Discrepancy in Antimicrobial Susceptibility Test Results Obtained for Oral Streptococci with the Etest and Agar Dilution[▽]

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A total of 270 viridans group streptococci (VS) isolated from healthy children, identified to the species level, were tested for their susceptibilities to penicillin, imipenem, erythromycin, and vancomycin. A total of 270 isolates and 1,080 organism-antibiotic combinations were evaluated. The overall susceptibility rates of all isolates obtained by the Etest (ET) versus agar dilution (AD) were 60.4% versus 61.8% for penicillin, 63.8% versus 63.9% for erythromycin, 90.6% versus 96% for vancomycin, and 99.1% versus 96.0% for imipenem, respectively. Major discrepancies occurred in the testing of the susceptibility of *Streptococcus mutans* to vancomycin, with 59.5% (ET) versus 100% (AD), followed by *S. salivarius*, with 84.1% versus 100%; *S. oralis*, with 82.1% versus 96.4%; and *S. mitis*, with 90% versus 100%, respectively. There were also differences in the rates of susceptibility of *S. mutans*, 66.5% (ET) versus 85.1% (AD), and *S. intermedius*, 82.9% versus 72.1%, respectively, to penicillin. General agreement between the results of ET and AD was obtained for 973 organism-antibiotic combinations out of 1,080 antibiotic combinations, i.e., 90.1%. Very major errors were found for 6.8% of isolates, and major errors were found for 3.2% of isolates; the minor errors were negligible. Agreement between the results of the two methods was 98.7% for penicillin, 94.6% for vancomycin, 96.9% for imipenem, and 99.9% for erythromycin. The highest rate of very major errors was for vancomycin, at 5.4%. The ET appears to be as efficient as AD for susceptibility testing of VS, except for vancomycin, where very major errors in the results were relatively high.

The oral cavity, a home to a wide variety of microorganisms, is a large reservoir of antibiotic-resistant bacteria. Antibiotic resistance among the oral microflora has been well documented, and it is gaining increasing attention, especially among the viridans group streptococci (VS). VS are a major cause of morbidity and mortality associated with bloodstream infections, shock, and respiratory distress syndrome in febrile neutropenic patients and play a leading role in the etiology of infective endocarditis (12, 13, 15, 16, 19, 20). Their portal of entry is oral and gastrointestinal mucosal lesions and intravascular catheters (7, 20). A major factor associated with infective endocarditis is orodental procedures, which may lead to hematogenous dissemination of VS into many extraoral sites (1, 14).

Penicillin and, to a lesser extent, erythromycin are the mainstays of therapy for infections caused by VS, and VS have been uniformly susceptible to β -lactam antimicrobial agents (3, 16, 20). However, antimicrobial resistance among VS is now becoming an obstacle to the antibiotic therapy of, for example, endocarditis (13, 15, 16). The emerging increasing rate of microbial resistance to β -lactam antimicrobial agents, macrolides, and tetracyclines among VS poses a serious clinical problem in the management of therapy (1, 3, 12, 15, 16, 20). Currently, about 25% and 33% of all VS isolated from subjects in Kuwait are resistant to penicillin and erythromycin, respectively (13). Hence, the determination of susceptibility of an

isolate of VS to penicillin and other antimicrobial agents is a clinical priority.

Antimicrobial susceptibility testing can be done by a variety of methods. The agar dilution (AD) method is considered to be the reference against other methods (2, 6). However, this conventional method is time-consuming, cumbersome, and inadequate for routine testing in many clinical laboratories (5, 6). A novel method, the Etest (ET) susceptibility testing method, developed based on the diffusion of the antibiotic from a preformed continuous antibiotic gradient from a plastic strip, appears to be much simpler and less time-consuming than reference methods (2, 5, 6, 12). This method has been recommended for susceptibility testing of *Streptococcus pneumoniae* and *Enterococcus* sp. isolates in clinical diagnostic laboratories (17, 18).

Very few studies have compared the results of the ET susceptibility method for testing VS isolates to those of the reference methods. This study was undertaken to assess the reliability of the results obtained by ET compared to those obtained by the reference AD method.

