</sub>SMP sequential auscultation method was used in the experimental group. Under the premise of fulfilling the needs of the teaching materials and curriculum, the six main elements of sequential cardiac auscultation (heart sounds, heart rate, heart rhythm, extra-heart sounds, murmurs, and pericardial friction sounds) were successively emphasized and explained. In addition, the specific details of the SR<sub>2</sub>SMP sequential auscultation method were continuously reinforced during classroom lectures, simulation practice, and ward rounds (Figure 1).



**Figure 1:** Sequential cardiac auscultation method is conducted according to the sequence of heart sounds, heart rate, heart rhythm, extra heart sounds, murmurs, and pericardial friction sounds (SR2SMP).

The traditional teaching method was used in the control group. Content relevant to cardiac auscultation was explained according to the needs of the teaching materials and curriculum. Although content relating to heart rate, heart rhythm, heart sounds, extra-heart sounds, murmurs, and pericardial friction sounds was delivered, the teaching sequence was not emphasized during the explanation process. Each teaching unit was mutually independent of the others.

### 2.5. Evaluation methods

The two groups were tested on their knowledge of the educational content at different stages. Evaluation was conducted through cardiac auscultation using a simulator. The students were primarily evaluated on typical aspects of cardiac auscultation and their ability to recognize, distinguish, analyze, and identify various heart sounds. Each test was composed of 20 standard cardiac auscultation problems. Some problems included a brief summary of a medical history and required the student to make a comprehensive, accurate diagnosis by combining clinical data with cardiac auscultation. There was a 1.5-min time limit for each problem, equaling a total examination time of 30 min. The problems in the tests for both groups

were completely consistent. Evaluation time points were designated as immediately after teaching intervention, 1 month after teaching intervention (T2), and 6 months after teaching intervention (T3). The material at each of the three evaluations was consistent but the order in which knowledge was tested and the specific heart sounds for the same evaluation objective differed. Figure 2 shows the process of simulation teaching and examination of cardiac auscultation. Comparisons and statistical analyses were performed on the evaluation grades of the two groups. Differences in cardiac auscultation ability between the two groups of students were evaluated at different points.

In addition, a self-reported survey was also used to determine the attitude of the students toward the teaching methods used. The survey included 10 items assessed using a 5-point Likert scale, with 1 representing “strongly disagree” and 5 representing “strongly agree”. One item was a subjective free-response question to obtain subjective assessments of and recommendations for the teaching methods used.

### 2.6. Statistical methods

SPSS 17.0 was used for all statistical analysis. Data are shown as the mean  $\pm$  standard deviation or

percentage, with  $\alpha = 0.05$  as the level of statistical significance. Measurement data were first

tested for the normal distribution using the Kolmogorov-Smirnov test and for homoscedasticity using Levene's test for equality of variances. Student's *t*-test was performed after the data passed these tests. The rank-sum test was used for non-

**3. Results**

**3.1. Baseline student characteristics**

A total of 180 students were enrolled in the present study. No student declined to participate. There were no statistically significant differences in

normally distributed data. The independent samples *t*-test was used to compare test results after teaching intervention at different time points and survey results between the two groups. The paired *t*-test was used to compare the test results of each group at different time points.

baseline conditions such as age, male/female ratio, and average grade in the previous year's core classes between the groups (Table 2).

