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Effect of bacterial flora on staphylococcal colonisation of the newborn

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SUMMARY The umbilical and nasopharyngeal flora of newborn infants was examined on days 3, 14, and 42 of life. An analysis of the bacteriological findings suggests that colonisation by either *Staphylococcus aureus* or *Staph. epidermidis* prevents colonisation by the other staphylococcus. Similarly, colonisation by Gram-negative bacteria prevents colonisation by staphylococci. Further, this bacterial interference lasts for as long as 42 days, which suggests the possibility of artificially colonising newborns with nonpathogens to prevent subsequent colonisation and disease by virulent microorganisms.

Widespread interest in neonatal bacterial colonisation developed in the 1940s when epidemics of staphylococcal pyoderma first appeared in significant numbers (Allison and Hobbs, 1947; Colbeck, 1949; Shaffer *et al.*, 1956; Shaffer *et al.*, 1957). It was soon established that most infants born in a hospital became nose carriers of *Staphylococcus aureus* within the first few days of life and the most important hospital sources of these microorganisms were the hands of nursery attendants (Wolinsky *et al.*, 1960; Mortimer *et al.*, 1966). Moreover, it also became apparent that bacterial colonisation began at the umbilicus and subsequently spread to involve other body sites (Mortimer *et al.*, 1966) and that a correlation existed between the rate of bacterial colonisation and the incidence of serious bacterial infection (Smith and Bloomfield, 1950; Forfar *et al.*, 1968; *Lancet*, 1968). Numerous procedures were developed to limit neonatal colonisation and disease; none of these measures, however, proved completely satisfactory. Recently, therefore, additional attempts were made to develop more effective measures for controlling bacterial colonisation in the newborn nursery (*British Medical Journal*, 1970).

Recent investigations carried out in our laboratories (Speck *et al.*, 1976, 1977) dealt with the effect of certain topical antimicrobial agents on bacterial colonisation of the newborn. As part of that study we carried out a detailed analysis of the bacterial flora present at various times in our study population. The present report deals with an analysis of the data with a view to determining whether the presence of one sort of bacterial flora prevented colonisation with another. Moreover, because of the topical agents studied (one selectively prevented colonisation with Gram-negative microorganisms while the other possessed activity mainly against Gram-positive bacteria) it was possible to determine whether the establishment of one type of flora prevents a different one from taking root.

Because of its relevance to the health of the newborn, the present analysis focuses on the effect of the nature of the bacterial flora on colonisation with *Staph. aureus*.

Material and methods

The procedure for selecting newborn infants for inclusion in the study has been described previously (Speck *et al.*, 1977). Two hundred and eighty-six babies were included. The anterior nares and umbilical area of these infants were cultured on days 3, 14, and 42 of life. Cultures were plated on blood agar, McConkey agar, and mannitol-salt agar, and recovered microorganisms were identified by standard bacteriological procedures.

Results

On day 3 of life, 25% of the babies were colonised by *Staph. aureus* while an equal number were colonised with *Staph. epidermidis*. Significantly, however, only 3% of the babies were colonised by

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Table 1 *Interference between staphylococci*

Day	*Site*	*Per cent of babies colonised*			*No. of babies*
		Staph. aureus *only*	Staph. aureus + Staph. epidermidis	Staph. epidermidis *only*	
3	Umbilicus	25·4	3·4	24·6	268
14	Umbilicus	26·7	11·6	27·5	251
42	Umbilicus	18·4	21·1	48·4	223
14	Nasopharynx	44·2	6·4	39·0	251
42	Nasopharynx	50·7	6·3	29·2	223