MATERIALS AND METHODS

Bacterial isolates. VS obtained from culturing plaque samples collected from teeth and tongue surfaces of 6- to 12-year-old healthy children in Kuwait were pooled and studied for their susceptibilities to four standard antibiotics. Isolates were stored at -70°C in skim milk prior to testing. When needed, they were thawed, inoculated onto tryptic soy agar (Difco, Becton Dickinson & Co., Sparks, MD) supplemented with 5% sheep blood, and incubated in the presence of 5% CO_2 at 35°C for 18 to 24 h; the isolates were then serially passaged three times using the same culture medium to ensure adequate growth and purity. They were identified by colonial and Gram-stained cell morphologies as well as by enzymatic/biochemical profiling by using commercially available API 20S kits (bio-Merieux, Marcy l'Etoile, France) and glycosidase activities with 4-methylumbel-

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liferyl-linked fluorogenic substrate profiling (4) designed for the identification of streptococci to the species level. The preparation and incubation of the kit were carried out according to the manufacturer's recommendations. The results, in numerical code, were input into a database system for identification (API Plus; bioMérieux).

Susceptibility testing. Susceptibility testing of penicillin, erythromycin, vancomycin, and imipenem against VS was performed by determining their MICs using the AD method (15) and the ET method (AB Biodisk, Solna, Sweden). Tests were performed using Muller-Hinton agar (Oxoid, Basingstoke, United Kingdom) supplemented with 5% sheep blood (Oxoid). Media and inoculum preparation and MIC interpretations for AD were performed in accordance with previously reported guidelines (11). ET inoculum preparation and plating, strip application, and subsequent MIC determinations were carried out in accordance with the manufacturer's instructions. AD and ET were performed in parallel and in triplicate. The antibiotic concentrations in AD tests ranged from 0.002 to 256 µg/ml, while those in the ET ranged from 0.016 to 256 µg/ml. These concentrations applied to all the test antibiotics except imipenem, whose concentration ranged from 0.002 to 32 µg/ml. The AD and ET results were analyzed and compared using statistical methods. The breakpoints used to define susceptible and resistant categories for each antimicrobial agent were those recommended by the Clinical and Laboratory Standards Institute (CLSI; formerly NCCLS) (11). *Streptococcus pneumoniae* ATCC 49619 was included in each run as a quality control.

The results obtained by the ET and AD were analyzed for their agreement and correlation, where agreement was defined as the ET MIC equals the AD MIC plus or minus a single twofold dilution (12). The degree of agreement between the ET MIC and the AD MIC assessed by interclass correlation coefficients and intraclass correlation coefficients was used to assess the consistency of the MICs of the tested antibiotics for individual VS over serial ETs and serial ADs; coefficients of 0.90 meant excellent correlation between or among tests. Discrepancies observed when the ET and AD did not assign the same susceptibility category to an isolate were described as follows: very major error, when the isolate was categorized as being susceptible by ET but resistant by AD; major error, when the isolate was categorized as being resistant by ET but susceptible by AD; and minor error, when there were other mismatches of ET and AD susceptibility results.

Statistical analysis using SPSS. The AD and ET results were statistically analyzed using a *t* test to find out any significant differences between (i) the two methods, (ii) the susceptibilities of the isolates to the tested antimicrobial agents, and (iii) the test of agreement for susceptibility classification by ET and that by AD using an interclass coefficient of correlation.

RESULTS

A total of 270 VS were studied. Table 1 shows the average ranges of MICs and MIC₉₀s of penicillin, vancomycin, imipenem, and erythromycin against individual species and the average percentage of isolates susceptible to each of the antibiotics as determined by the ET and AD methods. As there was good growth of all isolates for MIC determinations, the tests were not incubated beyond 24 h and were performed in triplicate. Two of us carried out the reading and interpretation of the tests independently and compared the results thereafter. Any discrepancies were resolved by consensus. Categorization of an isolate as being resistant or sensitive was done in accordance with the recommended breakpoint criteria (11). There was good agreement in the overall susceptibility rates of all isolates obtained by the ET and AD for penicillin, 60.4% and 61.8%, and erythromycin, 63.8% and 63.9%, respectively, but some discrepancies were observed with vancomycin, 90.6% and 96%, and imipenem, 99.1% and 96.0%, respectively. Analysis of the individual species showed variation in their susceptibilities by the two methods. Major discrepancies occurred in the susceptibility testing of *S. mutans* against vancomycin, with 59.5% (ET) versus 100% (AD), followed by *S. salivarius*, with 84.1% versus 100%; *S. oralis*, with 82.1% versus 96.4%; and *S. mitis*, with 90% versus 100%, respectively. There were also

differences in the rates of susceptibility of *S. mutans*, 66.5% (ET) versus 85.1% (AD), and *S. intermedius* 82.9% versus 72.1%, respectively, to penicillin.

