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Utility of rapid antigen detection test in group A ß-haemolytic streptococcal pharyngitis at a tertiary hospital, Gombe, North-East, Nigeria

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Abstract: *Background:* Throat swab cultures still remain the gold standard for the confirmation of Group A Streptococcal (GAS) pharyngitis but Rapid Antigen detection Test (RADT) are increasingly becoming popular. Recent studies show RADT can be as sensitive and specific as the throat swab culture. This study aimed to determine the utility of RADT in the diagnosis of GAS pharyngitis in children at the Federal Teaching Hospital Gombe.

Methods: A cross-sectional study that included 324 children aged 3-18 years presenting with sore-throat at the out-patient department, consecutively recruited between April and September 2018. Socio-demographic and clinical findings were document. Throat swab samples were taken for RADT using Encode strep A Rapid antigen test and culture on 5% sheep blood agar. The sensitivity, specificity and positive and negative predictive values of the RADT was determined against throat swab cultures, which is the gold standard.

Results: There were 190 (58.6%) females and Male to Female ratio of 1:1.4. The mean age was 8.3± 3.9 years. Only 125 (38.6%) of the participants were from low social class and 162 (62.3%) are from overcrowded households. Socio-demographic factors have no effect on the performance of the RADT, however, the use of antibiotics within 2 weeks prior to presentation significantly affected the performance of the RADT 1.4% (1/64) compared to 28.1% (73/260) for those without prior antibiotics use $p<0.001$.

GAS was isolated in 73 (28.1%) of the children with pharyngitis. The RADT had sensitivity and specificity of 84.6% and 96.2% respectively and a PPV and NPV of 90.4% and 93.6% respectively.

Conclusion: The RADT is reasonably sensitive and specific and can be used in the diagnosis of GAS pharyngitis in clinics as a substitute for throat swab cultures.

Keywords: Rapid test, streptococcus, Pharyngitis,

Introduction

Globally, an estimated 600 million symptomatic cases of acute pharyngitis occur annually.[1] Acute pharyngitis is one of the most frequent illnesses for which paediatricians and primary care physicians are consulted.[2,3] In the United States, approximately 7.3 million outpatient visits in children are attributable to acute pharyngitis each year.[3,4] The burden of acute pharyngitis in developing countries is over 90% of the global estimates.[1,5,6] In Nigeria, acute pharyngitis was diagnosed in 23.8% of children with upper respiratory infections at an outpatient clinic.[7] β-haemolytic streptococcus was isolated in 48.7% of children with pharyngitis while Group A β-haemolytic Streptococcus (GAS) was the aetiologic agent in 10.5% of cases.[8,9]

Early diagnosis and treatment of GAS pharyngitis are critical in preventing complications.[10] However, GAS pharyngitis is clinically indistinguishable from other causes of pharyngitis making laboratory confirmation necessary.[6,11] Clinical guidelines recommended laboratory confirmation using a rapid antigen detection test (RADT) with follow-up cultures in RADT-negative cases.[11, 13] Other guidelines however, suggested that the highly sensitive newer methods of RADT can be used alone without culture.[14] Cost-effectiveness analysis show the use of RADT alone to be the most cost-effective of any approach involving a laboratory investigation.[14,16] In resource-limited settings, the RADT could be a feasible option for timely diagnosis and treatment of GAS pharyngitis.[15] Rapid Antigen Detection Test has been shown to be cheaper and more cost-effective than

throat swab cultures and some guidelines in developed countries recommended its use alone in the diagnosis of GAS pharyngitis.[11,13,15]

In Nigeria, a majority of physicians have been shown to treat acute pharyngitis empirically with antibiotics.[17] This practice is associated with high cost, increased risk of adverse effect and development of antimicrobial drug resistance.[18,19] A RADT, if validated, may reduce the cost associated with empirical treatment, reduce the emergence of antimicrobial drug resistance and decrease the risk of adverse effects of the used antimicrobial agents.

This study aimed to determine the performance of a RADT as a point of care test compared to the goal standard throat swab cultures in the diagnosis of GAS pharyngitis in children.