Table 2 *Interference between staphylococci and Gram-negative bacteria*

Day	*Site*		*Per cent of babies colonised*			*No. of babies*
			Staphylococcus only	*Staphylococcus in presence of Gram-negative*	*Gram-negative only*	
3	Umbilicus	*Staph. aureus*	20·5	8·2	20·5	268
		Staph. epidermidis	20·9	7·5	20·5	
14	Umbilicus	*Staph. aureus*	31·9	15·9	24·3	251
		Staph. epidermidis	30·7	17·1	24·7	
42	Umbilicus	*Staph. aureus*	30·5	9·4	10·8	223
		Staph. epidermidis	55·2	14·4	7·2	
14	Nasopharynx	*Staph. aureus*	45·0	4·8	6·0	251
		Staph. epidermidis	38·7	5·2	5·6	
42	Nasopharynx	*Staph. aureus*	53·8	1·4	2·7	223
		Staph. epidermidis	34·0	1·8	2·2	

both staphylococcal species (Table 1). (Data for nasopharyngeal colonisation on day 3 are not included because of the very low colonisation rate of that site (Speck *et al.*, 1976, 1977; unpublished results)). On day 14 the umbilical colonisation rate with either *Staph. aureus* or *Staph. epidermidis* remained essentially unchanged while the colonisation rate with both species had increased to 12%—still significantly lower than by either species alone (Table 1). Although staphylococcal colonisation of the nasopharynx was higher than that of the umbilicus on day 14, the extent of dual colonisation (6·4%) was remarkably low (Table 1). Even though bacterial interference at the umbilical site was no longer evident on day 42, it was still maintained in the nasopharynx (Table 1).

Staphylococcal colonisation was significantly reduced in the presence of Gram-negative bacteria. This antagonism, which occurred with both *Staph. aureus* and *Staph. epidermidis*, continued for at least 42 days (Table 2). Conversely, staphylococci suppressed colonisation with Gram-negative bacteria. However, this inhibition was evident for the first two weeks of life only and was limited to the umbilical area (Table 2).

Discussion

Antagonism between microorganisms has been recognised since 1874 when Roberts noted the 'recognised antagonism between the growth of certain strains of bacteria' (*Lancet*, 1975). Subsequently there were many other studies dealing with *in vitro* bacterial interference. Thus diGiacinto and Frazier (1966) were able to document interference between certain coliforms and *Staph. aureus* while Iandolo and his colleagues (1965) demonstrated antagonisms between streptococci and staphylococci. Bacterial interference occurring *in vivo* has received less attention. It was reported that strains of *Staph. epidermidis* inoculated into experimental burns of laboratory animals reduce the severity of infection with hospital strains of *Staph. aureus* (Wickman, 1970) and that the inoculation of *Staph. epidermidis* into embryonated hen's eggs 24 hours before challenge with *Staph. aureus* greatly decreased the mortality rate (Ribble, 1965). The clinical application of bacterial antagonism to humans has received even less attention despite the success of early work with *Staph. aureus* 502A which showed that this strain inhibited subsequent challenge with more virulent

epidemic staphylococcal strains (Boris *et al.*, 1963; Shinefield *et al.*, 1963).

The mechanism whereby one organism interferes with the growth of another has not been demonstrated in all instances. However, it has been shown that certain microorganisms are capable of elaborating substances which inhibit the growth of other bacteria and/or deplete the environment of specific nutrients which are required for growth of other bacterial species (Reeves, 1965; Ribble, 1965). The role of these phenomena in human colonisation remains speculative; however, the recent demonstration of antibiotic-producing bacteria in the skin flora of 23% of a normal population and the correlation between the presence of these antibiotic-producing strains and an absence of secondary infections suggests that microbial antagonism may be more than a laboratory phenomenon (Selwyn, 1975).

The present data suggest that in the newborn there is interference between *Staph. aureus* and *Staph. epidermidis* as well as between either staphylococcus and Gram-negative bacteria. These findings lead one to consider the attractive possibility of using the natural body flora to control colonisation and combat infection. It should be noted that the concept of seeding with non-pathogens to prevent colonisation with disease-causing microorganisms is not new (Boris *et al.*, 1963; Shinefield *et al.*, 1963). However, the present data indicate that this concept should be applied to a new site.

References

Allison, V. D., and Hobbs, B. C. (1947). An inquiry into the epidemiology of pemphigus neonatorum. *British Medical Journal*, **2**, 1-7.