Data analysis using CLSI breakpoints. When evaluated using CLSI breakpoint guidelines, overall agreement between the results of ET and those of AD was obtained in 973 antibiotic combinations out of 1,080 antibiotic combinations, i.e., 90.1%. Very major errors were found for 6.8% of isolates, and major errors were found for 3.2% of isolates; there was no minor error. Agreement between the results of the two methods was 98.7% for penicillin, 94.6% for vancomycin, 96.9% for imipenem, and 99.9% for erythromycin. The highest rate of very major errors was found for vancomycin, 5.4%, followed by penicillin, 1.3%, and erythromycin, 0.1%, while major errors were found for imipenem, 3.2%. In individual species, the highest rates of very major errors were found with vancomycin in *S. salivarius* (*n* = 9), followed by *S. mutans* (*n* = 8), *S. oralis* (*n* = 4), and *S. mitis* (*n* = 3). The highest rates of major errors were for penicillin against *S. mutans* (*n* = 4) and imipenem against *S. oralis* (*n* = 4).

Statistical analysis of MICs determined by ET and AD. Statistical analysis was performed to compare the MICs obtained by the two methods. Table 2 shows the mean MICs of the four antibiotics determined by the two methods; there was no significant difference between the two methods in determining the MICs for all the antibiotics except for vancomycin, where the difference reached statistical significance (*P* < 0.001). The mean MICs of imipenem were almost identical by both methods (0.13 ± 0.16 µg/ml by ET and 0.12 ± 0.16 µg/ml by AD). A test of agreement between the ET and AD was done, and the results are as follows. There was agreement in the MICs of imipenem determined by both ET and AD, with an interclass value of 0.92 (95% confidence interval, 0.89 to 0.93). This was followed by penicillin, with an interclass value of 0.84 (95% confidence interval, 0.80 to 0.87), and erythromycin, with an interclass value 0.79 (95% confidence interval, 0.73 to 0.83). The MICs of vancomycin determined by ET showed poor agreement with those determined by AD, with a value of 0.62 (95% confidence interval, 0.10 to 0.83).

DISCUSSION

MIC determination is the standard quantitative technique for testing of antimicrobial susceptibility of bacteria (6) in many clinical laboratories. The recommended method for MIC determination is the AD method, which is regarded as the reference method against which other methods are assessed (2, 5, 6). However, this test is time-consuming and requires careful control of the technique to produce reliable results (6). Since the advent of ET, its versatility and ease of use make the method an attractive alternative to conventional methods. Thus, we undertook a deliberate attempt to evaluate the reliability of ET for the routine testing of susceptibilities of VS by comparing ET with the conventional AD method.

Comparison of the ET with AD showed a very good agreement with imipenem susceptibility testing, which gave a high positive correlation coefficient (>0.92), followed by penicillin (>0.84) and erythromycin (>0.79). In contrast, vancomycin had relatively low agreement (>0.62). This finding is supported by a previous report by Rosser et al. (12), who also