Materials and methods

The study was conducted at the General Out-Patient Department (GOPD) of the Federal Teaching Hospital (FTH), Gombe, a 450-bed capacity tertiary health institution located in Gombe Local Government Area, Gombe state, Nigeria. The study population comprised children aged between 3-18 years presenting to the GOPD with a clinical diagnosis of acute pharyngitis. Children less than 3 years were excluded due to the difficulty of taking an optimum throat swab sample and the rarity of GAS pharyngitis in this age group.[6,20]

For the purpose of this study, all patients with complaints of sore throat and/or any of the signs of inflamed Pharynx/ tansils (swelling, erythema and or exudates) were categorized as having clinical pharyngitis.

The study was a cross-sectional descriptive study and the minimum sample size was determined using the Fisher formula for descriptive study, $N = z^2pq/d^2$ Where:N = desired sample size, z = standard normal deviate set at 1.96 which correspond to 95% confidence interval or 5% significance level. P = prevalence of pharyngitis in a study of children at an Acute Respiratory Infection clinic of the Department of Paediatrics, University of Port Harcourt Teaching Hospital, Nigeria = 23.8%.[7] A minimum sample size of 307 was arrived at.

A consecutive non-probability sampling method was used. Participants presenting at the GOPD were consecutively recruited until the desired sample size was achieved.

Inclusion Criteria

1. Children aged between 3-18 years with acute pharyngitis.
2. Parents/caregivers who gave consent for their children to participate in the study
3. Assent from children 7years and above.

Exclusion criteria

- Children with other upper respiratory infections as the primary diagnosis such as:
- Otitis media, Acute Epiglottitis and Peritonsillar Abscess

Ethical clearance was obtained from the Research and Ethics Committee (REC) of the Federal Teaching Hospital Gombe. Ethical approval certificate number: NHREC/25/10/2013.

The study was carried out from April to September 2018. Participants were recruited on all clinic days from 9am to 2pm. The researcher provided detailed information to parents and caregivers about the study and informed them about voluntary participation.

A study Proforma was used to document the date, patient's age, sex, ethnicity and the parent's socioeconomic status using the method of Olusanya *et al*.[21] The duration of illness, types and duration of medications taken before visit and the symptoms and signs were recorded and throat swab samples were collected using a double–tipped swab stick after the throat examination. The throat swab specimens were taken with child sited for older children and supported on the care givers laps for younger children. The child is asked to open their mouths widely as was demonstrated by the examiner. A wooden spatula was inserted to depress the tongue while the swab stick is swiftly inserted and rubbed on the tonsils and pharynx with emphasis on inflamed surfaces. The oral mucosa was avoided to minimize contamination

The Rapid Antigen detection test: Specimen from one of the throat swab sticks was used within 5 minutes for the RADT in the consulting room. The RADT kit used was Encode Rapid Strep A Test kit model-ISA-502 (Guangdong, China Mainland). The test cassettes were removed from the sealed foil pouch and kept horizontally on the table. The extraction test tube is kept vertically and 6 full drops of red in coloured reagent A added (approximately 240µl). Then, 4 full drops of Reagent B (approximately 160µl) were added to the extraction test tube. The addition of reagent B to reagent A changes the colour from red to yellow.

Immediately, the throat swab stick was inserted into the extraction test tube of the yellow solution, the stick agitated and held in the tube for 1 minute and withdrawn.

Three full drops of the solution in the extraction tube were then added into the sample well of the test cassette and the timer started. The appearance of the red coloured line(s) was observed and results read within 5 minutes. There were two indicator lines, one for control and the second for the streptococcus positivity. The appearance of two lines was recorded as positive, only control line means negative and absence of any line is repeated.

The Throat swab culture: The second throat swab stick was used to inoculate a 5% sheep's blood agar plate by gently rolling the swab over an area of 3x2cm at

the edge of the plate.[20] The plates were then placed into a carbon dioxide (CO_2) rich environment provided by a sealed glass jar (candle jar method). All inoculated plates were then transported to the microbiology laboratory within one hour of inoculation.