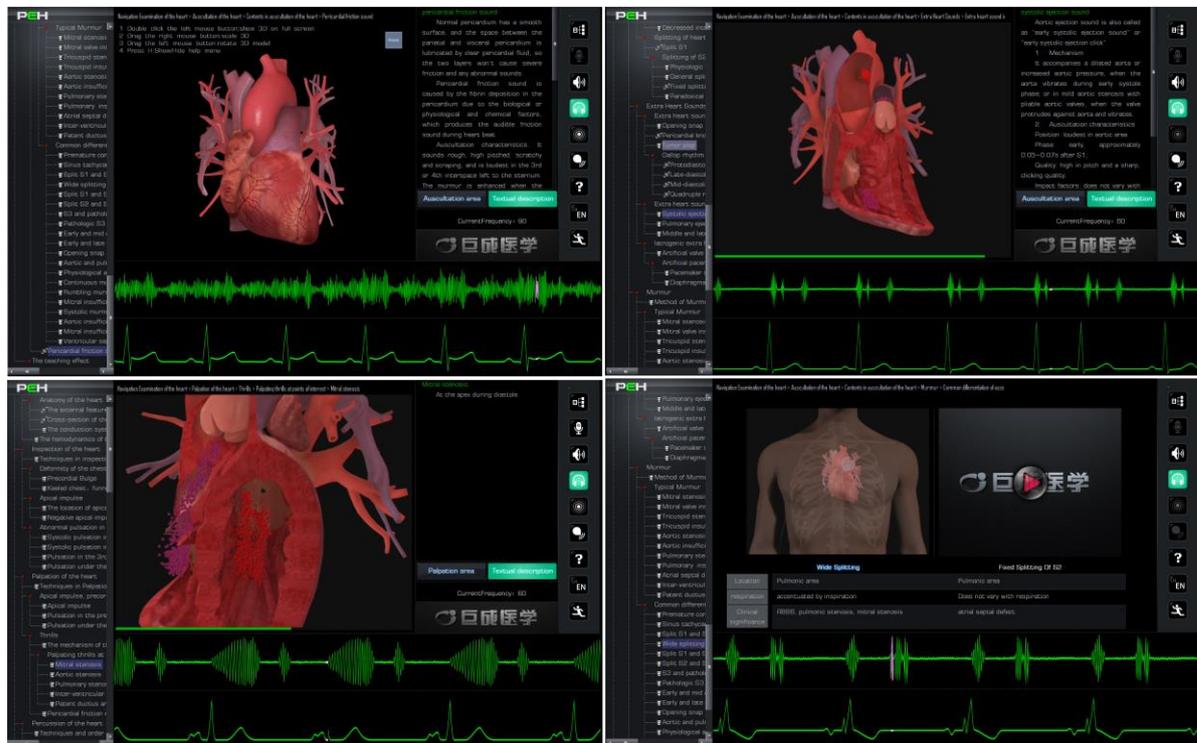


Figure 2: Simulation teaching and examination of cardiac auscultation was conducted by Physical Examination of Heart (PEH) simulator.

Table 2: Baseline student characteristics

Characteristics of baseline	Experimental group (n=90)	Control group(n=90)	t or $\chi^2$ value	P value	95% Confidence Interval of the Difference
Age	20.17 ± 0.70	20.23 ± 0.63	-0.389	0.699	(-0.410,0.276)
Gender(male/female)	42/48	43/47	0.069	0.793	
Anatomy	76.10 ± 10.55	79.23 ± 11.18	-1.116	0.269	(-8.752,2.485)
Pathophysiology	80.77 ± 9.57	79.30 ± 11.41	0.539	0.592	(-3.977,6.91)
Pathology	77.30 ± 10.02	77.23 ± 10.46	0.025	0.980	(-5.228,5.361)

### 3.2. Comparison of evaluation grades between the two groups

Cardiac auscultation testing revealed no significant difference in evaluation grade between the two groups immediately after teaching intervention. Later, the grades of the two groups decreased to some extent. However, the decreasing trend in the experimental group was more gradual than that in the control group (Figure 3). At 1 month and 6 months after teaching intervention, the grades of the students in the experimental group were significantly better than those of students in the control group (T2:  $16.80 \pm 1.79$  vs.  $15.53 \pm 2.19$ ,  $t = 2.451$ ,  $p = 0.017$ ; T3:  $15.40 \pm 2.22$  vs.  $13.37 \pm 3.09$ ,  $t = 2.926$ ,  $p = 0.005$ ) (Table 3).

### 3.3. Survey of student feedback about cardiac auscultation teaching methods

A total of 180 answered surveys were received.

The response rate was 100%. The results of the survey showed that students were generally satisfied with the sequential cardiac auscultation teaching method and at the same time approved of the traditional teaching method; the difference between the two groups was not statistically significant (5 [4,5] vs. 5 [4,5],  $t = 1.881$ ,  $p = 0.066$ ). For the survey item "This teaching method can enhance the effectiveness and maintenance of my memory, helps my understanding of the methods and procedure, and increases my clinical technique and self-confidence", more students selected "agree or strongly agree" for the sequential auscultation method than for the traditional teaching method. The survey item "I hope this teaching method is promoted in future coursework" also received higher scores among the sequential auscultation group than among the traditional group (5 [4,5] vs. 4 [3,4],  $t = 6.501$ ,  $p = 0.000$ ) (Figure.4, Table 4).

