

***Klebsiella* species in hospital food and kitchens: a source of organisms in the bowel of patients**

By E. MARY COOKE, TAHERE SAZEGAR,* A. S. EDMONDSON,
JANET C. BRAYSON AND DIANA HALL

Department of Microbiology, University of Leeds, Leeds LS2 9NL

(Received 28 May 1979)

SUMMARY

Hospital food was examined for the presence of *Klebsiella* species. Salads and cold meat were most frequently contaminated, often containing more than 10^8 organisms per g. *Klebsiella* species were also widely distributed in the kitchen environment which was considered, at least in part, to be the source of the organisms in food.

By the use of serological and bacteriocin typing, intestinal carriage of strains ingested in food could be demonstrated.

INTRODUCTION

The epidemiology of klebsiella infections in hospitals is not well defined (Lancet, 1971). Selden *et al.* (1971) have demonstrated that an important source of klebsiella strains causing infections may be the patient's own bowel. In addition to being a potential source of autoinfection, the acquisition of a strain in the bowel during hospitalization provides a possible source for transmission of the organism.

If intestinal carriage of klebsiella is an important source of infection, then the sources of the strains colonizing the patient's bowel are of interest. Hospital food has been shown to contain klebsiellas in numbers great enough to suggest that colonization of the bowel might occur (Montgomerie *et al.* 1970; Shooter *et al.* 1971) and infant feeds have also been shown to be contaminated (Ayliffe, Collins & Pettit, 1970).

The three surveys reported in this paper were designed to examine the extent of klebsiella contamination of food prepared in a hospital kitchen, to define in greater detail the sites at which food became contaminated with klebsiella by examining the kitchen environment and food both before and after preparation, and to determine whether ingestion of klebsiellas in food could lead to intestinal colonization.

* Present address: Medical Centre, Ferdowsi University, Meshed, Iran.

MATERIALS AND METHODS

Survey 1

Food prepared in the hospital kitchen was sampled daily for 16 weeks and examined for klebsiellas.

Survey 2

During a 7-week period work surfaces, floors, utensils and sinks in a hospital kitchen were examined twice weekly for klebsiellas. Food at different stages of preparation was also sampled.

Survey 3

Every portion of food and drink and any medicaments taken in 1 week by five patients in a female orthopaedic ward was sampled. During the week faecal specimens were obtained from each patient and examined for klebsiellas. Faecal specimens were also sampled during the week before the food sampling and during the week after food sampling had ended. Patients' hands were examined for klebsiellas on one occasion. The patient's locker and articles on it were sampled weekly.

Bacteriology

In Survey 1 a volume of sterile water equal to the weight of the food sample in grams was added to each sample and suspensions of food were made using either a wrist-action shaker or a stomacher. Each suspension was inoculated on MacConkey agar (Difco), Endo agar (Difco) and deoxycholate agar (Difco) and plates were incubated at 37 °C for 48 h. The number of klebsiella colonies was counted and klebsiellas were identified biochemically as previously (Cooke *et al.* 1979).

In Surveys 2 and 3 solid food samples were mixed with 1/4 strength Ringer's solution in the proportion of 4 ml of Ringer's solution to 1 g of food, and were homogenized in a stomacher. Volumes of 0.1 ml of suspension were spread on MacConkey agar, citrate agar (Difco) and MacConkey-inositol-carbenicillin (MIC) agar (Cooke *et al.* 1979) and were also inoculated into citrate broth. Liquid samples were diluted tenfold in 1/4 strength Ringer's solution and plated out as for solid food samples.

When examining environmental sites hard surfaces were sampled by swabbing with a cotton wool swab moistened in nutrient broth, which was then immersed in a tube containing 1 ml of 1/4 strength Ringer's solution. The tube was agitated on a vortex mixer and the resulting suspensions were inoculated to the same media as for food.

All plates were incubated at 37 °C for 24 h and up to ten klebsiella colonies were selected from each food and environmental sample. Where plate cultures were negative after 24 h, the citrate broth was subcultured on MacConkey, citrate and MIC agar plates.