At the microbiology laboratory, the samples were removed from the candle jar and three to four sets of streaks were made on the agar plates from the initial inoculum.[20] A bacitracin disk (0.04 U) was placed in the area of the primary inoculum and the plates were incubated at 35–37°C under CO2 rich environment. Each plate was checked for growth at 24 and 48 hours. Group A β-haemolytic streptococci were identified morphologically as beta haemolytic colonies characterized by circular, glistening colonies measuring 0.5-2.0 mm in diameter with a zone of complete haemolysis 2-4 times the size of the colonies.[20] Colonies sensitive to bacitracin (a clear zone of inhibition around the disc) were classified as GAS positive.[20] Culture plates that show no growth and those that grow any other organisms were reported as GAS negative cultures.

Data generated were processed and analysed using the IBM SPSS statistical software version 23(CDC, Atlanta Georgia USA 2016). The performance of the RADT (sensitivity, specificity and predictive values) was determined against throat swab cultures using a 2X2 contingency table.

Results

There were 190 (58.6%) females with a male to female ratio of 1:1.4. The mean age of the study population was 8.3± 3.9 years. Participants aged 3-6 years constituted the majority 137 (42.3%), while 15-18years age group was the lowest 25 (7.7%). About 125 (38.6%) of the participants were from low socio-economic class, 129 (39.8%) were from households with 6-8 persons. Table 1.

Table 1: Socio-demographic characteristics of study participants

Socio-demographic characteristics	Frequency (%) N=324
Age Groups	
3-6years	137 (42.3)
7-10years	97 (29.9)
11-14years	65 (20.1)
15-18years	25 (7.7)
Gender	
Male	134 (41.4)
Female	190 (58.6)
Socio-economic level	
High	111 (34.2)
Middle	88 (27.2)
Low	125 (38.6)
Household size (number of persons)	
3- 5	80 (24.7)
6-8	129 (39.8)
>9	115 (35.5)

Performance of RADT against throat swab culture

The performance of the RADT was determined against the gold standard throat swab cultures were shown in Table 2. The overall prevalence of GAS pharyngitis using the RADT was 22.8% (74/324). Exposure to antibiotics within 2 weeks prior to presentation, found in 64 (19.8%) of participants, was shown to have a statistically significant effect on the prevalence of GAS positive RADT results, 1.4% (1/64) compared to 28.1% (73/260) for those without prior antibiotics use $p<0.001$. These patients were excluded from further analysis. No statistically significant effect of socio-demographic factors; age, gender, socio-economic status and overcrowding on the performance of the RADT, see Table 3.

The sensitivity, specificity, PPV and NPV were presented in Table IV. The sensitivity, specificity, PPV and NPV of the RADT analyzed among the 260 participants that had no antibiotic exposure prior to presentation were 84.6%, 96.2%, 90.4% and 93.6% respectively. The sensitivity of the RADT was significantly affected by prior antibiotic use (0% compared to 84.6% in patients without prior antibiotics), $p = <0.001$. However, there was no statistically significant difference in the specificity of the RADT between the overall study group, participants with and those without prior antibiotic use (96.7%, 98.3%and 96.2% respectively) Fisher's Exact p= 0.906.

The performance of the RADT was also significantly affected by the clinical likelihood or probability of GAS pharyngitis as assessed by a Clinical Prediction Rule (CPR) $p<0.001$. The CPR scores patients on the clinical likelihood of having GAS pharyngitis on a scale of 0-5 with 0 been least likely and 5 been most likely.[22] The sensitivity of the RADT increased as the CPR score increase from 0-5 as shown in Figure1

Table 2: Performance of RADT against throat swab cultures in the diagnosis of GAS pharyngitis

	Throat swab culture results		
	Positive	Negative	Total
Rapid Antigen test			
Positive	66	7	73
Negative	12	175	187
Total	78	182	260

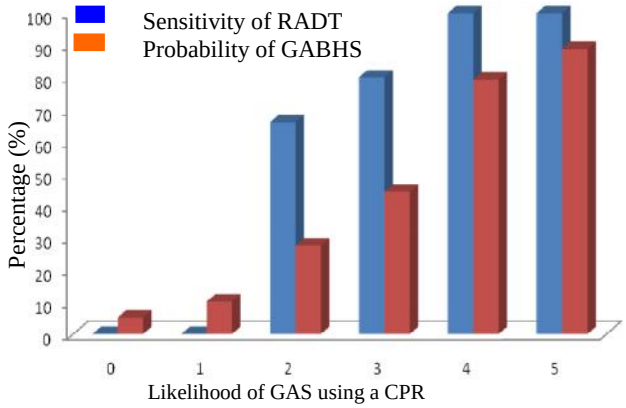
Table 4: Prior antibiotic use and the performance measures of RADT

