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ERYTHROMYCIN-RESISTANT *STREPTOCOCCUS PYOGENES* IN ARGENTINA

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Abstract Erythromycin (ERY) resistance in *Streptococcus pyogenes* has recently emerged as a problem of growing concern all through the world. We are presenting the comparison of results of the continuous surveillance of erythromycin resistance in *S. pyogenes* performed since 1989 in the Hospital de Pediatría J.P.Garrahan of Buenos Aires City, with independently observed rates in other five centers of Buenos Aires and seven centers of six other Argentinian cities, obtained between 1999 and 2001. A significant increase of erythromycin resistance was observed among *S. pyogenes* isolated in the Hospital Garrahan (6.6% in 1998-1999 to 9.9% in 2000). Similar trends were also detected in other centers of other Argentinian cities when recent data were compared to results of a multicenter study performed in 1995. However, lower rates of resistance were recorded in Mendoza, Cipolletti and Neuquén in comparison with data of 1995, 1998 and 1998 respectively. The reason of such decreasing resistance rates deserves to be investigated. The average of ERY-resistance rates obtained in the surveyed centers was 6.7% (range 0.5 - 14.1%). Control of antimicrobial use should be performed to warrant the future effectiveness of macrolide antibiotics regarding the positive association between use and resistance. These results also suggest that susceptibility tests for macrolides should be performed whenever *S. pyogenes* is isolated in Argentina.

Key words: erythromycin, *Streptococcus pyogenes*, group A streptococci, macrolides, resistance

Resumen ***Resistencia a la eritromicina en* Streptococcus pyogenes *en la Argentina*.** La resistencia a la eritromicina en *Streptococcus pyogenes* ha emergido en los últimos tiempos como un problema creciente en todo el mundo. En este trabajo se presenta la comparación de los resultados de la vigilancia continua de la resistencia a la eritromicina que se viene realizando en el Hospital de Pediatría J.P.Garrahan de Buenos Aires desde 1989, con resultados independientes de otros cinco centros de la ciudad de Buenos Aires y siete de otras seis ciudades argentinas, obtenidos entre 1999 y 2001. Se observó un aumento significativo en el Hospital Garrahan (6.6% en 1998-1999 a 9.9% en el año 2000) y una tendencia similar en otros centros de diversas ciudades argentinas si se comparan estos datos con los de un estudio multicéntrico realizado en 1995. No obstante, se registraron menores porcentajes de resistencia en Mendoza, Neuquén y Cipolletti, en relación a lo hallado en 1995, 1998 y 1998 respectivamente. La razón de esta disminución merece ser investigada. El porcentaje promedio de resistencia a eritromicina obtenido en los distintos centros participantes de este estudio fue de 6.7% (rango 0.5-14.1%). Debe efectuarse un control en el uso de estos antibióticos para garantizar la efectividad futura de los macrólidos, teniendo en cuenta la asociación estrecha entre uso y resistencia. Estos resultados sugieren que deberían realizarse pruebas de sensibilidad a los macrólidos para todos los aislamientos de *S. pyogenes* en la Argentina.

Palabras clave: eritromicina, *Streptococcus pyogenes*, estreptococos grupo A, macrólidos, resistencia

Erythromycin (ERY) has been extensively used in the management of pediatric infections since it was discovered in 1952. Mainly, it is alternative in cases of penicillin allergy or in infections due to penicillin-resistant organisms. Macrolide use was enhanced by the recent introduction of new compounds with better pharmaco-kinetic properties, gastrointestinal tolerability and decreased interactions with commonly used drugs.

ERY-resistant group A streptococci (ERGAS) have already been described in several countries from differ-

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TABLE 1.– *Erythromycin resistance in different Argentinian centers (1998-2001)*

City (Center)	Period	Pharyngeal exudates	*Streptococcus pyogenes* tested isolates	ERGAS[$] (% of tested isolates)	Phenotype M (% of ERGAS)
B. Aires (HPJPG) *	Ago 98 - Jul 99	4474	884 (884)	58 (6.6)	94.4
B. Aires (HPJPG)	Jan - Dec 00	4942	772 (742)	73 (9.9)	99.6
B. Aires (HNRG) [†]	Mar 00 - Feb 01	5140	804 (488)	16 (3.3)	87.6
B. Aires (C[¶]+ FF[#])	Oct 99 - Mar 00	N.A.°	329	20 (6.0)	95.0
B. Aires (HAP) **	Jun 99 - Aug 99	783	114 (114)	7 (6.1)	100
B. Aires (HF) [‡‡]	Apr 00 -May 01	N.A.	64 (64)	9 (14.1)	100
Neuquén (PN[§§]+BR[¶¶])	Jun 00 - Jun 01	576	168 (98)	8 (8.2)	100
Cipolletti (HAC) ***	Jan 99 - Jul 01	423	55 (24)	1 (4.1)	0
Bariloche (HC +SS) [§]	Jan 00 - Jul 01	N.A.	395 (395)	2 (0.5)	50
Córdoba (CRF) [##]	Jan 99 - Jul 01	284	47 (47)	2 (4.2)	100
Mendoza (HN)[‡]	Jan 99 - Jun 01	3483	544 (544)	30 (5,5)	100
Esquel (HZ) [††]	Jan - Jun 01	484	113 (83)	10 (12.0)	100

[$] *ERGAS = Erythromycin-resistant group A streptococci*
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° *N.A.: Non-available data*

ent continents in rates higher than 10%[1-3]. However in some cases the problem disappeared as macrolide use diminished[3]. Overuse of macrolides was identified as the main cause of resistance increase[1, 3-5]. There are two well-characterized mechanisms that may be responsible of such resistance in beta-hemolytic streptococci: target site modification (ribosome methylation) and active drug efflux[6, 7].