Faeces were examined as previously (Cooke *et al.* 1979).

Patients' hands were sampled by agitation in 1/4 strength Ringer's solution, which was filtered through 0.45 µm membrane filters; the filters were then incubated on MacConkey agar at 37 °C for 24 h.

Table 1. Isolation of *Klebsiella* from food

Type of food	No. of samples	No. of samples containing <i>Klebsiella</i> (percentage in parentheses)
Salads	211	49 (23.2)
Cold meat	54	14 (25.9)
Other cold food	443	13 (2.9)
Hot food	637	18 (2.8)

Table 2. Isolation of *Klebsiella* from the kitchen environment

Source of sample	No. of samples	No. of samples containing <i>Klebsiella</i> (percentage in parentheses)
Surfaces	53	23 (43.4)
Utensils	87	34 (39.1)
Sinks	57	37 (65.0)
Washing-up water	16	8 (50.0)
Water used for soaking vegetables and salads	12	8 (66.7)

Typing methods

All *klebsiella* strains were serotyped by the quellung method (Kauffmann, 1949), using 77 capsular antisera produced in this laboratory. Strains which were thought to be related were also bacteriocin typed by the method of Edmondson & Cooke (1979).

RESULTS

The results from the three surveys of the isolation of *Klebsiella* from food prepared in the hospital kitchen are given in Table 1. The number of *klebsiellas* in different types of food varied from 10 to $> 10^8$ /g, salads being the most heavily contaminated.

Food was also sampled as it entered the kitchen. Of 136 salad constituents sampled before preparation, 14 (10.3%) contained *Klebsiella* while 6 of 13 (46%) raw meat samples contained *Klebsiella*.

The contamination of the kitchen environment with *Klebsiella* is shown in Table 2. Estimations of the numbers of *klebsiella* organisms in the kitchen environment could not be made in many cases as there was frequently confluent growth on the isolation media. Where estimations could be made there were 10^3 – 10^4 organisms/ml of suspension prepared from an environmental sample.

Forty-six different serotypes were isolated from food and 50 types were isolated from the kitchen environment. Thirty-four serotypes were found in both situations.

Table 3 shows the serotypes of *klebsiella* strains isolated from food, faeces and the immediate environment (i.e. the locker and any articles on it) of four patients studied in Survey 3. No *klebsiellas* were isolated from any source from the remaining patient examined in this survey.

There was no evidence of acquisition of *klebsiellas* from food in patients A and B, for the non-typable strains isolated from the food and faeces of the two patients had distinct bacteriocin typing patterns. Similarly the two strains of serotype 35

Table 3. *Klebsiella* serotypes in the food, faeces and environment of four hospital patients

Patient	Klebsiella serotypes isolated from different sources (No. of isolations in parentheses, when more than one)		
	Food	Faeces	Environment
A	18 (2), 23, 31, 33 41 (4), 47, NT	9, 30, 39, NT	66
B	18, 35, 38 (2), 47 (2) 66, 74, NT	24, 33, 35, NT	2, 27
C	18, 23, 24 (3), 27 (2)	24 (4), 31, 33/35, 47, 54, 62 (2), NT	—
D	20 (2), 36, 38 (2), 62 72 (2), G23 (2)	8 (5), G23 (2), NT (2)	—

NT = Non-typable.

G23 may be a new capsular serotype (Edmondson & Cooke, 1979).

isolated from patient B had distinct bacteriocin typing patterns and had differing antibiotic sensitivity patterns and biochemical reactions. Evidence of acquisition was obtained for patient C, as this patient ingested klebsiellas of serotype 24 on three occasions, and the same serotype was isolated from the faeces of the patient on four occasions. The first isolation of type 24 from faeces was made on the day following the first ingestion of this type, and the final isolation was made three days after the examination of food had ended. The bacteriocin typing patterns, biochemical reactions and antibiotic sensitivity patterns of these seven cultures of serotype 24 suggested that they were identical. Patient D ingested klebsiellas of type G23 on two occasions and klebsiellas of the same serotype and bacteriocin typing pattern were isolated from the faeces of this patient on two later occasions.

