Abo-Hashish *et al.*
Egyptian Pediatric Association Gazette (2023) 71:61
https://doi.org/10.1186/s43054-023-00208-4

Egyptian Pediatric Association Gazette

RESEARCH

Open Access

Tenascin-C: as a diagnostic biomarker for rheumatic heart disease

Maha M. A. Abo-Hashish[1*], Azza Mohamed Ahmed[1], Mohammad Ali Hegazi[2], Naglaa Abdel Rahman Mosaad[2], Mona Hammed Ibrahim[3] and Nagwan Yehia Abdel Salam[1]

Abstract

Background Rheumatic fever is a long-term inflammatory disease that can happen if group A beta-hemolytic streptococci bacteria are not treated well enough. Rheumatic fever is recognized globally as the leading cause of heart disease in the pediatric population. This disease destroys the heart muscle, progressively deteriorating its structure and impairing the function of its valves over time.

Aim The aim of this study is to determine the role of serum tenascin-C in the diagnosis of acute rheumatic fever and chronic rheumatic heart disease.

Methods This case–control study involved a group of 60 Egyptian children. Among them, 20 were diagnosed with acute rheumatic fever, identified using the updated Jones criteria from 2015. Another 20 children, who were suffering from chronic rheumatic heart disease, were also act as a part of the study. The remaining 20 participants, healthy children carefully matched in age and sex, served as the control group.

Results Serum tenascin-C level was significantly increased in acute rheumatic fever (ARF) and highly significantly increased in chronic rheumatic heart disease (CRHD) groups when compared with control group ($P=0.04$, 0.01), respectively. There were highly significant difference between and within the studied groups regarding the mean of serum tenascin-C. Serum tenascin-C mean of ARF, CRHD, and control was 4.82 ± 18.7, 5.46 ± 1.6, and 3.78 ± 2.4, respectively, $P=0.02$. Level of serum tenascin-C was lower in cases with severe mitral valve insufficiency. No significant link was found between the level of serum tenascin-C and C-reactive protein (CRP), ESR, and ASO titer, with a *P*-value greater than 0.5. ROC curve for serum tenascin-C in ARF patients was area under the curve = 0.682 ($P=0.05$) with optimal serum tenascin-C cut-off point (> 3.76 ng/ml); ROC curve for serum tenascin-C in CRHD patients was $AUC=0.73$ ($P=0.01$) with cut-off point level (73.76 ng/ml).

Conclusion Patients with ARF and CRHD have increased level of serum tenascin-C. Serum tenancin-C is superior in the diagnosis of ARF in comparison to CRP, ESR, and ASOT. Tenascin-C level can be used as a diagnostic marker for ARF and CRHD.

Keywords Serum tenascin-C, Acute rheumatic fever, Chronic rheumatic heart disease

Background

Rheumatic fever, a persistent inflammatory condition, can be a severe complication from insufficient treatment of group A beta-hemolytic streptococci bacteria. This disease progressively damages the heart valves. It is the leading cause of heart disease among children worldwide [1]. There have been three major groups of ideas, which have been advanced during the past five

*Correspondence:
Maha M. A. Abo-Hashish
mahaabohashish66@gmail.com
[1] Pediatric Department, National Research Centre, Giza 12622, Egypt
[2] Pediatric Department, Faculty of Medicine, Cairo University, Cairo, Egypt
[3] Clinical Pathology Department, National Research Centre, Giza 12622, Egypt

decades to clarify streptococcal etiology for rheumatic fever. These include the following: direct infection, impacts of streptococcal toxin, and, most plausibly, the concept of antigenic mimicry in combination with an abnormal immune response [2].

Duckett Jones developed the diagnostic criteria (major and minor) for acute rheumatic fever in 1944, and they have since been revised in 1965, 1984, 1992, 2002, and 2015 [3]. For a disease diagnosis, it is necessary to have either two major criteria or one major and two minor criteria present, along with evidence of a recent streptococcal infection [3].

The majority of heart valves consist of various components such as the extracellular matrix (ECM), smooth muscle, fibroblasts, and endothelial cells [4]. Tenascin-C (TnC), a hexameric glycoprotein, serves a multitude of functions. It is a crucial part of the ECM, produced by interstitial fibroblasts, and its levels are notably increased in inflammatory diseases [5]. During embryo development, only low levels of tenascin-c are present for a short time. In places where wounds heal, cancer grows, and myocarditis, higher levels of TnC expression have been found [6]. The goal of this research is to find out if serum tenascin-C plays a role in the diagnosis of acute rheumatic fever and chronic rheumatic heart disease.