Table 3: The test scores of two groups of students in different learning stages

Scores	Experimental group (n=90)	Control group (n=90)	t value	P value	95% Confidence Interval of the Difference
T1	$17.80 \pm 1.40$	$17.40 \pm 1.63$	1.019	0.312	(-0.386, 1.186)
T2	$16.80 \pm 1.79$	$15.53 \pm 2.19$	2.451	0.017	(0.232, 2.301)
T3	$15.40 \pm 2.22$	$13.37 \pm 3.09$	2.926	0.005	(0.642, 3.424)

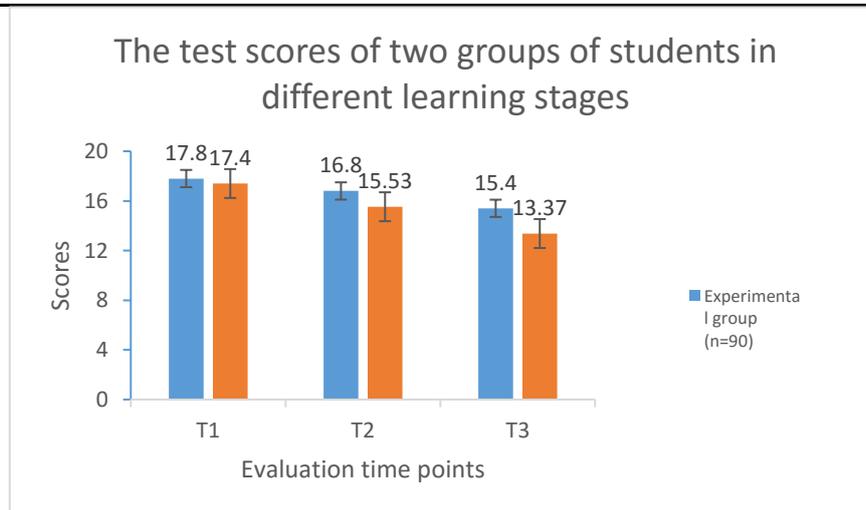


Figure 3: Cardiac auscultation performance trends of two groups. Testing showed that there was no significant difference in evaluation grade between the two groups immediately after teaching intervention (test 1). At 1 month (test 2) and 6 months (test 3) after teaching intervention, the grades of the experimental group were significantly better than those of the control group.

Table 4: Students' feedback on the teaching model

Items	Score (Mean score $\pm$ standard error)		t value	P value	95% Confidence Interval of the Difference
	experimental group (n=90)	control group (n=90)			
The teaching method can stimulate my interest in learning	4.67 $\pm$ 0.55	4.33 $\pm$ 0.80	1.881	0.066	(-0.022,0.689)
The teaching method can enhance the memory effectiveness	4.87 $\pm$ 0.35	3.80 $\pm$ 0.81	6.667	0.000	(0.743,1.390)
The teaching method can increase the permanence of my memory	4.97 $\pm$ 0.18	3.57 $\pm$ 0.82	9.158	0.000	(1.089,1.711)
The teaching method can help my understanding	4.43 $\pm$ 0.77	4.03 $\pm$ 0.72	2.075	0.042	(0.014,0.786)
The teaching method can improve my study efficiency	4.47 $\pm$ 0.68	4.30 $\pm$ 0.75	0.901	0.371	(-0.204,0.537)
The teaching method is benefit for me to master the operation rules, methods and procedures	4.73 $\pm$ 0.45	4.30 $\pm$ 0.47	3.664	0.001	(0.197,0.670)
The teaching method can increases my clinical technique and self-confidence	4.93 $\pm$ 0.25	4.03 $\pm$ 0.85	5.555	0.000	(0.571,1.229)
The teaching method is more active for the classroom atmosphere	4.30 $\pm$ 0.60	4.13 $\pm$ 0.73	0.968	0.337	(-0.178,0.511)
How satisfied am I with this method of cardiac auscultation teaching?	4.67 $\pm$ 0.48	4.33 $\pm$ 0.48	2.693	0.009	(0.086,0.581)
I hope this teaching method is promoted in future coursework	4.73 $\pm$ 0.45	3.77 $\pm$ 0.68	6.501	0.000	(0.668,1.265)

#### 4. Discussion

##### 4.1. Main findings

In 1885, the German psychologist Ebbinghaus discovered and formulated the forgetting curve through experimentation and believed that meaningful memorization of material was retained more rapidly than learned memorization material that was not significant. Compared to insignificant and irregular memorization material, the human brain memorizes rhythmic material and mnemonics more rapidly and retains them for a longer period of time (Murre JM & Dros J., 2015). The SR<sub>2</sub>SMP sequential cardiac auscultation method simplifies many complicated points of knowledge into a text description through highly generalized knowledge

and summaries. The method reduces the absolute amount of material to be memorized and divides the material into modules for memorization. Increasing the density of material can reduce the burden on the brain, increasing memory consolidation and retention and guiding regular operation. As shown by the results of the present study, although the difference in test grades immediately after intervention was not statistically significant between the two groups, testing at 1

month and 6 months after intervention showed that the SR<sub>2</sub>SMP sequential cardiac auscultation teaching method resulted in significantly better test grades.