There was no correlation between the serotypes found in the environment of the patients and those isolated from the food and faeces of the patients. No klebsiellas were isolated from the patients' hands, nor from any medicaments.

DISCUSSION

The results of the three surveys described in this paper show that all types of hospital food may be contaminated with large numbers of klebsiella organisms. Klebsiellas were most frequently isolated from salads, as has been reported in other surveys (Shooter *et al.* 1971; Wright, Kominos & Yee, 1976) but were also very common in cold meat. There was a considerable difference between the number of klebsiella isolations from salads which had been prepared in the hospital kitchen and salads before preparation since 23.2% of prepared salads contained klebsiella while only 10.3% of salads sampled as they entered the kitchen were contaminated with klebsiella. This suggests that salads became contaminated during preparation. The similarity of the serotypes of klebsiella strains isolated from food and the kitchen environment also suggests that contamination of food from the environment may have occurred.

A total of 62 different serotypes were found in food and the kitchen environment from the three surveys, and no one serotype predominated. A similar range of

serotypes has been isolated from infections in the hospital, again with no particularly common single serotype.

The acquisition in the gastro-intestinal tract of klebsiella serotypes ingested in food has previously been reported in renal-transplant patients (Montgomerie *et al.* 1970) and in intensive-care patients (Casewell & Phillips, 1978). In this paper we demonstrate intestinal colonization by klebsiella strains ingested in food in two patients using two sensitive epidemiological typing methods, capsular serotyping and bacteriocin typing. The use of more than one typing method is valuable in studies of this kind as it improves discrimination between strains.

The results obtained here confirm those of other workers showing that hospital food may be contaminated with klebsiellas, and indicate that the source of the organisms may, at least in part, be the kitchen environment.

It is a pleasure to acknowledge the assistance given by the nursing staff of the Leeds General Infirmary and by Miss Jones, Catering Manager. One of us, A. S. E, was supported by an M.R.C. project grant.

REFERENCES

- AYLIFFE, G. A. J., COLLINS, B. J. & PETTIT, F. (1970). Contamination of infant feeds in a Milton milk kitchen. *Lancet* *ii*, 559–60.
- CASEWELL, M. & PHILLIPS, I. (1978). Food as a source of *Klebsiella* species for colonisation and infection of intensive-care patients. *Journal of Clinical Pathology* **31**, 845–9.
- COOKE, E. M., BRAYSON, J. C., EDMONDSON, A. S. & HALL, D. (1979). An investigation into the incidence and sources of klebsiella infections in hospital patients. *Journal of Hygiene* **83**, 473–80.
- EDMONDSON, A. S. & COOKE, E. M. (1979). The development and assessment of a bacteriocin typing method for klebsiella. *Journal of Hygiene* **82**, 207–23.
- KAUFFMANN, F. (1949). On the serology of the *Klebsiella* group. *Acta pathologica et microbiologica scandinavica* **26**, 381–406.
- LANCET (leading article) (1971). Epidemiology of klebsiella infections. *Lancet* *i*, 416–17.
- MONTGOMERIE, J. Z., DOAK, P. B., TAYLOR, D. E. M., NORTH, J. D. K. & MARTIN, W. J. (1970). *Klebsiella* in faecal flora of renal-transplant patients. *Lancet* *ii*, 787–92.
- SELDEN, R., LEE, S., WANG, W. L. L., BENNETT, J. V. & EICKHOFF, T. C. (1971). Nosocomial klebsiella infections: intestinal colonization as a reservoir. *Annals of Internal Medicine* **74**, 657–64.
- SHOOTER, R. A., FAIERS, M. C., COOKE, E. M., BREADON, A. L. & O'FARRELL, S. M. (1971). Isolation of *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella* from food in hospitals, canteens, and schools. *Lancet* *i*, 390–2.
- WRIGHT, C., KOMINOS, S. D. & YEE, R. B. (1976). *Enterobacteriaceae* and *Pseudomonas aeruginosa* recovered from vegetable salads. *Applied and Environmental Microbiology* **31**, 453–4.