TABLE 1. Susceptibility patterns of oral VS determined by ET and AD

Streptococcus (no. of isolates) and antibiotic (breakpoint in µg/ml)	ET			AD		
	MIC (µg/ml)		% Susceptible	MIC (µg/ml)		% Susceptible
	Range	90%		Range	90%	
<i>S. salivarius</i> (58)						
Penicillin G (0.125)	0.016–4	1.5	24	0.0125–6.4	1.6	27.9
Vancomycin (1)	0.016–4	1.5	84.1	0.1–0.8	0.8	100
Imipenem (0.5)	0.003–1	0.38	96.4	0.013–1	0.6	93.7
Erythromycin (0.25)	0.016–>256	4	67.4	0.025–12.8	6.4	67.4
<i>S. sanguis</i> (44)						
Penicillin G (0.125)	0.016–6	0.75	49.8	0.005–3.2	0.6	53.3
Vancomycin (1)	0.38–2	1.25	91.2	0.2–1.6	1.2	91.2
Imipenem (0.5)	0.008–0.38	0.25	100	0.006–0.8	1.2	94.8
Erythromycin (0.25)	0.016–256	6	49.5	0.006–12.8	8	49.5
<i>S. mutans</i> (20)						
Penicillin G (0.125)	0.023–1	0.625	66.5	0.025–1.6	0.45	53.3
Vancomycin (1)	0.5–1.5	1.5	59.5	0.2–0.8	0.8	100
Imipenem (0.5)	0.008–0.125	0.125	100	0.006–0.8	0.1	96.2
Erythromycin (0.25)	0.016–6	4	77.5	0.0125–12.8	6.6	73.6
<i>S. oralis</i> (28)						
Penicillin G (0.125)	0.016–1	0.875	32.7	0.005–1.6	0.8	35.7
Vancomycin (1)	0.38–1.5	1.5	82.1	0.4–1.6	0.8	96.4
Imipenem (0.5)	0.008–0.5	0.38	100	0.013–0.8	0.4	89.3
Erythromycin (0.25)	0.023–4	1.5	50	0.025–3.2	1.6	53.6
<i>S. mitis</i> (28)						
Penicillin G (0.125)	0.016–8	1.5	39.7	0.0125–6.4	1.2	35.9
Vancomycin (1)	0.008–1.5	1.25	90	0.1–1.6	0.8	100
Imipenem (0.5)	0.016–0.75	0.29	100	0.005–1.6	0.45	88.5
Erythromycin (0.25)	0.016–256	4.5	66.1	0.006–12.8	4.0	64.61
<i>S. intermedius</i> (19)						
Penicillin G (0.125)	0.016–1	0.19	82.9	0.0125–0.8	0.3	72.2
Vancomycin (1)	0.38–1.5	1	89.2	0.4–1.6	0.8	93.8
Imipenem (0.5)	0.004–0.19	0.1	100	0.006–0.4	0.15	100
Erythromycin (0.25)	0.016–0.75	0.31	87.5	0.0125–0.8	0.25	81.3
<i>S. bovis</i> (17)						
Penicillin G (0.125)	0.023–1	0.38	76.4	0.025–0.8	0.4	76.4
Vancomycin (1)	0.38–1	1	100	0.4–0.8	0.8	100
Imipenem (0.5)	0.008–0.25	0.16	100	0.0125–0.4	0.1	100
Erythromycin (0.25)	0.064–1	0.75	41.7	0.05–1.6	0.8	67.5
<i>S. anginosus</i> (9)						
Penicillin G (0.125)	0.023–0.75	0.625	67.5	0.025–0.8	0.8	67.5
Vancomycin (1)	0.38–1	1	100	0.4–0.8	0.8	100
Imipenem (0.5)	0.008–0.125	0.125	100	0.006–0.1	0.1	100
Erythromycin (0.25)	0.016–1	0.875	55	0.0125–0.8	0.8	55
<i>S. suis</i> (5)						
Penicillin G (0.125)	0.032–1.5	0.81	83.3	0.005–1.6	0.85	83.3
Vancomycin (1)	0.38–1	0.875	100	0.4–0.8	0.8	100
Imipenem (0.5)	0.008–0.5	0.25	100	0.0125–0.4	0.21	100
Erythromycin (0.25)	0.016–1	0.625	83.3	0.0125–0.8	0.5	83.3
<i>S. acidominimus</i> (24)						
Penicillin G (0.125)	0.016–1	0.625	68.8	0.025–0.8	0.6	68.8
Vancomycin (1)	0.38–1	1	100	0.2–0.8	0.8	100
Imipenem (0.5)	0.008–0.75	0.44	96.9	0.0125–0.8	0.6	96.9
Erythromycin (0.25)	0.016–>256	1.375	71.9	0.0125–12.8	0.6	75
<i>S. constellatus</i> (18)						
Penicillin G (0.125)	0.016–0.75	0.5	73.2	0.025–0.8	0.21	73.2
Vancomycin (1)	0.125–1	1	100	0.1–0.8	0.8	100
Imipenem (0.5)	0.008–0.5	0.5	96.4	0.006–0.8	0.4	96.4
Erythromycin (0.25)	0.016–12	5	53.6	0.0125–12.8	2	53.6