The teaching of cardiac auscultation is associated with many complications. Because of the widespread development of echocardiography, cardiac angiography, and other medical technologies, clinical physicians have neglected the importance of cardiac auscultation, which in turn has caused awareness of cardiac auscultation among medical students to weaken (Issenberg S.B. et al., 1999)(Fraser K. et al., 2009)(Butter J. et al., 2010)(Sverdrup Ø. et al., 2010)(Fraser K. et al., 2011)(Norman G. et al., 2012)(McKinney J. et al., 2013)(Chen R. et al., 2015). Cardiac anatomy and electrophysiology are complicated, cardiac auscultation material is abstract and difficult to understand, and the vast majority of students believe that cardiac auscultation is very difficult, making it easy for them to feel overwhelmed (Chizner M.A., 2008)(Anderson R.H. et al., 2016)(Barrett M.J. et al., 2004)(Collins J.A. et al., 2014)(Voin V. et al., 2017). Because the current medical environment is limited, patients have a strong sense of self-protection, and the incidence of some diseases has decreased significantly, there is a lack of typical clinical cases and patients (Barrett M.J. et al., 2004). The use of multimedia teaching software or electronic simulations to teach auscultation have enhanced the standardization and repeatability of auscultation teaching to some extent, but the learning process is still passive (Fraser K. et al., 2009)(Butter J. et al., 2010)(Sverdrup Ø. et al., 2010)(Fraser K. et al., 2011)(Norman G. et al., 2012)(McKinney J. et al., 2013). A study on cardiac auscultation based on the use of a model showed that even if only the time of simulation-based teaching was increased, the auscultation ability of students could not be improved significantly (Hatala R. et al., 2008). In response to the above problems, the SR<sub>2</sub>SMP sequential cardiac auscultation teaching method was proposed and merged with simulation teaching and bedside teaching with actual patients to allow students to fully understand the specific details of the method. It significantly increased the auscultation skill of students and made their grasp of

location, time course, quality, strength, and clinical significance more comprehensive, systematic, and organized. The grades earned during comprehensive testing of auscultation were also significantly better than those in the control group. This teaching method is attractive to students who have just entered the clinical phase of their education. Students' interest and motivation to practice are intensified through step-by-step practice of the sequential cardiac auscultation method. By breaking down and refining points of knowledge, students can understand key points of knowledge more deeply and comprehensively. Not only does this procedure stimulate students to take initiative in participating in practice and discussion, it also reinforces their awareness of auscultation. In addition, this is a teaching method with clear instructions and procedures, making it easier to grasp and apply and therefore providing a solid foundation for students to become skilled at auscultation and achieve good teaching outcomes.

Feedback questionnaires from students regarding the teaching of auscultation showed that although students were also comfortable with the traditional teaching method, multiple items assessing the SR<sub>2</sub>SMP sequential cardiac auscultation method all received widespread agreement among students, with an average score of over 4 on a Likert 5-point evaluation scale. In addition, for the items "I gained more from this teaching method" and "I hope this teaching method is promoted in future coursework", students in the SR<sub>2</sub>SMP group gave higher scores than those in the traditional group. Student feedback fully affirmed the SR<sub>2</sub>SMP teaching method because its simplicity and practicality facilitated memorization. Even at 1 month and 6 months after the end of training, the students in

the experimental group could still clearly remember the sequence, content, differentiation between heart sounds, heart sounds, and extra heart sounds, and characteristics of murmurs common during cardiac auscultation. Although the

test grades of students in the control group immediately after intervention approached the level of those in the experimental group, memory loss due

to long-term disuse caused test grades at 1 month and 6 months to be far lower than those in the experimental group.