ERY resistance in *S. pyogenes* was recognized as a rare event in Argentina by 1994. Macrolide resistance may be due to either inducible or constitutive 23S ribo-somal dimethylation (MLS_B), active efflux (M phenotype) or modification of protein L4 or RNA sequences by mutation. MLS_B may be coded either by the *ermTR* gene (now classified as an *ermA* variant) or by the *ermB* gene. Active efflux is coded by *mefA* or *mefE* genes in streptococci. Only few isolates showing an inducible MLS_B phenotype were obtained during 1989[8]. Fifty eight centers from 27 Argentinian cities participated in two monthly studies on prevalence of ERY susceptibility of *S. pyogenes*. Prevalences of 0.14% (1 isolate) and 0.28% (3 isolates) in May and October 1994 were respectively observed from a total of 1767 (713 and 1054) tested isolates[9]. Three isolates showed the MLS_B phenotype, two of them having the *ermTR* gene and one strain had the efflux mechanism (M phenotype coded by *mef A* gene)[10].

Later, in 1995, 11.1% (15/135) of resistant isolates have been reported in Mendoza[9] and 12% (30/251) in 1998 in Neuquén and Cipolletti[11]. All those resistant isolates showed the M phenotype. Some of them were characterized as having the *mefA* gene[10].

We are describing now the evolution of erythromycin resistance in *S. pyogenes* isolated in one pediatric hospital of Buenos Aires comparing these results with independently observed rates in other five centers of Buenos Aires and seven centers of six other Argentinian cities.

Isolates obtained from pharyngeal exudates of children hospitalized or assisted as outpatients in any of the 13 centers were identified primarily by pyrrolidonyl arylamidase and bacitracin tests. Doubtful results were confirmed by serological tests, Voges-Proskauer and Rapid ID32 profiles (*bioMérieux*, Marcy l'Étoile, France) at the Hospital de Pediatría Prof. Dr. Juan P. Garrahan[12].

Susceptibility tests for ERY and clindamycin (CLI) were performed by the double disc method in 5% sheep blood Mueller Hinton agar[11]. Susceptibility to penicillin (PEN), ceftriaxone (CRO), ERY, CLI and azithromycin (AZI) of ERGAS isolated in the Hospital de Pediatría Prof. Dr Juan P. Garrahan were tested by the agar dilution method following National Committee for Clinical Laboratory Standards (NCCLS) guidelines[13]. ERGAS were defined as ERY non-susceptible *S. pyogenes* (zone < 21mm).

Eight hundred and eighty four isolates of *S. pyogenes* were obtained from 4474 pharyngeal exudates during one year (August 1998 - July 1999). Fifty eight of them (6.6%) were ERGAS. Eighteen ERGAS were tested by the dilution method. Minimal inhibitory concentrations (MICs) ranges in µg/ml were: PEN (<0.007-0.015), CRO (<0.007), ERY (4-16), CLI (<0.06-8) and AZI (32-64). Most of ERGAS (94.4%) had an M phenotype suggesting that an efflux mechanism was involved. Only one isolate (5.6%) was recognized as having the MLS_B inducible susceptibility pattern.

Seven hundred and forty two group A beta-hemolytic streptococci out of 772 isolated from 4 942 pharyngeal exudates during 2000 in the same hospital were studied in the same way.

Higher rates of ERGAS (9.9%) were observed in this instance as compared with those of 1988-89 (6.6%) (chi square, P = 0.01). A comparison with recent data from other Argentinian cities is presented in Table 1. ERY resistance range was 3.3-14.1% in the period 1998-2001 for the Buenos Aires area (average = 7.7%). In six other cities the range was 0.5 - 12% (average = 5.8%). Range in all the country was 0.5-14.1% (average 6.7%).

Concluding, a significant increase of ERY resistance was observed among *S. pyogenes* isolated in one pediatric hospital of Buenos Aires. Similar trends were also detected in other centers of other Argentinian cities when recent data were compared to previous results of a multicenter study (9). However, lower rates of ERGAS were recorded in Mendoza, Cipolletti and Neuquén in comparison with data of 1995, 1998 and 1998 respectively. The reason of such decreasing resistance rates deserves to be investigated.

The average of ERY-resistance rates obtained in the surveyed centers was 6.7% (range 0.5-14.1%). Control of antimicrobial use should be performed to warrant the future effectiveness of macrolide antibiotics regarding the possitive association between use and resistance, as previously was demonstrated in Finland[3].

These results also suggest that susceptibility tests for macrolides should be performed whenever *S. pyogenes* is isolated in Argentina. Monitoring susceptibility rates will eventually lead to rationale recommendations for treating upper respiratory infections.

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