TABLE 2. Comparison of mean MICs for ET with those for AD

Antibiotic	Mean MIC ($\mu\text{g/ml}$) $\pm$ SD		<i>P</i> value ^a
	ET	AD	
Penicillin	0.45 $\pm$ 0.78	0.42 $\pm$ 0.91	0.43
Vancomycin	0.85 $\pm$ 0.33	0.60 $\pm$ 0.26	0.0001*
Imipenem	0.13 $\pm$ 0.16	0.12 $\pm$ 0.16	0.23
Erythromycin	1.41 $\pm$ 4.45	1.35 $\pm$ 2.83	0.74

^a *, statistically significant.

found a relatively low agreement of 86.7% for vancomycin compared with 92, 96, and 91% agreement for penicillin, cefuroxime, and cefotaxime, respectively, against clinical isolates of VS. The mean MICs of all the four antimicrobial agents used in the study were slightly higher for ET than for AD, with the difference being higher in the case of vancomycin (0.85 $\mu\text{g/ml}$ for ET and 0.60 $\mu\text{g/ml}$ for AD).

The often-quoted criteria for choosing an antimicrobial susceptibility testing system are as follows: 90% of the test MICs should be within ± 1 twofold dilution of the MIC obtained by the reference method, i.e., agreement; very major errors should occur in $<3\%$ of all comparisons for isolates shown to be resistant by the reference method; and combined major and minor error rates should be $<7\%$ (9). In our study, the criterion for agreement was met for tests with imipenem, penicillin, and erythromycin. However, the agreement for vancomycin susceptibility testing was not as good as those for the other antibiotics, suggesting that for many of the isolates, the MICs obtained by the ET were a 1.5 dilution greater than the corresponding AD MICs. Our present findings are somewhat in agreement with data from previous reports by other authors (2, 8, 10, 11, 17). In those reports, all the other antibiotics tested using ET and other reference methods also gave a good agreement except for vancomycin. It was rationalized that this finding could be due to the narrow range at which the MICs for vancomycin usually fall (0.5 $\mu\text{g/ml}$ to 2 $\mu\text{g/ml}$), which in our hands were from 0.5 $\mu\text{g/ml}$ to 1.5 $\mu\text{g/ml}$. In their study, Hashemi et al. (8) pointed out that the CLSI guidelines for the breakpoints are valid only for the interpretation of results of susceptibility testing by broth dilution and AD methods, and many laboratories use them to interpret results derived by other susceptibility testing methods. While there were differences between the two methods, all of the differences occurred at the breakpoints between susceptibility categories, traditionally problematic when one is comparing results based on a twofold dilution scheme. This difficulty is compounded by the fact that the ET provides results on a linear scale, so these results are not directly comparable to the twofold dilution results provided by the reference methods (10). This may explain the reason why, in our study, many strains that are susceptible to vancomycin by AD were not so by ET.

In conclusion, we found that the ET susceptibility method is a reliable method for determinations of MICs of imipenem, erythromycin, and penicillin but may produce an overestimation of the MICs of vancomycin for many VS. Thus, some strains that are vancomycin susceptible by the AD method may appear to be resistant by the ET method. A simpler and less

expensive susceptibility testing method, such as the disk diffusion method, augmented with MIC determinations for isolates with equivocal results may be more useful.