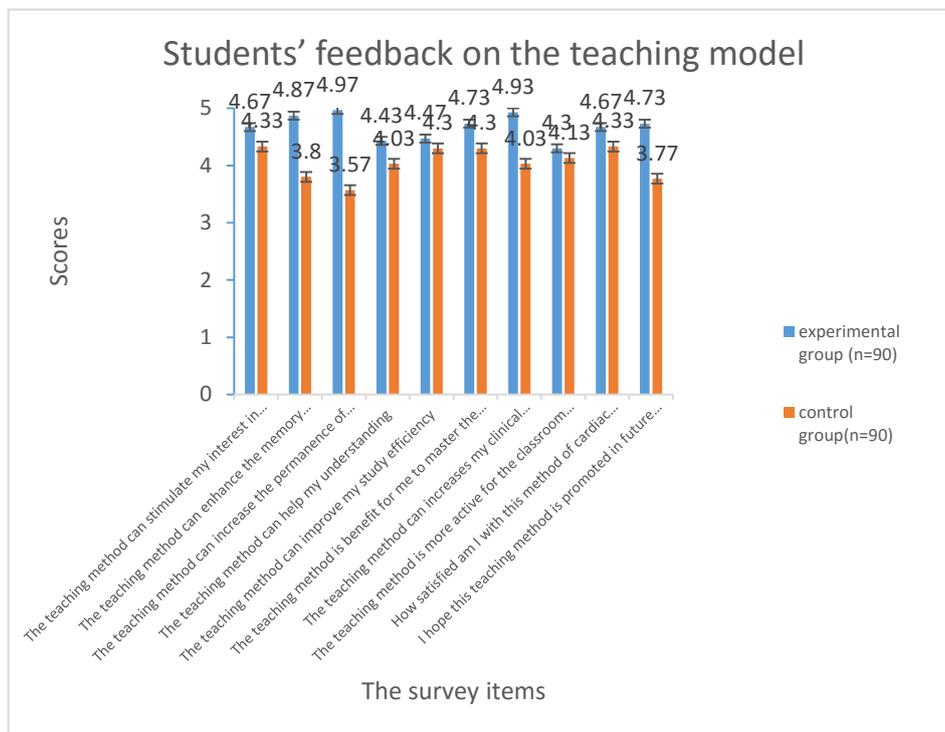


Figure 4: Students' feedback on the teaching model.

#### 4.2. Limitations

Our study had a number of limitations. First, due to the limited teaching hours, therefore, the results should be generalized with caution. Further large-scale and more in-depth studies are needed to verify

#### 5. Conclusions

Many young physicians still take basic physical examination skills very seriously and the results of physical examinations are generally very accurate. However, physicians are becoming increasingly reliant on results obtained using supplementary examinations because of a lack of confidence in their auscultation skills. It should be emphasized that in the early stages of some diseases or due to heterogeneity between patients, differences in supplementary examinations may be very small or lag compared to differences in signs, which can appear more rapidly. The power of clinical medicine lies not only in technological solutions, which are improving rapidly, but also in the traditional stethoscope skills of physicians, which allow the

these results. Second, balance within groups was not ensured, which might have resulted in in-group differences regarding experiences and achievements during the teaching process.

early signs of disease to be detected with limited distraction by complex machines. For cardiac auscultation to be effective, the full confidence of the clinical physician is required and the physician must trust his/her own ears (Tavel M.E. et al., 2006)(Tavel M.E. et al., 2010). As mentioned above, cardiac auscultation is a necessary skill for

physicians. Although the procedure involves many different skills and is associated with a high level of difficulty, the introduction of the SR2SMP sequential cardiac auscultation teaching method can effectively solve these problems, allowing young medical students to simply and effectively grasp the knowledge and methods of cardiac auscultation. The method provides the student with even more

familiarity and confidence when they eventually deal with real patients.

### **Contributors**

ZR participated in the design of the study, analysis of the results, discussion, and writing of the manuscript. ZZF contributed to the writing of the Results and Discussion section of the manuscript. JY contributed to the study design, selection of students, and critical review and writing of the manuscript. LFH participated in the analysis and interpretation of the data. ZZF also participated in the design of the study and collected student data. CF and ZJ participated in the analysis and interpretation of the data, and also in data collection and result management. ZC, who was the program director, conceived the study, participated in its design and coordination, and helped draft the manuscript. All authors read and approved the final version of the manuscript.

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### **Competing interests**

No competing interests exist.

### **Ethical approval**

Experimental protocols and written informed participants' consent were approved by the Human Subject Ethics Committee of West China Medical Centre, Sichuan University (Ethics consent No. HSEC20151748-11554) according to the requirements of the Chinese Prevention of Cruelty to Human Subjects Act and the Code of Practice for the Care and Use of Human Subjects for Scientific Purposes. All students and teachers participated in this study clearly understood each step of the study and signed an informed consent form before participating in our study.

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