Patients and methods

Our present study was carried out in New Children Hospital-Cairo University and National Research Centre, Egypt. The study is a case–control one in which all participants were 60 Egyptian children and adolescent of both sex with their ages ranging between 3 and 18 years old stratified into 3 groups:

- Group 1: In twenty patients with acute rheumatic fever, the diagnosis was based on Jones criteria update (2015) [3].
- Group 2: Twenty patients with chronic rheumatic heart disease
- Group 3: Twenty age- and sex-matched healthy children and adolescent who served as a control group

All patients were recruited from rheumatic fever outpatient clinic, inpatient ward of Children Hospital-Cairo University, and outpatient pediatrics clinic of the National Research Center. An informed consent was approved and signed by parents of all our study participants according to guidelines of Ethical Committees of Children Hospital, Cairo University, and National Research Centre, Egypt, approval no. 15077.

Patients eligible for the study showed the following:

Inclusion criteria

1. Age of patients ranged between 3 and 18 years old
2. Diagnosis of acute rheumatic fever is based on Jones criteria updated 1992.
3. Chronic rheumatic heart disease group

Exclusion criteria

Patients with other chronic systemic diseases.

All participants were subjected to the following:

1. Full history including demographic data, age of disease onset, disease duration, clinical manifestations of the disease, and treatment given to each patient
2. Full physical examination specific for cardiac aspect, with measurement of the blood pressure and heart rate
3. Laboratory investigations are as follows:

    a. Complete blood count by Sysmex.
    b. Erythrocyte sedimentation rate.
    c. C-reactive protein by latex.
    d. Antistreptolysin O titer by latex.
    e. Serum tenascin-C by ELISA technique.

4. Twelve lead electrocardiogram and full echocardiographic (ECG) study.

Echocardiographic examination

Echocardiographic studies were performed according to methods established by the American Society of Echocardiography [7] with 2.5–3.8 MHz transducer frequencies using a Vivid 5 GE ultrasonic imager. Pathological valvular insufficiency was evaluated according to World Health Organization criteria [8]. Doppler methods, including assessment of regurgitant jet characteristics, were used in the assessment of the severity of valvular regurgitation [9].

Methodology

Six milliliters of peripheral venous blood samples was withdrawn from each participant under complete aseptic conditions. Portion of blood samples were anticoagulated with EDTA for performing CBC and ESR. Then sera were left to clot at room temperature and centrifugated. Sera were aliquated and uniquely labeled for

assessment of C-reactive protein and antistreptolysin O titer; part of sera were stored at − 20 °C for further assessment by ELISA. Serum tenascin-C was assayed using MyBioSource ELISA Kit, cat no. MBS177649 [10].

Statistical analysis

Statistical Package for Social Science (SPSS) program version 16 was used for analysis of data. Data were summarized as mean ± SD. Student's *t*-test for quantitative independent variables was used for analysis of difference between two groups. Comparison of multiple groups was done using one-way ANOVA. Correlation between quantitative variables was done using Pearson's bivariate test. In all test, $P < 0.05$ was considered statistically significant. Receiver operator characteristic curves (ROC curves) were drawn for ARF and CRHD groups.

Results

The study encompassed 20 patients diagnosed with acute rheumatic fever (ARF), including 12 males and 8 females, with fifteen cases showing isolated arteritis and five cases presenting with carditis. Furthermore, 20 patients (15 males, 5 females) were dealing with chronic rheumatic heart disease (CRHD). A control group was also part of the study, consisting of 20 individuals (14 males, 6 females).

Comparison between values of serum tenascin-C (ng/ml) in the studied groups were demonstrated in Fig. 1. Serum tenascin-C level was significantly increased in ARF and GRHD groups when compared with control group ($P = 0.04$, 0.01), respectively. The mean levels of serum tenascin-C in ARF, CRHD, and control were 7.16 ± 9.7, 5.46 ± 1.6, and 3.78 ± 2.4, respectively (Fig. 1).