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REFERENCES

- Amrouche, M. L., L. Schlegel, and A. Bouvet. 2004. Impact of susceptibility to antibiotics of streptococci and enterococci isolated from patients with infective endocarditis on antibiotic treatment. *Ind. J. Med. Res.* **119**(Suppl.): 80–83.
- Baker, C. N., S. A. Stocker, D. H. Culver, and C. Thornsberrry. 1991. Comparison of the E-test to agar dilution, broth dilution, and agar diffusion susceptibility testing techniques by using a special challenge set of bacteria. *J. Clin. Microbiol.* **29**:533–538.
- Bancescu, G., S. Dumitriu, A. Bancescu, C. Defta, M. Pana, D. Ionescu, S. Alecu, and M. Zamfirescu. 2004. Susceptibility testing of *Streptococcus mitis* group isolates. *Ind. J. Med. Res.* **119**(Suppl.):257–261.
- Beighton, D., J. M. Hardie, and R. A. Whaley. 1991. A scheme for the identification of viridans streptococci. *J. Med. Microbiol.* **35**:367–372.
- Bonventura, G. D., E. Ricci, N. D. Loggia, G. Catamo, and R. Piccolomini. 1998. Evaluation of the E-test for antimicrobial susceptibility testing of *Pseudomonas aeruginosa* isolates from patients with long-term bladder catheterization. *J. Clin. Microbiol.* **36**:824–826.
- Brown, D. F. J., and L. Brown. 1991. Evaluation of the E-test, a novel method of quantifying antimicrobial activity. *J. Antimicrob. Chemother.* **27**:185–190.
- Doern, G. V., M. J. Ferraro, A. B. Brueggemann, and K. L. Ruoff. 1996. Emergence of high rates of antimicrobial resistance among viridans group streptococci in the United States. *Antimicrob. Agents Chemother.* **40**:891–894.
- Hashemi, F. B., G. E. Schutze, and E. O. Mason, Jr. 1996. Discrepancies between results by E-test and standard microbroth dilution testing of *Streptococcus pneumoniae* for susceptibility to vancomycin. *J. Clin. Microbiol.* **34**:1546–1547.
- Jorgensen, J. H. 1993. Selection criteria for an antimicrobial susceptibility testing system. *J. Clin. Microbiol.* **31**:2841–2844.
- Macias, E. A., E. O. Mason, Jr., H. Y. Ocera, and M. T. Larocco. 1994. Comparison of E-test with standard broth microdilution for determining antibiotic susceptibilities of penicillin-resistant strains of *Streptococcus pneumoniae*. *J. Clin. Microbiol.* **32**:430–432.
- National Committee for Clinical Laboratory Standards. 2002. Performance standards for antimicrobial susceptibility testing. Informational supplement: approved standard M100-S12, p. 114–115. NCCLS, Wayne, PA.
- Rosser, S. J., M. J. Alfa, S. Hoban, J. Kennedy, and G. K. Harding. 1999. E-test versus agar dilution for antimicrobial susceptibility testing of viridans group streptococci. *J. Clin. Microbiol.* **37**:26–30.
- Rotimi, V. O., N. O. Salako, E. M. Mokaddas, L. Philip, and P. Rajan. 2005. High frequency of isolation of antibiotic-resistant oral viridans streptococci from children in Kuwait. *J. Chemother.* **17**:493–501.
- Salako, N. O., V. O. Rotimi, S. M. Adib, and S. Al-Mutawa. 2004. Pattern of antibiotic prescription in the management of oral disease among dentists in Kuwait. *J. Dent.* **32**:503–509.
- Smith, A., M. S. Jackson, and H. Kennedy. 2004. Antimicrobial susceptibility of viridans group streptococci blood isolates to eight antimicrobial agents. *Scand. J. Infect. Dis.* **36**:259–263.
- Teng, L. J., P. R. Hsueh, Y. C. Chen, S. W. Ho, and K. T. Luh. 1998. Antimicrobial susceptibility of viridans streptococci in Taiwan with emphasis on the high rates of resistance to penicillin and macrolides in *Streptococcus oralis*. *J. Antimicrob. Chemother.* **41**:621–627.
- Tenover, F. C., C. N. Baker, and J. M. Swenson. 1996. Evaluation of commercial methods for determining antimicrobial susceptibility of *Streptococcus pneumoniae*. *J. Clin. Microbiol.* **34**:10–14.
- Tenover, F. C., J. M. Swenson, C. M. O'Hara, and S. A. Stocker. 1995. Ability of commercial and reference antimicrobial susceptibility testing methods to detect vancomycin resistance in enterococci. *J. Clin. Microbiol.* **33**:1524–1527.
- Thomas, J. P. 2003. Antibiotic resistance. *Dent. Clin. N. Am.* **47**:623–639.
- Wisplinghoff, H., R. R. Reinert, O. Cornely, and H. Seifert. 1999. Molecular relationships and antimicrobial susceptibilities of viridans group streptococci isolated from blood of neutropenic cancer patients. *J. Clin. Microbiol.* **37**: 1876–1880.