P1046

Colonization by multidrug-resistant bacteria in long-term care facilities in Italy: a point-prevalence study.

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Introduction and Purpose

- Multidrug-resistant organisms (MDROs) constitute a major public health concern worldwide (1).
- Residents in long-term care facilities (LTCFs) are at increased risk for colonization/infection with MDROs because of age-associated morbidities, exposure to recurrent antibiotic courses and frequent referral to and from acute-care hospitals (2).
- This study aimed to determine the prevalence of colonization in elderly LTCF residents in Italy, by:
- extended-spectrum β-lactamase (ESBL)- and/or carbapenemase-producing Enterobacteriaceae;
- hypervirulent antibiotic resistant *Clostridium difficile*;
- methicillin-resistant *Staphylococcus aureus* (MRSA)

Methods

Study design and participants. A point-prevalence study was conducted at 12 LTCFs located in 4 different Italian regions (1 February 2015–15 March 2015). The total number of residential beds was 856. Only LTCF residents aged ≥ 65 years who were not admitted in sub-intensive care units and/or special care wards were enrolled in the study. Socio-demographic and clinical data of the residents were collected.

Specimen collection, bacteria identification and antibiotic susceptibility testing. After obtaining informed consent, clinical samples were collected: i) faecal samples for *C. difficile*; ii) faecal samples/rectal swabs for ESBL- and/or carbapenemase-producing Enterobacteriaceae and iii) nasal/axillary swabs for MRSA. Faecal samples/swabs were streaked onto appropriate chromogenic agar plates for the detection of the microorganisms under study. Identification at species level, antibiotic susceptibility testing and confirmation of ESBL production were carried out according to standard laboratory procedures using a shared protocol. The interpretative criteria were based on the EUCAST clinical breakpoints.

Carbapenemase production confirmatory testing. Phenotypic testing was performed with the agar tablet/disc diffusion method (KPC/MBL and OXA-48 ConfirmKit, ROSCO Diagnostica A/S, Denmark). Identification of carbapenemase encoding genes (bla_{VIM}-, bla_{IMP}-, bla_{NDM}- and bla_{KPC}-type) and their variants was achieved by PCR and sequencing (3).

Genotypic characterization of MDROs

- **ESBL- and/or carbapenemase–producing *E. coli:*** phylogenetic typing, multilocus sequence typing (MLST) and *fimH*-based subtyping of the ST131 isolates were performed following procedures previously described (4, 5).
- **ESBL- and/or carbapenemase–producing *K. pneumoniae* isolates:** MLST was carried out.
- ***C. difficile:*** Multiplex PCR was used for the detection of toxin A and B genes (6). Capillary gel electrophoresis and PCR-ribotyping were performed (7).
- **MRSA:** *S. aureus* species and methicillin-resistance status were confirmed by a PCR assay targeting *nuc* and *mec*A genes, respectively (8).

Risk factor analysis

Association of resident's socio-demographic variables and clinical data with outcomes was assessed by $\chi 2$ test or Fisher's exact test, as needed. Multivariate analysis was performed by stepwise logistic regression; variables associated with *p*-value ≤ 0.2 in the univariate analysis were included in the multivariate analysis. The associations to rare events Poisson regression model with a robust error variance and relative risk was used.

Results

Prevalence of MDROs

Overall, a total of 489 (489/856, 57.1%) LTCF residents aged ≥65 years were enrolled. Demographics and clinical details of enrolled residents are summarized in table 1.

Table 1. Data of the 489 LTCF residents enrolled in the study.

Factor	Description	N (%)
Age	Mean	85 years
	Median	86 years
Length of stay in LTCF	Mean	41 months
	Median	18 months
Gender	Female	338 (69.1%)
	Male	143 (29.2%)
Hospitalization	Previous 3 months	104 (21.3%)
Surgery	Previous month	12 (2.5%)
Medical devices	Urinary catheter	73 (14.9%)
	PEG*	25 (5.1%)
	Vascular catheter	18 (3.7%)
Presence of	Wound	21 (4.3%)
	Decubitus ulcer	41 (8.4%)
Mobility	Autonomy	170 (34.8%)
	Non autonomy	210 (42.9%)
	Immobility	104 (21.3%)
Incontinence	Urinary	316 (64.6%)
	Faecal	261 (53.4%)
Temporal/spatial disorientation		217 (44.4%)
Antimicrobial therapy	Previous month	129 (26.4%)
	Current	35 (7.2%)

*PEG, Percutaneous endoscopic gastrostomy

The overall prevalence of colonization by ESBL-producing Enterobacteriaceae, *C. difficile* and MRSA was 57.3%, 5.1% and 17.2% (for at least one sample site), respectively (table 2).

Table 2. Prevalence of colonization by MDROs in LTCF residents.