Figure 2 clarifies the level serum tenascin-C in the CRHD regarding the severity of mitral regurge (MR) according to echo cardiac valve affection. The mean levels of serum tenascin-C were 6.36 ± 1.5 in patients with mild MR ($n = 10$), 5.84 ± 1.04 in patients with moderate MR ($n = 5$), and 3.88 ± 0.86 in patients with severe MR ($n = 5$).

Correlation between serum tenascin-C and CRP, ESR, and ASOT in ARF is shown in Table 1. There was no significant correlation between scrum tenascin-C level and CRP, ESR, and ASOT ($P > 0.5$). ROC curve for serum tenascin-C in ARF patients was area under the curve = 0.682 ($P = 0.05$) with optimal scrum tenascin-C cut-off point (> 3.76 ng/ml), sensitivity 75%, and specificity 65%. Positive predictive valve is 68.2% and negative predictive valve 72.2% (Fig. 3).

ROC curve for serum tenascin-C in CRHD patients was $AUC = 0.73$ ($P = 0.01$) with cutoff point level 73.76 ng/ml, sensitivity 90%, specificity 65%, positive vindictive valve 72%, and negative predictive valve 86.7% (Fig. 4), i.e., ROC curve for serum tenascin-C in ARF patients and CRHD patients indicates that serum tenascin-C is useful test for diagnosis of ARF and CRHD.

Discussion

Tenascin-C is believed to have a significant role in the immune system and the development of inflammatory diseases. In this specific study, it was observed that the levels of serum tenascin-C were notably elevated in cases of ARF and CRHD, with statistical significance marked by a *P*-value of 0.04 and 0.01, respectively.

In a study parallel to ours, Golledge et al. [5]. discovered that patients with rheumatic heart disease had significantly elevated levels of serum tenascin-C when