RADT	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Antibiotic Use				
Prior Antibiotics	0.0	98.3	0.0	90.5
No Antibiotics	84.6	96.2	90.4	93.6
Overall	78.6	96.7	89.2	92.8

Table 3: Effect of Socio-demographic factors on the Performance of the RADT

	RADT (+)	RADT(-)	Total	
Variables	N=73(%)	N=187(%)	N=260(%)	χ2 (P-values)
Age Groups				3.241 (0.072)
3-6years	35(47.9)	87 (46.5)	122 (100)	
7-10years	26 (35.6)	63(33.7)	89 (100)	
11-14years	10 (13.7)	24 (12.8)	34 (100)	
15-18years	2 (2.7)	13 (7.0)	15 (100)	
Gender				1.724(0.632)
Male	22 (30.1)	79(42.2)	101 (100)	
Female	51 (69.9)	108 (57.8)	159 (100)	
Overcrowding				3.478 (0.324)
Yes	29 (39.7)	65 (34.8)	69 (100)	
No	44 (60.3)	122 (65.2)	166 (100)	
Socio-economic class				0.624 (0.732)
Low	22(30.1)	66 (35.3)	88 (100)	
Middle	22 (30.1)	52 (27.8)	74 (100)	
High	29 (39.7)	69 (36.9)	98 (100)	

Fig 1: Relationship between the sensitivity of RADT and clinical likelihood of GAS pharyngitis using a Clinical Prediction Rule (CPR)



Discussion

The prevalence of group A β-hemolyte streptococcal pharyngitis in this study (22.8%) was similar to the 21.6%reported by Engel *et al*[23] in South Africa, 25.0% by Gonsu *et al*[24] in Cameroon, 28.2% by Ether *et al*[25] in Sudan. Although Gonsu *et al*[24] assessed GAS using a RADT, the prevalence was similar to Engel*et al*[23] and Ether *et al*[25] who used the Gold standard throat cultures. All these studies where among children 3-15year which were similar to the age range in this study.

The sensitivity and specificity of the RADT in this study was 84.6% and 96.2% respectively were similar to the 83.3% and 96.0% reported by Gonsu*et al*[24] in Cameroon. Gonsu*et al*[24] used the same type of RADT method used in this study, the enzyme immunoassay. Rimoin *et al*[15] reported the performance of a RADT in four developing countries and the sensitivity ranged from 72.4% to 91.8% and specificity ranged from 85.7% to 96.4%. Although Rimoin *et al*[15] used the more sensitive optical immunoassay method of RADT, the results are not different from that observed in this study. Two meta-analysis of the performance of RADT by Lean *et al*.[26] and Stewart *et al*.[27] Both reported pooled sensitivities of 86% and pooled specificities of 96% and 92% respectively which were similar to the findings in this study.

Several factors influence the performance of a RADT.[28,30] In this study, the sensitivity of the RADT increased from 66% to 100% as the likelihood of GAS pharyngitis using the CPR increased from two to four. The phenomenon of spectrum bias which is the increase in the sensitivity of the RADT with increasing likelihood of GAS pharyngitis has been demonstrated by previous studies.[31,32] Therefore, the RADT is likely to be more sensitive in patients identified to be at higher risk of GAS pharyngitis. This study was carried out in a hospital setting and may have included more severe cases of acute pharyngitis that are more likely to be due to GAS pharyngitis. This may have explained the high sensitivity of the RADT obtained in this study.

The RADT in this study would have missed only 12 (6.4%) of true GAS positive cases compared to the throat swab culture and will inappropriately treat 7 (9.6%) of true GAS negative cases. These findings are comparable to previous studies.[15,24,33] Rimoin *et al*[15] in a multicenter study of the utility of RADT for the diagnosis of GAS pharyngitis in four low-income countries found a missed diagnosis rate of 3.2% – 8.1% and inappropriate antibiotics use of 2.7% – 10.1%. McIsaac *et al* reported 14.2% and 1.0% for missed GAS cases and unnecessary antibiotics use respectively while Gonsu *et al* reported 25% and 4% respectively.[33] The higher misclassification rates reported by Gonsu *et al*[24] may be because the study includes both children and adults. Adults have a lower risk of GAS pharyngitis which will reduce the overall performance of the RADT.

