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IN-VITRO RESISTANCE OF GRAM-NEGATIVE ENTERIC BACILLI FROM WOUND INFECTIONS TO HONEY

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RUNNING TITLE: RESISTANCE OF ENTERIC BACILLI TO HONEY

Honey was found by some workers to possess antibacterial activity. It has never been reported to cause any tissue damage, and appears to actually promote the healing process. Our work however showed that all the aerobic Gram-negative bacilli tested produced an *in-vitro* resistance to the honey used.

Sir,

The concept of prophylactic antibiotics was established in the 1960s when experimental data established that antibiotics had to be in the circulatory system at high enough dosage at the time of incision to be effective (1). With the use of antibiotics, a new era in the management of wound infections commenced. Unfortunately, eradication of the infective plague affecting surgical wounds has not ended because of the insurgence of antibiotic-resistant bacterial strains and the nature of more adventurous surgical intervention in immunocompromised patients and in implant surgery (2). Honey was found by some workers to possess antibacterial activity where antibiotics were ineffective (3, 4, 5). Much of the effectiveness of honey as a dressing appears to be due to its antimicrobial properties. Honey is reported to cause no tissue damage, and appears to actually promote the healing process (6).

Since honey is used extensively in Nigeria, even at University College Hospital, Ibadan (a premier hospital in Nigeria), we felt it desirable to reassess its effectiveness in view of the prevailing multiple antibiotic resistance reported by many authors (7, 8, 9). We isolated aerobic Gram-negative enteric bacilli from wound specimens at the University College Hospital (Out-patient and in-patient department) (Table 1) and were identified by standard procedures (10, 11). The minimum inhibitory concentration of honey for each isolate was determined using punch-

hole agar diffusion method. Sensitivity pattern was compared with the standard *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853. All isolates flourished at 70%^{v/v} and below, very poor inhibition of the bacteria was noticed at 80%^{v/v}, 90%^{v/v} and 100%^{v/v} (Table 1). The water activity of the honey (a_w) was 0.6 and the pH is 3.6.

All the aerobic Gram-negative bacilli isolated showed an *in-vitro* resistance to the honey used. This study is similar to the finding of Adefule *et al* (12) in which the disc assay method employed showed no zone of inhibition for the undiluted honey (100%^{v/v}). Nzeako and Hamdi (3) and Subrahmanyam *et al* (13) reported *in-vitro* antibacterial activity of honey at various concentrations against Gram-negative bacilli isolated from wound infections. The honey used in this study was obtained from Saki in Oyo State and Pharmacy department, University College Hospital, Ibadan. Saki is known to produce a very high quality honey in south-western Nigeria, and that used in the study of Adefule *et al* (12) was obtained from Edo State, south-south region of Nigeria. It is possible that the honey obtained from these regions may be synthesized from the same family of flower nectar that shows less antibacterial activity unlike the honey in the study of Nzeako and Hamdi (3) and Subrahmanyam *et al* (13) which were obtained from Saudi-Arabia and India, respectively. Allen *et al* (14) showed that there are many types of honey with and without antibacterial activity and postulated that the type of the flower that

was the source of the nectar determines the nature of the antibacterial activity of honey.

The water activity (a_w) and pH of honey are also important factors in their antimicrobial properties; a_w (0.6) of this honey showed it is ripe enough to inhibit bacteria growth (11). Similarly, honey is characteristically quite acidic, the pH 3.6 obtained in this study is low enough to be inhibitory to many of

these pathogens (6). It is also very possible that aerobic Gram-negative enteric bacilli have developed resistance to honey which was hitherto sensitive. In a related study carried out by Ogbolu *et al* (15); *Candida* isolates had better susceptibility to coconut oil than fluconazole and their results elucidated the use of coconut oil in complementary and alternative medicine especially in this era of emerging drug-resistant *Candida* species.

TABLE 1: Number and percentage of strains inhibited on MH with different concentrations of honey

Organisms	5%-70% x (%)	80% x (%)	90% x (%)	100% x (%)
<i>Escherichia coli</i> n =10	0 (0)	0 (0)	0 (0)	3(80)
<i>Klebsiella species</i> n=15	0 (0)	0 (0)	0 (0)	0 (0)
<i>Proteus vulgaris</i> n=2	0 (0)	0 (0)	0 (0)	2 (25)
<i>Pseudomonas aeruginosa</i> n=21	0 (0)	1 (4.8)	1 (4.8)	2 (9.5)
Key: - n = number of strains	x= number of	strain that show	inhibition	

Having observed a lesser antibacterial activity of honey against the aerobic gram-negative enteric bacilli *in-vitro* further studies to investigate the *in-vivo* antibacterial activity of honey against the enteric bacteria from wound infections will be fundamental to the well being of the populace, as treatment of wound with honey is still widely practiced in Nigeria. Essentially, study into the antibacterial activities of honeys produced by various flowers that are the sources of the nectar is also imperative to further clarify the ambiguity in the use of honey as an antimicrobial agent.

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