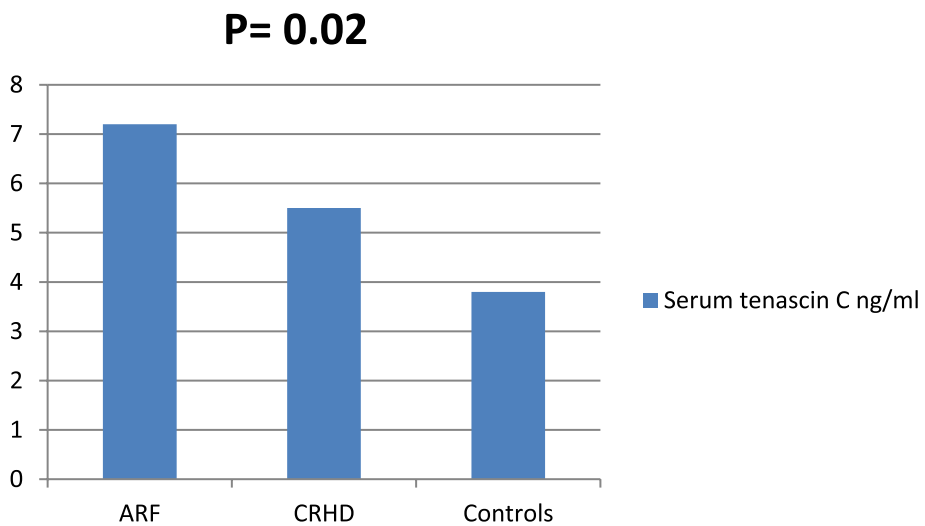

Fig. 1 Comparison between values of tenascin-C (ng/ml) in the studied groups

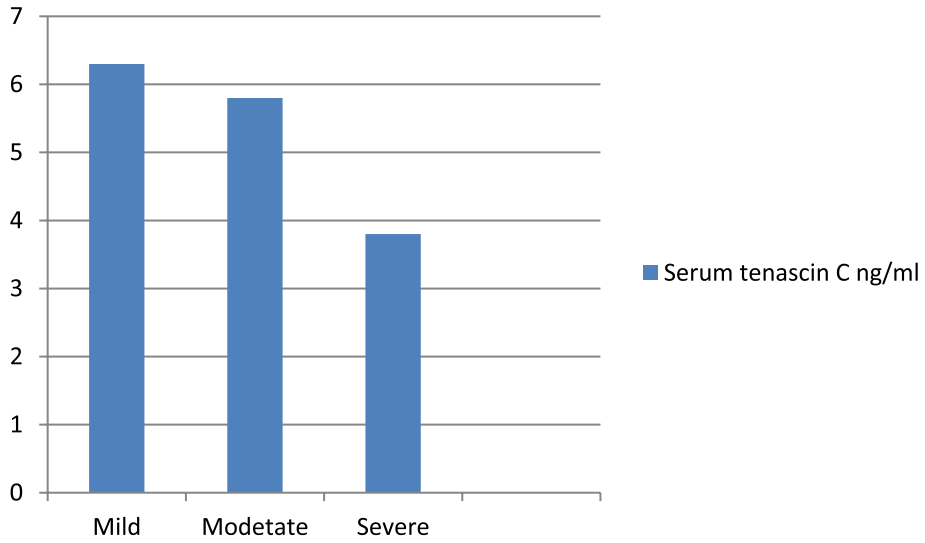

Fig. 2 Comparison between values of tenascin-C (ng/ml) in chronic group according to echo mitral valve affection

Table 1 Correlation between serum tenascin-C and CRP, ESR, and ASOT in acute rheumatic fever group ($n=20$)

Items	Spearman's correlation (R)	*p*-value
CRP	0.181	0.43
ESR	0.224	0.34
ASOT	0.27	0.23

$P>0.05$, not significant

$P<0.05$, significant

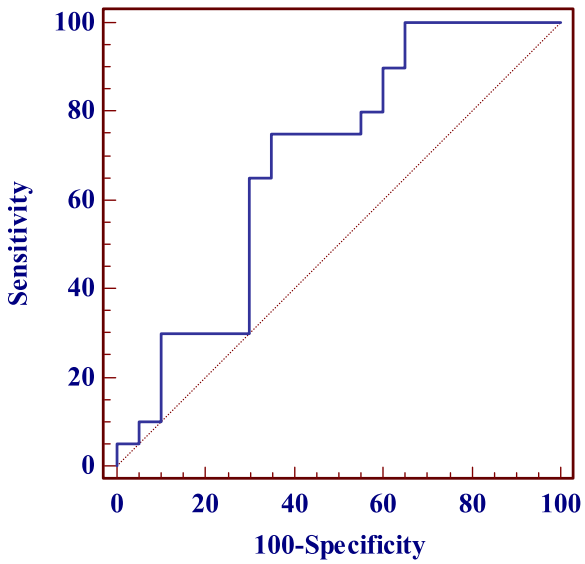

Fig. 3 ROC curve of serum tenascin-C in acute rheumatic fever group

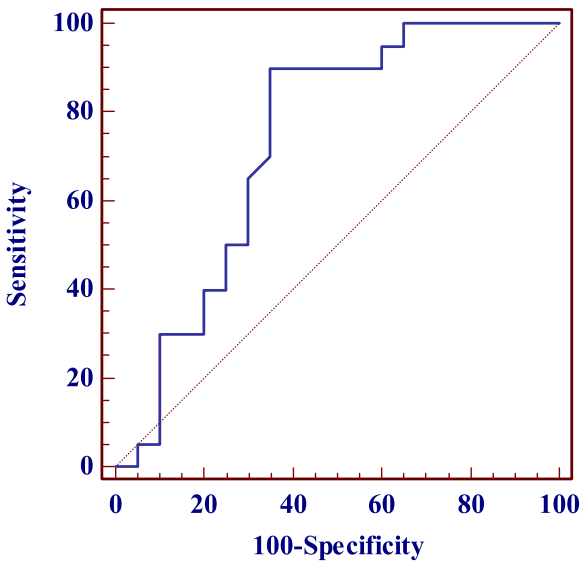

Fig. 4 ROC curve of serum tenascin-C in chronic rheumatic heart disease group

compared to control subjects ($P<0.05$). This observation was further corroborated by Karatas and colleagues [11], who found a notable increase in tenascin-C levels in patients with ARF and CRHD compared to their control group ($P<0.05$). They even suggested that the extent of the rise in serum TnC levels can be used as a dependable predictor for ARF.