Resistant organisms	Colonized/enrolled residents
ESBL *E. coli*	239/487 (49.1%)
ESBL *K. pneumoniae*	35/487 (7.2%)
Total ESBL Enterobacteriaceae	279/487 (57.3%)
CPE*	5/487 (1%)
MRSA	84/487 (17.2%)
C. difficile	21/409 (5.1%)

*CPE, carbapenemase-producing Enterobacteriaceae

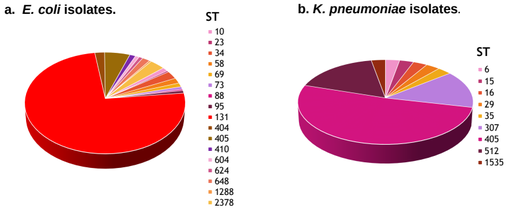
Carriage rate of carbapenemase-producing Enterobacteriaceae was low (1%, 5/487). In fact, 5 isolates only were found to produce carbapenemase (3 *K. pneumoniae* isolates harboured bla_{KPC-3} and 2 *E. coli* isolates carried bla_{VIM-1})

Considering residents carrying ESBL-producing Enterobacteriaceae, the prevalence of colonization by *E. coli*, *K. pneumoniae* and other Enterobacteriaceae ESBL-producing was 49.1%, 7.2% and 4.1%, respectively.

Genotypic characterization of MDROs

A total of 417 MDRO isolates (247 *E.coli,* 35 *K. pneumoniae,* 21 other Enterobacteriaceae species, 93 MRSA and 21 *C. difficile*) were collected.

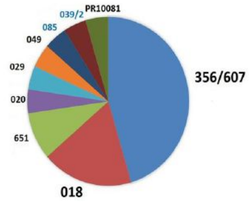
Figure 1. MLST genotyping of ESBL-producing
a. *E. coli* isolates. b. *K. pneumoniae* isolates.



The MDR H30-ST131 sub-clone strongly predominated among *E. coli* isolates (175/247, 70.9%). The remaining 72 E. coli isolates were distributed among 26 different STs (figure 1a). Apart from ST 131, ST405 (13 isolates), ST 2378 (8 isolates) and ST34 (6 isolates) were common.
K. pneumoniae isolates were dispersed in 9 different STs, mainly ST405 (18 isolates), ST512 (6 isolates) and ST307 (5 isolates) (figure 1 b).

Significant differences in *C. difficile* colonization rates were observed among the different LTCFs, with no residents colonized in 5 LTCFs. All but one *C. difficile* isolates were toxigenic. Most *C. difficile* isolates (85%) were MDR; of these 82% were resistant to erythromycin, clindamycin, rifampicin and moxifloxacin. The predominant PCR-ribotype was 356/607 (50%) (figure 2).

Figure 2. *C. difficile* PCR-ribotyping.



Genotyping of MRSA isolates including *spa* typing, SCCmec typing and MLST is reported in poster P204.

Risk factor analysis

According to multivariate analysis, significant risk factors for carriage of ESBL Enterobacteriaceae were being bedridden (OR 1.85; 95% CL 1.02 – 3.34; *p*=0.043), being urinary and/or faecal incontinent (OR 1.57; 95% CL 1.00 – 2.45; *p*=0.048) and the facility (*p*=0.0010). For *C. difficile* carriage, significant risk factors were being bedridden (RR 3.99; 95% CL 1.66 – 9.63; *p*=0.002) and recent hospitalization (RR 3.79 ; 95% CL 1.60 – 8.97; *p*=0.002). Length of stay in the LTCF ≤4 months (OR 2.05; 95% CL 1.22 – 3.43; *p*=0.0007) and being bedridden (OR 1.84; 95% CL 1.06 – 3.17; *p*=0.029) were independently associated with MRSA colonization.

Conclusions

- ✓ This study documents a high prevalence of colonization by ESBL-producing Enterobacteriaceae, especially ESBL-producing *E. coli*, among LTCF residents in Italy
- ✓ The high carriage of ESBL- producing *E. coli* was associated with the spread of the pandemic H30-ST131 sub-clone that was found to predominate not only among infected residents (see poster P1045) but also among colonized ones.
- ✓ On the contrary, carbapenemase-producing Enterobacteriaceae was rarely detected in both carriage and infections (see poster P1045)
- ✓ The prevalence of *C. difficile* colonization was significantly different among the LTCFs enrolled. MDR *C. difficile* PCR-ribotype 356/607 was the most frequently detected, as also observed for the infected patients (see poster P1045).
- ✓ The rate of MRSA colonization in the LTCFs under study was high, although it varied according to the facility.
- ✓ A low level of autonomy and consequent exposure to frequent contacts with caregivers appears a common risk factor for all MDROs. This element underlines the needs for improving specific measures of prevention capable to control MDRO transmission.
- ✓ MDROs represent a huge problem in Italian LTCFs, confirming the importance of these facilities as reservoir of MDROs for acute-care hospitals.

ACKNOWLEDGMENTS

This study was conducted with the financial support of the Italian Ministry of Health-Centro Controllo Malattie project "Infezione e colonizzazione da patogeni multi-resistenti nell'anziano in residenze sanitarie assistenziali".

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