Conclusions

The RADT had good performance measures and therefore, can be used as a substitute for throat swab cultures where they are not readily available. The RADT should not be performed in patients who have already taken antibiotics within two weeks prior to presentation

Conflict of interest: None
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References

1. Carapetis JR. The Current Evidence for the Burden of Group A Streptococcal Diseases. WHO. 2004;1–57. Available from: http://apps.who.int/iris/bitstream/10665/69063/1/WHO_FCH_CAH_05.07.pdf. Accessed on 6th April 2016.–57.
2. Cherry DK, Wood well DA, Rechtsteiner EA. National Ambulatory Medical Care Survey: 2005 summary. *Advanced Data* 2007;(387):1–39. Available from: http://www.ncbi.nlm.nih.gov/pubmed/17703793. Accessed 17th May 2017.
3. Pfoh E, Wessels MR, Goldmann D, Lee GM. Burden and economic cost of group A streptococcal pharyngitis. *Paediatrics* 2008;121(2):229–34.
4. Linder JA. Antibiotic Treatment of Children With Sore Throat. *JAMA 2005;294 (18):2315.*
5. May PJ, Bowen AC, Carapetis JR. The inequitable burden of group A streptococcal diseases in indigenous Australians. *Med J Aust 2016;205(5):201–4.*
6. Bisno AL. Acute Pharyngitis : Etiology and diagnosis. *Paediatrics* 1996;97:949.
7. Yaguo-Ide LE, Uchenwa-Onyenegecha TA. Burden of Acute Respiratory Tract Infections as Seen in University of Port Harcourt Teaching Hospital Nigeria. *J US-China Med Sci 2015;12(4):158–62.*
8. Mawak HJ, Ewelike JD, Lar IC, ZumbesPM. Bacterial Aetiologic Agents Associated with Upper Respiratory Tract Infections in Children (under five years) attending selected clinics in Jos, Nigeria. *Highl Med J 2006;4(1):13–6.*
9. Sadoh WE, Sadoh AE, Oladipo AO, Okunola OO. Bacterial isolates of tonsillitis and pharyngitis in a paediatric casualty setting. *J Med Biomed Res 2011;7(1–2):37–44.*
10. Zühlke L, Mirabel M, Marijon E. Congenital heart disease and rheumatic heart disease in Africa: Recent advances and current priorities. *Heart 2013;99(21):1554–61.*
11. Bisno AL, Gerber MA, Gwaltney Jr. JM, Kaplan EL, Schwartz RH. Diagnosis and management of group A streptococcal pharyngitis: a practice guideline. Infectious Diseases Society of America. *Clin Infect Dis 1997;25(3):574–83.*
12. Shulman ST, Bisno AL, Clegg HW, Gerber MA, Kaplan EL, Lee G, e*t al*. Clinical practice guideline for the diagnosis and management of group A streptococcal pharyngitis: 2012 update by the Infectious Diseases Society of America. *Clin Infect Dis 2012;55(10):1279–82.*
13. American Academy of Pediatrics. Group A streptococcal Infections. In: Pickering LK, Baker CJ, Kimberlin DW, Long SS, eds. Red Book: 2009 Report of the Committee on Infectious Diseases. 28th ed. Elk Grove Village, IL: American Academy of Pediatrics; 2009:p616-27.
14. Webb KH. Does culture confirmation of high-sensitivity rapid streptococcal tests make sense? A medical decision analysis. *Pediatrics* 1998;101 (2):E2.Available from http://www.ncbi.nlm.nih.gov/pibmed/9445512. DOI:10.1542/peds.101.2.e2.
15. Rimoin AW, Walker CF, Hamza HS, Elminawi N, Ghafar HA, Vince A, *et al*. The utility of rapid antigen detection testing for the diagnosis of streptococcal pharyngitis in low-resource settings. *Int J Infect Dis 2010;14(12):p1048–53.*
16. Gerber MA, Shulman ST. Rapid Diagnosis of Pharyngitis Caused by Group A Streptococci. *Clin Microbiol Rev 2004;17(3):571–80.*
17. Sadoh WE, Akinsete AM. Physicians management of sore throat in children in Benin City , Nigeria. *Niger J Clin Pract 2009;12(4):407–11.*
18. Smeesters PR, Campos D, Van Melderen L, de Aguiar E, Vanderpas J, Vergison A. Pharyngitis in Low-Resources Settings: A Pragmatic Clinical Approach to Reduce Unnecessary Antibiotic Use. *Pediatrics 2006;118(6):1607–11.*
19. Howard DH, Scott RD. The Economic Burden of Drug Resistance. *Clin Infect Dis* 2005;41(s4):S283–6. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16032567%5Cnhttp://cid.oxfordjournals.org/lookup/doi/10.1086/430792. Accessed 23rd February 2016.
20. Johnson DR, Kaplan EL. Laboratory diagnosis of group A streptococcal infections. *WHO Geneva 1996. p4-12.*
21. Olusanya O, Okpere EE, Ezimokhai M. The Importance of socioeconomic class in voluntary fertility in developing countries. *West Afr Med J 1985;4:205–7.*
22. Mclsaac WJ, White D, Tannenbaum D, Low DE. A clinical score to reduce unnecessary antibiotic use in patients with sore throat. *CMAJ 1998;158(1):75–83.*
23. Engel ME, Muhamed B, Whitelaw AC, Musvosvi PM, Mayosi BM and Dale JB. Group A Streptococcal emm Type Prevalence among Symptomatic Children in Cape Town and Potential Vaccine Coverage. *Pediatr Infect Dis J 2014;33(2):208–10.*
24. Gonsu HK, Bomki CM, Djomou F, Toukam M, Ndze VN, Lyonga EE, *et al*. A comparative study of the diagnostic methods for group A streptococcal sore throat in two reference hospitals in Yaounde, Cameroon. *Pan Afr Med J 2015;20:1–7.*