Davutoglu et al. [12]. explored the progression of rheumatic carditis in their study, emphasizing its association with immunological and biochemical disturbances. They pointed out that the development of rheumatic heart

valve disease in carditis is a complex process, influenced by genetic factors that make an individual more susceptible to autoimmune reactions [13]. The link between heart valve damage, the development of valve fibrosis, and immune-inflammatory response is well-recognized [14]. Degenerative valve lesions exhibit several characteristics typical of active pathological processes, such as chronic inflammation, lipoprotein deposition, and active calcification [15]. Moreover, the role of expressional tenascin-C in the development of fibrosis during tissue repair has also been established [14].

In this study, it was found that serum tenascin-C levels were lower in children with severe mitral valve insufficiency compared to those with mild and moderate insufficiencies, a finding that aligns with results reported by Karatas et al. Vollmer et al. [16] suggested that in the absence of tenascin-C, neutrophil invasion remains unaffected, but monocyte chemotaxis and macrophage migration are inhibited, potentially leading to insufficient tissue repair. This was supported by a study on tenascin-deficient mice, which showed less neointimal proliferation, indicating that impaired tenascin-C production could result in failed tissue repair.

Karatas et al. [11] also found in their chronic rheumatic heart disease (CRHD) group that valve replacement was performed in three patients with very low tenascin-C levels in the chronic phase. This suggests that low serum tenascin-C levels could be a useful predictor for surgical treatment. They concluded that the immune system-mediated tissue repair process is completed appropriately in rheumatic carditis in cases with normal tenascin-C levels, and that serum tenascin-C could be used to predict the disease's prognosis.

In the present study, no significant correlation was found between serum tenascin-C and CRP, ESR, and ASOT in ARF. This is in agreement with data from Karatas et al. [11], who reported no statistically significant difference between patients with ARF in terms of serum tenascin-C and other acute phase reactants (CRP, ASOT, ESR). This suggests that serum tenascin-C is more sensitive in diagnosing ARF than other acute phase reactants.

In terms of ARF, the area under the curve (AUC) in our study was 0.682 ($P=0.05$) with a cutoff point of 73.76 ng/ml, a sensitivity of 75%, a specificity of 65%, a positive predictive value of 68.2%, and a negative predictive value of 72.2%. For CRHD, the AUC was 0.73 ($P=0.04$) with a cutoff point of > 3.76 ng/ml, a sensitivity of 90%, a specificity of 65%, a positive predictive value of 72%, and a negative predictive value of 86.7%. These results suggest that serum tenascin-C is a very useful test for identifying ARF and CRHD.

Karatas et al. [11] reported similar findings in a study conducted on 25 patients with ARF and 25 patients with CRHD. For ARF, the ROC analysis AUC was 0.953, with a cutoff point of 2.08 ng/ml, a sensitivity of 93.3%, and a specificity of 95%. For CRHD, the AUC was 0.92 ($P<0.001$) with a cutoff point of 1.56 mg/ml, a sensitivity of 83.3%, and a specificity of 85%. These findings support the use of serum tenascin-C as a new biomarker for the diagnosis of ARF and CRHD.

Conclusion

Patients with acute rheumatic fever (ARF) and chronic rheumatic heart disease (CRHD) exhibit elevated levels of serum tenascin-C. Serum tenascin-C may be more helpful for diagnosing ARF when compared to CRP, ESR, and ASOT. Therefore, serum tenascin-C can be utilized as a marker for diagnosing ARF and CRHD. Future prospective studies which include larger population are required to ascertain if serum tenascin-C can serve as a reliable biochemical test for predicting the prognosis of CRHD.

Abbreviations

RHVD	The association between rheumatic heart valvular disease
TnC	Tenascin-C
NYHA	New York Heart Association
ECM	Extracellular matrix
CRHD	Chronic rheumatic heart disease
ASOT	Antistreptolysin O titer
ECM	Extracellular matrix
PMBV	Percutaneous mitral balloon valvotomy
CRP	C-reactive protein
CBC	Complete blood count
ESR	Erythrocyte sedimentation rate

Acknowledgements

Not applicable.

Authors' contributions

MMAA put the design of the work, wrote and revised methodology, wrote and revised the article, and supervised the study. AMA supervised the study and revised the article. MAH supervised the study and revised the article. NAM supervised the study and revised the article. MHI, methodology, validation, and data curation. NYA shared in the analysis and interpretation of the data, wrote and revised the methodology, revised the article, and supervised the study. All authors have read and approved the manuscript in its final form. The authors certify that the manuscript is original and has not been published before, has been seen and approved by all authors involved, and is neither being published in any other peer-reviewed journal nor being considered for publication elsewhere. The article contains nothing that is unlawful, libelous, or which would, if published, constitute a breach of contract or of confidence or of commitment given to secrecy. The authors are responsible for all parts of the work.