Boris, M., Shinefield, H. R., Ribble, J. C., Eichenwald, H. F., Hauser, G. H., and Caraway, C. T. (1963). Bacterial interference: Its effect on nursery-acquired infection with *Staphylococcus aureus*. IV. The Louisiana epidemic. *American Journal of Diseases of Children*, **105**, 674-682.

British Medical Journal. (1970). Infection in the nursery (Leading article). *British Medical Journal*, **3**, 235-236.

Colbeck, J. C. (1949). An extensive outbreak of staphylococcal infections in maternity units. *Canadian Medical Association Journal*, **61**, 557-568.

DiGiacinto, J. V., and Frazier, W. C. (1966). Effect of coliform and Proteus bacteria on growth of *Staphylococcus aureus*. *Applied Microbiology*, **14**, 124-129.

Forfar, J. C., Gould, J. C., and MacCabe, A. F. (1968). Effect of hexachlorophane on incidence of staphylococcal and gram-negative infection in the newborn. *Lancet*, **2**, 177-179.

Iandolo, J. J., Clark, C. W., Bluhm, L., and Ordal, Z. J. (1965). Repression of *Staphylococcus aureus* in associative culture. *Applied Microbiology*, **13**, 646-649.

Lancet. (1968). Hexachlorophane in the nursery. *Lancet*, **2**, 205.

Lancet. (1975). Antibiosis amongst skin microorganisms (Editorial). *Lancet*, **2**, 1080-1081.

Mortimer, E. A., Jr., Wolinsky, E., Gonzaga, A. J., and Rammelkamp, C. H., Jr. (1966). Role of airborne transmission in staphylococcal infections. *British Medical Journal*, **1**, 319-322.

Reeves, P. (1965). The bacteriocins. *Bacteriological Reviews*, **29**, 24-45.

Ribble, J. C. (1965). A mechanism of bacterial interference. *Journal of Clinical Investigations* (Abstract), **44**, 1091.

Selwyn, S. (1975). Natural antibiosis among skin bacteria as a primary defence against infection. *British Journal of Dermatology*, **93**, 487-493.

Shaffer, T. E., Baldwin, J. N., Rheins, M. S., and Sylvester, R. F., Jr. (1956). Staphylococcal infections in newborn infants. I. Study of an epidemic among infants and nursing mothers. *Pediatrics*, **18**, 750-760.

Shaffer, T. E., Sylvester, R. F., Jr., Baldwin, J. N., and Rheins, M. S. (1957). Staphylococcal infections in newborn infants. II. Report of 19 epidemics caused by an identical strain of *Staphylococcus pyogenes*. *American Journal of Public Health*, **47**, 990-994.

Shinefield, H. R., Ribble, J. C., Boris, M., and Eichenwald, H. F. (1963). Bacterial interference: its effect on nursery acquired infection with *Staphylococcus aureus*. I. Preliminary observations on artificial colonization of newborns. *American Journal of Diseases of Children*, **105**, 646-654.

Smith, J. W., and Bloomfield, A. L. (1950). The development of the aerobic bacterial flora of the throat in newborn babies. *Journal of Pediatrics*, **36**, 51-60.

Speck, W. T., Driscoll, J. M., Jr., Polin, R. A., O'Neill, J., and Rosenkranz, H. S. (1976). Bacterial colonization in the newborn: effect of cord care (Abstract). *Pediatric Research*, **10**, 335.

Speck, W. T., Driscoll, J. M., Jr., Polin, R. A., O'Neill, J., and Rosenkranz, H. S. (1977). Bacterial colonization of the newborn infant—Effect of antiseptic cord care. *American Journal of Diseases of Children*. (In press.)

Wickman, K. (1970). Studies of bacterial interference in experimentally produced burns in guinea pigs. *Acta Pathologica et Microbiologica Scandinavica*, **78B**, 15-28.

Wolinsky, E., Lipsitz, P. J., Mortimer, E. A., Jr., and Rammelkamp, C. H., Jr. (1960). Acquisition of staphylococci by newborns: direct versus indirect transmission. *Lancet*, **2**, 620-622.