25. Malik EM, Ali SKM. Prediction of bacterial pharyngitis in children using clinical features. *Khartoum Med J. 2014;07 (02):967–71.*
26. Lean WL, Arnup S, Danchin MM, Steer AC, Danchin MM, Rogers S, *et al*. Rapid diagnostic tests for group A streptococcal pharyngitis: a meta-analysis. *Paediatrics 2014;134 (4):771–81*.
27. Stewart EH, Davis B, Clemans -Taylor BL, Littenberg B, Estrada CA, Centor RM. Rapid Antigen Group A Streptococcus Test to Diagnose Pharyngitis: A Systematic Review and Meta-Analysis. *PLoS One 2014;9(11):1–10.*
28. Fox JW, Cohen DM, Marcon MJ, Cotton WH, Bonsu BK. Performance of rapid streptococcal antigen testing varies by personnel. *J Clin Microbiol 2006;44(11):3918–22.*
29. Kurtz B, Kurtz M, Roe M, Todd J. Importance of inoculum size and sampling effect in rapid antigen detection for diagnosis of Streptococcus pyogenes pharyngitis. *J Clin Microbiol 2000;38(1):279–81.*
30. Penney C, Porter R, O'Brien M, Daley P. Operator Influence on Blinded Diagnostic Accuracy of Point-of-Care Antigen Testing for Group A Streptococcal Pharyngitis. *Can J Infect Dis Med Microbiol 2016;2016: ID 1710561. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27579047.Accessed 23rd Nov 2016.*
31. Tanz RR, Gerber MA, Kabat W, Rippe J, Seshadri R, Shulman ST. Performance of a rapid antigen-detection test and throat culture in community pediatric offices: implications for management of pharyngitis. *Paediatrics 2009;123(2):437–44.*
32. Hall MC, Kieke B, Gonzales R, Belongia EA. Spectrum bias of a rapid antigen detection test for group A beta-hemolytic streptococcal pharyngitis in a pediatric population. *Pediatrics 2004;114(1):182–6.*
33. McIsaac WJ, Kellner JD, Aufricht P, Vanjaka A, Low DE. Empirical validation of guidelines for the management of pharyngitis in children and adults. *JAMA 2004;291 (13):1587–95.*