Funding

None.

Availability of data and materials

The data used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate
An informed consent was approved and signed by parents of all our study participants according to guidelines of Ethical Committees of Children Hospital, Cairo University, and National Research Centre, Egypt, approval no. 15077.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

Received: 20 July 2023 Accepted: 18 August 2023
Published online: 06 December 2023

References

1. Chen RS, Bivens MJ, Grossman SA (2011) Diagnosis and management of valvular heart disease in emergency medicine. Emerg Med Clin North Am 29(4):801–810
2. Kaplan EL (2005) Pathogenesis of acute rheumatic fever and rheumatic heart disease: evasive after half a century of clinical, epidemiological, and laboratory investigation. Heart 91:3–4
3. Gewityz MH, Baltimore RS, Tani LY, Sable CA, Shulman ST, Taulbert KA (2015) revision of Jones criteria for the diagnosis of acute. Rheumatic fever in the era of Doppler echo cardiography a scientific statement from the American Heart Association. Circulation 131:1806–1818
4. Fan H, Liu W, Yan P (1996) Pathological observation and immunohistochemistry study of type I, III, IV collagen in mitral valve and cardiac interstitium of rheumatic disease. Zhonghua Yi Xue Za Zhi 76(3):183–186
5. Golledge J, Clancy P, Maguire J, Lincz L, Koblar S (2011) The role of tenascin C in cardiovascular disease. Cardiovasc Res 92(1):19–28
6. Imanaka-Yoshida K. (2021) Tenascin-C in Heart Diseases—The Role of Inflammation. Int J Mol Sci 22:5828
7. Guilherme L, Cury P, Demarchi LMF, Coelho V, Abel L, Lopez AP et al (2004) Rheumatic heart disease: proinflammatory cytokines play a role in the progression and maintenance of valvular lesions. Am J Pathol 165(5):1583–1591
8. Guilherme L, Faé K, Oshiro SE, Kalil J (2005) Molecular pathogenesis of rheumatic fever and rheumatic heart disease. Expert Rev Mol Med 7(28):1–15
9. Tsukada B, Terasaki F, Shimomura H, Otsuka K, Otsuka K, Katashima T et al (2009) High prevalence of chronic myocarditis in dilated cardiomyopathy referred for left ventriculoplasty: expression of tenascin C as a possible marker for inflammation. Hum Pathol 40(7):1015–1022
10. Khomtchouk BB, Lee YS, Khan ML, Sun P, Mero D, Davidson MH (2022) Targeting the cytoskeleton and extracellular matrix in cardiovascular disease drug discovery. Expert Opin Drug Discov 17:1–18
11. Karatas Z, Baysal T, Alp H, Toker A (2013) a novel biomarker for diagnosis and predicting prognosis of rheumatic carditis? J Trop Pediatr 59(6):476–482
12. Davutoglu V, Celik A, Aksoy M (2005) Contribution of selected serum inflammatory mediators to the progression of chronic rheumatic valve disease Subsequent valve calcification and NYHA functional class. J Heart Valve Dis 14:251–256
13. Toni LV (2008) Rheumatic fever and rheumatic heart disease. In: Allen HD, Driscoll DJ, Shaddy RE, Feltes TF (eds) Moss and Adam's heart disease in infants, children and adolescent's, 7th edn. Lippincott Williams and Wilkins, Philadelphia, pp 1256–1280
14. Lincoln J, Yutzey KE (2011) Molecular and developmental mechanisms of congenital heart valve diseases. Birth Defect Res A Clin Mol Teratol 91:526–534
15. Chen MC, Change JP, Liu WH, Yang CH, Chen CJ, Fang CY (2009) Increased serum oxidative stress in patients with severe mitral regurgitation: a new finding and potential mechanism for atrial enlargement. Clin bio chem 42:943–948
16. Vollmer T, Hinse D, Kleesiek K, Dreier J (2010) Interactions between endocarditis derived Streptococcus gallolyticus subsp. Isolates and human endothelial cells. BMC Microbial 10:78–89

Publisher's Note